EPIDEMIOLOGY AND INTEGRATED MANAGEMENT OF ANTHRACNOSE OF GREENGRAM

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By

SUNIL A. KULKARNI

DEPARTMENT OF PLANT PATHOLOGY
COLLEGE OF AGRICULTURE, DHARWAD
UNIVERSITY OF AGRICULTURAL SCIENCES,
DHARWAD – 580 005
FEBRUARY 2009

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1. INTRODUCTION

Greengram [*Vigna radiata* (L.) Wilczek], is an important pulse crop of India. It is also commonly known as mungbean, which is an ancient and well known leguminous crop of Asia. It is quite versatile crop grown for seeds, green manure and forage and it is also considered as "Golden Bean" because of its nutritional values and suitability for increasing the fertility of the soil, by the way of addition of nitrogen to the soil. On an average, pulse crops add upto 30 kg nitrogen per hectare per year. Because of its short duration, it fits well in crop rotation and mixed cropping systems.

Greengram is a rich source of protein (23 - 24%), carbohydrate (54 - 56%), minerals and vitamines. It has high digestibility due to which it is fed to babies, convalescents and elders. Unlike other pulses, it is free from flatulent effects in stomach. It is consumed in many forms including boiled dhal, sprouts, bean cakes, noodles and pudding.

Presently, the per capita share of pulses in nutrition supply in India with respect to energy, protein and fat is 117.4 K cal, 6.9 g and 1.0 g per day respectively. An adult male and female requires 80 and 70 g per capita per day, respectively for balanced diet (Anon., 2004). Greengram crop covers a total world area of 5 m ha with a total production of 3 mt (John, 1991). It is widely cultivated throughout the South Asia including India, Pakistan, Bangladesh, Sri Lanka, Thailand, Cambodia, Vietnam, Indonesia, Malaysia and South China. India is an important pulse growing country contributing 28 per cent to the global pulse basket from an area of about 37 per cent (Masood Ali and Shivkumar , 2000)

Greengram is grown mainly as a *kharif* season crop. However, its cultivation in *rabi* season is restricted to the eastern and southern parts of the country. The major greengram growing states are Orissa, Maharashtra, Andhra Pradesh, Rajasthan, Karnataka and Gujarat. It ranks third among all pulses grown in India after chickpea and pigeonpea. In India the total production of greengram is 13.74 lakh tonnes from an area of 32.99 lakh ha with a productivity of 417 kg ha^{-1}. The Hyderabad Karnataka area particularly Bidar and Gulbarga districts have an extensive cultivated area of greengram, pigeonpea and bengalgram. Hence, these regions are called "Pulse bowl" of Karnataka. In Karnataka, it occupies 1.77 lakh ha with a production of 0.71 lakh tonnes and average yield of 399 kg ha^{-1} (Rajendra Prasad, 2006).

The crop is generally grown during *kharif* as rainfed crop. It has the yield potential of 11 to 12 q ha^{-1} (Anon., 2004), as against the national average of 4.17 q ha^{-1}. Among various factors responsible for low yields, biotic and abiotic stresses take a heavy toll of the crop, out of which diseases cause an estimated yield loss of 20 to 30 per cent (Singh, 1995).

Greengram suffers from many diseases caused by fungi, bacteria, viruses, nematodes and also abiotic stresses. Among the fungal diseases, powdery mildew, anthracnose, *Cercospora* leafspot, web blight and dry root rot are the most prevalent ones. In recent years, greengram anthracnose caused by *Colletotrichum truncatum* (Schw.) Andrus and Moore has become one of the major diseases which is known to occur in many countries *viz.* India, Nigeria, Thailand, Philippines, Upper volta, Zambia, Palmira, Columbia, *etc*. (Agarwal, 1991). It occurs in all the parts of the world, wherever greengram is cultivated.

In India, the greengram anthracnose was first reported from Jorhat of Assam state in 1951 (Majid, 1953). The disease has been reported from all major mungbean growing regions of India in mild to severe form and in tropical and subtropical areas it causes considerable damage by reducing seed quality and yield. The disease causes qualitative as well as quantitative losses (Sharma *et al.,* 1971). Losses in yield due to anthracnose have been estimated to be in the range of 24 to 67 per cent (Deeksha and Tripathi, 2002a) and 18.2 to 86.57 per cent disease index of anthracnose have been reported in northern Karnataka (Laxman, 2006)

The yield losses caused by greengram anthracnose are proportional to the disease severity and vary remarkably depending on the stage of infection, genotypes and environmental conditions. The disease is characterized by serious leaf spotting ultimately resulting in 'shot hole' symptoms and finally

defoliation which affects the yield greatly. Infection of pods directly damages the seeds and reduces its germinability. Pod infection may result in complete loss in yield.

The disease of late has attained the economic status in the state. However, no much systematic research work has been carried out on loss assessment, cultural and physiological characteristics of the pathogen, epidemiology and integrated management of the hitherto neglected but important disease of greengram. A number of management approaches *viz*., use of tolerant varieties, application of fungicides, cultural practices and combination of approaches leading to integrated management of the disease have been evaluated and recommended (Rathaiah and Sharma, 2004, Mittal, 1998 and Laxman, 2006). In spite of all these measures, anthracnose continues to be one of the major constraints in greengram production. In addition, there are large variations among different genotypes and cultural practices in different regions. These spatial and temporal variations render it difficult to evolve common management strategies to control anthracnose epidemics. Therefore, it is necessary to know the severity of the disease and factors associated with disease development, which will help in devising suitable and effective management practices feasible to each location, looking into the prevailing conditions,. In Karnataka, research work on important aspects of the anthracnose disease of greengram has not been done systematically. Information on crop loss assessment, epidemiological studies and integrated disease management strategies is lacking.

The present investigations were, therefore, initiated on some of such neglected but important aspects of this disease and the pathogen. It is necessary to conduct a survey of the disease, so that its distribution and extent of severity can be understood and hot spots can be located, which would also help in natural screening for host plant resistance (HPR). The estimation of crop loss is an important parameter in determining the economic importance of the disease and in order to develop threshold for determining, when the exact cost effective management practices should be deployed

Epidemiological studies play an important role in developing prediction and forecasting models about disease progress in relation to disease severity and environmental factors. The pathogen survives in different forms during unfavourable environmental conditions. The methods of survival and spread of the pathogen need to be worked out to delink the infection chain at appropriate time in order to manage the disease effectively. Host plant resistance is considered as most practical, feasible and an economical method of plant disease management. Estimation of biochemical constituents help in detecting their role in the resistance mechanism. Effective management of this disease is a necessary strategy using host plant resistance, plant extracts, bio agents, Indigenous Technology Knowledge (ITK's) and fungicides. Therefore the present investigation was undertaken with the following objectives.

1. Survey and surveillance for the severity of anthracnose of greengram in northern parts of Karnataka.
2. Studies on loss assessment due to greengram anthracnose.
3. Studies on cultural and physiological characteristics of the pathogen.
4. Studies on epidemiological aspects of the disease.
5. Screening of greengram genotypes against anthracnose and identification of biochemical factors for resistance.
6. Integrated management of the disease.

2. REVIEW OF LITERATURE

Indians being basically vegetarian are dependent on plant based diet, where protein requirement is supplemented by pulses. Among the pulses, greengram is one of the important crops. Because of its higher protein content and easy digestibility, it has become an important diet. Greengram is affected by a number of fungal, bacterial and viral diseases, which cause heavy loss. In recent years, greengram anthracnose caused by *Colletotrichum truncatum* (Schw.) Andrus and Moore has become one of the major diseases occurring in all parts of the world wherever greengram is grown.

Literature is available on anthracnose disease of other crops like soybean, cowpea, blackgram and beans but little information is available on anthracnose of greengram. The review of literature on *Colletotrichum* species of different crops is as given hereunder.

2.1 History, distribution and economic importance

The term anthracnose literally means 'coal like' and was coined first time in 1833 by Fabra and Dunal to describe the disease of grape in which blackening of tissues was the striking feature and pathogen associated was *Colletotrichum* (Sanjay Kumar and Srivastava, 1983). Atleast, four species of *Colletotrichum* have been found associated with greengram and blackgram, infecting leaves, stem and pods severely in different parts of the world (Saxena and Sinha, 1977; Hiremath, 1981). Three different species of *Colletotrichum viz., C. truncatum*, *C. lindemuthianum* and *C. dematium* were reported to cause leaf spots or anthracnose in mungbean in India (Saxena and Gupta, 1981; Bharadwaj and Singh, 1986 and Thakur and Khare, 1989). *C. truncatum* was reported to cause leaf spot or anthracnose in greengram in Karnataka (Laxman, 2006). In India, the greengram anthracnose was first reported from Jorhat of Assam state in 1951 (Majid, 1953). Later, Rathaiah and Sharma (2004) reported anthracnose disease on mungbean, which is caused by *C. truncatum* in Assam and subsequently by Laxman (2006) from Karnataka.

Anthracnose has been reported to occur on greengram in several countries including India, Nigeria, Thailand, Philippines, Upper Volta, Zambia, Palmira, Columbia, *etc.* wherever greengram is cultivated (Agarwal, 1991).

Deeksha and Tripathi (2002a) reported the loss in yield ranging from 24 to 67 per cent. Laxman (2006) found 18.2 to 86.57 per cent severity of the disease in northern parts of Karnataka.

2.2 Symptomatology

Majid (1953) reported that *C. lindemuthianum* infects seedlings, stem, petioles and pods in greengram. Brown, sunken lesions are formed on the cotyledons and the young stems of seedlings. During wet conditions, the lesions increase in size and number and young plants get killed after attack. On pods, depressed, black cankers appear, sometimes with flesh coloured centres. Seeds may also be infected resulting in pre-emergence death. In severe infections, the affected parts may wither off. Tripathi and Beniwal (1977) described the symptoms of anthracnose as small, circular, brown spots appearing mainly on leaves. Later, the spots enlarge and develop concentric rings giving the target board effect. The concentric rings are dark brown, while the in between area is grey/dirty grey. The spots may be sickle shaped, circular to irregular in shape with grey centres and measure 4 to 8 mm in greengram, the spotted portion often becomes papery and fall off the leaves producing 'shot holes'.

Singh and Shukla (1988) reported that *C. truncatum* causing serious leaf spotting in greengram, the disease manifests mainly on the leaves but under severe conditions it may produce small, reddish brown, 1 to 4 mm diameter spots on petioles, light grey, water soaked, irregular, later on turning to rustic brown, 10 to 14 mm diameter spots on stems and small, reddish blotches on pods. The leaf spots later turn to form 'shot holes' and in severe cases pre-mature defoliation occurs.

Bharadwaj and Singh (1986) opined that in urdbean and mungbean, the symptom appeared as water soaked greenish lesions which assumed the shape of a horse shoe, such lesions eventually turned straw coloured with narrow reddish brown to dark brown margins. After a few days, necrotic areas were shed leaving behind the reddish brown margin intact. In case of mungbean, the margin around necrotic areas became irregular in shape as a result of

extension of infection on the lower leaves. Usually, such infected portions gave brittle appearance to the spot which resulted in the fall of a small necrotic area. The spots occasionally coalesced.

Bains *et al.* (1989) observed red/brown lesions on leaves, stems, branches and pods of different cultivars of *Vigna radiata* and *Vigna mungo* in Gurdaspur.

Rathaiah and Sharma (2004) indicated appearance of characteristic blood red ring like spots of 8 – 11 mm diameter on the upper surface of leaf. The spots on the lower surface of leaf were different from those on the upper surface, which are comparatively large (10 mm diameter) patches of bright blood red stains. The corresponding upper surface became chlorotic. Blood red stains measuring upto 2.5 cm in length appeared on the pods. Similar symptoms occurred on the petioles.

Laxman (2006) noticed that lesion or spots, which started on lower surface of leaf. At later stage it also appeared on upper surface and spots coalesced to form a big pustules. Spots also appeared on branches, stems and even on pods, showing brown lesions with reddish brown centre.

2.3 Etiology

Quimio (1975) reported some morphological features of an isolate of *C. lindemuthianum* from greengram. The mycelium is branched, septate, hyaline at first but becoming dark with age. The acervuli are saucer shaped, sub-cuticular and become erumpent. The conidia are borne acrogenously on short conidiophores and appear pink in mass. They are 1 celled, hyaline, oblong, cylindrical with rounded ends or with one end slightly pointed. Conidia measure 10 to 20 × 3 to 6 µ in size. The appressoria are sparse, pale to dark brown, clavate or circular in out line, regular and 8 × 6.7 µ in size.

Saxena and Sinha (1977) described morphology of *C. truncatum* from greengram and blackgram. The colonies are dark brown to black with septate, branched, brown and 3 to 4 µ thick mycelium. The acervuli are hemispherical to truncate, conical in shape and measure 150 to 250 µ. The setae are dark brown to black, 6 to 10 per acervulus, filliform, 1 to 3 septate, 60 to 300 µ long and 3 to 8 µ wide. Conidiophores are short, erect, simple, hyaline, non-septate and 12.5 to 20.5 × 2 to 3.2 µ in size. Conidia are hyaline, but pinkish in mass, unicellular, thin walled, falcate to lanceolate, bluntly tapering towards both ends and measure 16 to 20 × 3.0 to 3.5 µ.

Bharadwaj and Singh (1986) reported that isolates of *C. dematium* f. *truncata* from blackgram, greengram, horsegram and soybean appeared to differ from each other in regard of their pathogenicity on six leguminous hosts but blackgram and greengram isolates did not differ much. On PDA, both formed dirty white to dark colonies with abundant sporulation but mungbean isolate was darker with leathery colony.

Sinclair and Backman (1989) described morphological characters of *C. truncatum*. They found, black acervuli borne on well developed stomata. The acervuli were oval to elongate, hemispherical to truncate, conical and erumpent with numerous black, needle like intermixed long and short setae, 60 to 300 × 3 to 8 µ. Conidia were bluntly tapered, curved unicellular and hyaline and measuring 17 to 31 × 3 to 45 µ. Germtube in contact with solid surface produced dark, sticky approssoria, which penetrate directly.

Murthy (1997) observed morphological features of *C. dematium.* Acervuli were black in colour and oval to conical in shape measuring 145.0 × 210.7 µ. Conidia were single celled hyaline, curved and measuring 20.5 to 24.5 × 3.5 to 4.6 µ. Setae were black in colour, longer than conidia, broader at base and tapering at apex measuring 80.0 to 190.0 × 3.7 to 5.8 µ.

Madhusudhan (2002) reported morphological characters of *C. truncatum* as a hyaline mycelium, which is septate and branched. The acervuli were oval to conical and appeared single. They were dark brown to black in colour and measured 181.0 × 275.5 µ in size. The conidia were single celled, smooth, hyaline, curved and measured 21.0 to 23.5 × 3.80 to 4.10 µ in size.

2.4 Survey and surveillance

The survey carried out in the districts *viz.,* Farrukhabad, Kanpur and Hamirpur of Uttar Pradesh indicated that, mungbean and urdbean were found to be naturally infected by several leaf infecting fungi including *Erysiphae polygoni*, *Colletotrichum truncatum* and *Cercospora canescene* (Saxena and Gupta, 1981).

Lakshmi Ramakrishnan (1964) surveyed the seed borne infection intensity of *C. lindemuthianum* on *Dolichus lablab* and reported it to the extent of 13.40 per cent.

The seed infection due to *C. truncatum* ranged between 15.20 to 81.00 per cent and there was reduction in the weight of diseased seeds of about 22.4 to 61.7 per cent (Verma and Upadhyay, 1973). Zate *et al.* (1976) also reported the seed borne infection intensity of *C. lindemuthianum* to the extent of 10 to 30 per cent.

Roy (1982) conducted survey for seedling infection by *C. truncatum* on soybean in USA and observed 32 per cent plant emergence and 100 per cent hypocotyl infection and 3.8 per cent stunting index under field condition.

Khare and Chacko (1983) observed 30 per cent seed infection in soybean due to *C. truncatum* and there was drastic reduction of 50 per cent germination in such seed lots.

The observations recorded during 1989 at Asian Vegetable Research and Development Centre (AVRDC), Taiwan, on the incidence of *C. truncatum* on soybean plants in advanced varietal trial-I and II, showed mean incidence of 50 and 34 per cent, respectively (Anon., 1989).

Angadi (1999) carried out survey for the incidence of anthracnose of chilli caused by *C. capsici* in Raichur, Dharwad and Gadag districts. The disease was more prevalent in Raichur district than in Dharwad and Gadag districts.

Sanathkumar (1999) during his survey in and around Bangalore district observed that, the chilli varieties Chikkaballapur, Gauribidanur, Byadagi Kaddi and Pant C-2 showed anthracnose infection of 25, 35, 30 and 25 per cent, respectively.

Madhusudhan (2002) surveyed the incidence of soybean anthracnose caused by *C. truncatum* and collected 24 soybean seed samples of 15 genotypes from Bangalore, Dharwad, Nippani, Sankeshwar, Ugarkhurd, Coimbatore, Guntur and Indore. Further, he reported that seed infection by *C. truncatum* was of 2.0, 3.92, 6.75, 4.08, 4.25, 0.50, 5.17 and 3.25 per cent, respectively.

Laxman (2006) reported 18.2 to 86.57 per cent incidence of the greengram anthracnose in north Karnataka. Varaprasad (2000) conducted survey for chickpea blight caused by *C. dematium* and observed disease incidence in the range of 0 to 91.00 per cent in and around Gulbarga district.

2.5 Estimation of loss

Estimates of loss are a pre-requisite to the rational development of any agricultural research programme with an important component of plant protection (Campbell and Madden, 1990; Gaunt, 1987; Teng and Johnson, 1988). Reliable estimates of loss facilitate the objective identification of the relative importance of biotic pests. Consequently, limited resources can be assigned on a priority basis and optimize returns from a given effort. Accurate information concerning losses is also needed by growers and plant protection specialists to develop decision thresholds for determining, when cost effective control measures should be deployed (Nutter, 1993). The need for reliable crop loss assessment methodology (to develop reliable decision aids) assumes added importance given the current worldwide concern about improving or maintaining environmental quality by reducing the use of pesticides (Stern *et al.*, 1959).

Chowdhury (1957) reported that anthracnose of chilli was quite serious and wide spread in Assam. The disease has been recorded from that state wherever chilli was grown resulting in a loss of 12.30 per cent of the fruits. Bansal and Grover (1969) during their studies on *C. frutescens* Linn. reported that crop losses due to anthracnose disease ranged from 10 to 35 per cent of fruits in 1966 and 20 to 60 per cent of fruit during 1967 in six districts of Punjab and Haryana.

The seed infection due to *C. truncatum* in soybean was observed with a range of 15.20 to 81.00 per cent and there was reduction in weight of diseased seeds from 22.4 to 61.7 per cent (Verma and Upadhyay, 1973).

Schneider *et al.* (1976) estimated that yield loss upto 42 per cent due to *Cercospora* leaf spot in cowpea and a single application of benomyl gave adequate control of this disease in Nigeria.

Walter (1977) estimated overall yield loss of 25.3 per cent due to diseases in soybean and amongst them anthracnose was responsible for the yield loss of about 3.68 per cent.

Backman *et al.* (1982) reported the reduction in seedling stand, seed quality and yield by 16 to 20 per cent due to anthracnose in soybean.

Khare and Chacko (1983) observed 30 per cent soybean seed infection due to *C. truncatum* and there was drastic reduction of 50 per cent germination in such seed lots.

Sud and Singh (1985) opined that yield losses in blackgram were proportional to anthracnose disease severities at 50 per cent flowering.

Khan and Sinclair (1992) stated that anthracnose and pod blight of soybean caused yield losses upto 30 per cent.

Deeksha and Tripathi (2002a) reported maximum grain yields and minimum anthracnose severity in blackgram where treatments received four sprays of mancozeb (0.2%) at 10 days interval followed by 3 and 2 sprays. The increase in yield over check plot ranged between 12.1 to 65.8 per cent. However, maximum net profit of Rs. 7694 was recorded from plots where it received three sprays followed by four, two and one spray.

2.5.1 Crop loss model

Crop loss models are essential for estimation of losses due to the diseases. Accurate information, concerning losses is needed by growers and plant protection specialists to develop decision thresholds for determining when the cost effective control measures should be deployed (Nutter, 1993). Losses in grain yield due to disease may either be proportional to the area under the disease progress curve or to disease severity at some critical stages of host development. The relationship may be quantified in terms of critical point models (James, 1974).

Schneider *et al.* (1976) estimated cowpea yield loss due to *Cercospora* leaf spot by developing crop loss model in the form of Y = 0.43 × +14.95 (AUDPC) with a coefficient of determination of 70 per cent. Similarly, a crop loss model in the form of Y = 1553.8539 to 11.8442 × (AUDPC) with coefficient of determination of 75 per cent was developed to estimate yield loss due to blackgram leaf spot (Sud and Singh, 1985).

Benagi (1995) developed yield loss model in groundnut due to late leaf spot as Y = 1.318 + 0.03 (AUDPC) with coefficient of determination of 97.80 per cent.

2.6 Cultural and physiological studies

2.6.1 Cultural studies

2.6.1.1 Growth of fungus, effect of different solid and liquid media

Hegde (1967) obtained different monospore cultures of *Colletotrichum lindemuthianum* by isolating directly from typical lesions produced on bean pods while working with the cultivars and observed that semi-synthetic media were good for growth and sporulation of *C. lindemuthianum*.

Hiremath (1968) isolated *C. lindemuthianum* from infected bean pods by tissue culture technique on PDA.

Later Onesirosan and Sagay (1975) conducted experiment on anthracnose of cowpea, using acidified potato dextrose agar and living host. Accordingly, they found that, living host was found to be better than PDA for isolation of *C. lindemuthianum*. They obtained satisfactory results by using living host as selective medium for the isolation of the fungus.

Chacko *et al.* (1978) used seven different media for growth and sporulation of *Colletotrichum dematium* f. sp. *truncata*. The maximum growth of fungus was observed on Richard's agar (81.8 mm), while soil extract agar produced minimum growth (50.8 mm). Abundant acervuli were observed on Richard's agar, Czapek's agar and potato dextrose agar while they observed scanty acervuli on soil extract and maize meal agar media.

Genchev (1983) reported that glucose peptone agar medium supported the growth of *C. lindemuthianum* at pH 5.0 to 6.5.

Wong *et al.* (1983) during their cultural studies on *C. truncatum* found that oat meal agar medium was good for growth while Czapek's and potato dextrose agar for sporulation.

Hegde (1986) reported that the maximum radial growth of *C. gloeosporioides* was observed on Sabouraud's agar and potato dextrose agar. The fungus reached maximum growth after 10 days of incubation in potato dextrose broth. Highest mycelial weight was observed in Sabouraud's broth.

Singh and Shukla (1986) reported that Kirchoff's medium was found to be the best for growth and sporulation of *C. truncatum.* Czapeck's agar, Sabouraud's agar, Coon's Asthana and Hjawken's agar and Richard's agar media also supported good growth and sporulation.

Sinclair (1988) indicated that *C. truncatum* grew well on a variety of media like malt agar, potato dextrose agar, torula yeast agar, V-8 agar, oat meal agar as well as on non-synthetic media.

Drijfhout and Jansen (1989) evaluated the effect of culture media on spore production of *C. lindemuthianum.* Malt extract agar (MEA) gave the highest average spore production, over the ten races considered followed by neopeptone glucose agar (PGA), bean pod agar (BEA) and Czapeck's Dox agar (CDA).

Sinclair and Backman (1989) found potato dextrose agar and oat meal agar were good for *C. truncatum* growth.

Ekbote (1994) observed maximum radial growth of *C. gloeosporioides* on Richard's agar, potato dextrose agar and Browns agar. The fungus reached maximum growth after 12 days of incubation in potato dextrose broth. Highest dry mycelial weight was observed in Richard's broth and was least in Asthana and Hawker's broth.

Shirshikar (1995) opined that for *C. truncatum*, soybean seed extract dextrose agar medium was the best medium as it had recorded maximum mean colony diameter of 68.52 mm among 10 different media. Further, he reported that the fungus produced maximum mean mycelial dry weight of 99.85 mg on eighth day. Among the different liquid media tested, Richard's medium was found to be the best.

Mesta (1996) found that potato dextrose agar medium was the best solid medium for growth and sporulation in case of *C. capsici* and it reached maximum mycelial dry weight on 15^{th} day in potato dextrose broth. Among different liquid media tested, highest dry mycelial weight was observed in Richard's broth.

Murthy (1997) observed that potato dextrose agar medium was the best solid medium for *C. dematium*.

The induction of sporulation on different media for *C. lindemuthianum,* pod agar media was found best for growth and sporulation which was incubated at 20^0C in dark condition for 20 days (Pria *et al*., 1997).

Angadi (1999) found that among the solid media, potato dextrose agar was best for the growth of *C. capsici*. The maximum mycelial weight was observed after 16 days of incubation. Among the various liquid media used, maximum dry mycelial weight was observed in Richard's medium.

Varaprasad (2000) opined that Sabouraud's and Richard's agar supported good growth in *C. dematium*. Similarly, out of 10 liquid media tested, Richard's medium supported good mycelial growth and sporulation.

Mandeep and Munshi (2003) studied the effect of different media on the growth of *C. truncatum*. They concluded that the mycelial growth was more in potato dextrose and dox agar.

Laxman (2006) noticed that among semi-synthetic media, oat meal agar and synthetic media, Czapeck's dox agar media supported maximum growth of *C. truncatum*. The maximum mycelial weight was observed after 15 days of incubation.

2.6.2 Physiological studies

2.6.2.1 Effect of temperature

Lauritzen *et al.* (1933) noted that optimum temperature for infection of *C. lindemuthianum* was 22 to 25^{0}C and it could also infect the snap bean plants within a range of 7 to 33^{0}C. *C. lindemuthianum* required high humidity and comparatively low temperature (18 – 25^{0}C) for infection and sporulation.

Takimoto (1934) observed minimum and maximum temperatures for the growth of *C. lindemuthianum* on potato dextrose agar are 9 and 30^{0}C, respectively. Ujevic (1960) also found the range of temperature for the growth of the fungus as 5 to 30^{0}C, optimum being 22^{0}C.

According to Chowdhury (1957), the temperature required for optimum growth of *C. capsici* was 28^{0}C at 92 per cent relative humidity under laboratory condition.

Singh *et al.* (1977) reported that *C. capsici* grew best at 30^{0}C, but failed below 12^{0}C.

Chung and Bae (1979) while working with Ginsen anthracnose caused by *C. panacicala* observed that optimum temperature for mycelial growth and conidial formation was between 18 and 25^{0}C.

Mazhan and Sariah (1980) reported that the growth and sporulation of *C. capsici* were maximum at 30^{0}C with pH 5.0. Wong *et al.* (1983) reported optimum temperature for growth and sporulation of *C. truncatum* was 25 and 20^{0}C, respectively.

Ahmed (1982) reported that optimum temperature for growth and sporulation of *C. capsici* was found to be 30^{0}C, whereas temperature of 5, 10 and 40^{0}C did not favour growth of the fungus. Similarly, Asuti and Suhardi (1986) found that a temperature of 30^{0}C is favourable for the sporulation of *C. capsici*.

Kumarswamy (1983) studied percentage conidial germination of *C. capsici* which was maximum at 30^{0}C followed by 25^{0}C.

Hegde (1986) opined that maximum growth of *C. gloeosporioides* was obtained at 30^{0}C and temperature range for the good growth of fungus was 20 to 35^{0}C.

Singh and Shukla (1986) studied the influence of temperature on growth of the *C. truncatum* causing anthracnose of blackgram. They noticed increased mycelial weight with the increase in temperature upto 30^{0}C and maximum number of sporulation was found at 28^{0}C. Average mycelial growth was maximum at 25 to 30^{0}C and after 30^{0}C it decreased rapidly.

Abha Mishra and Om Gupta (1994) studied the effect of temperature on the radial growth, sporulation and spore germination of *C. dematium*, where they recorded maximum radial growth, sporulation and spore germination at 27^{0}C.

Ekbote (1994) opined that the optimum range of temperature for *C. gloeosporioides* was 20 to 29^{0}C. However, maximum growth of fungus was recorded at 29^{0}C temperature.

Shirshikar (1995) studied the effect of temperature on the growth of *C. truncatum*. The maximum mean colony diameter of 69.55 mm was obtained at 30^{0}C.

Murthy (1997) reported excellent mycelial growth and abundant acervuli production of *C. dematium* in culture at 25 and 30^{0}C.

Angadi (1999) observed that temperature of 30^{0}C was found to be favourable for growth of *C. capsici*.

Varaprasad (2000) studied the effect of temperature on the growth of *C. dematium*. The maximum growth of fungus was obtained at 27^0C.

Laxman (2006) found optimum range of temperature for the growth of *C. truncatum* as 25 to 30^0C. However, maximum growth of fungus was recorded at 30^0C.

2.6.2.2 Effect of relative humidity

Pepper anthracnose caused by *C. coccodes* was severe with increasing time of wetness duration from 0 to 60 hours. Fourty hours wetness was found optimum for disease severity (Hong and Byungkook, 1998).

Shirshikar (1995) studied the effect of different humidity levels for the recovery of *C. truncatum* and found that maximum recovery of 11.50 per cent was recorded in the seed samples which were exposed to 100 per cent humidity level.

Relative humidity of 95 to 100 per cent was found optimum for growth of *C. dematium* (Varaprasad, 2000).

Laxman (2006) observed that optimum range of relative humidity for the *C. truncatum* was 85 to 95 per cent. However, maximum growth of fungus was recorded at 95 per cent.

2.6.2.3 Effect of pH

Kumarsamy (1983) studied different pH levels on *C. capsici*, of which pH 6.0 was found to be optimum where maximum conidia were germinated.

Hegde (1986) reported that *C. gloeosporioides* grew well between the pH range of 5.0 to 7.0, but the growth was maximum at pH 6.0.

The effect of pH on the growth of *C. truncatum* was studied by Singh and Shukla (1986) and noticed the growth on wide range of pH 3.0 to 9.0. The optimum pH range was 5.5 to 7.5 and there was a significant reduction in growth of the fungus at pH lower than 5.5 and higher than 7.5.

Do and Paik (1987) studied the effect of pH on the approssorial formation and concluded that 6 to 8 pH was ideal.

Ekbote (1994) observed that the optimum range of pH for the *C. gloeosporioides* was 5.5 to 7.5. However, maximum growth of the fungus was recorded at 6.5 pH.

Shirshikar (1995) studied the effect of pH on the growth of *C. truncatum*. The pH range of 5.5 to 7.0 was found optimum for the fungal growth.

Angadi (1999) observed that *C. capsici* was favoured by acidic pH and maximum dry mycelial weight was obtained at pH 6.0.

Laxman (2006) found that optimum range of pH for the *C. truncatum* was 5.5 to 7.0. However, maximum growth of fungus was recorded at 6.5 pH.

2.6.2.4 Effect of light intensities

Ahmed (1982) reported that alternate cycles of light and darkness favoured both growth and sporulation of *C. capsici*.

Shirshikar (1995) studied the effect of light on the growth of *C. truncatum* and reported maximum growth of the fungus when exposed to alternate cycle of 12 h day light and 12 h darkness.

Murthy (1997) reported that the *C. dematium* exposed to alternate cycles of 12 h light and 12 h darkness recorded excellent mycelial growth and abundant acervuli production.

Varaprasad (2000) observed that alternate light (12 h) and darkness (12 h) supported good growth and sporulation in *C. dematium*.

2.7 Epidemiological studies

2.7.1 Aerobiology

Nair (1963) surveyed the fungal spore flora of vellore showing *Alternaria*, *Helminthosporium* and *Mucor* round the year. Baruah and Chettia (1966) used slides and

culture plates in Gauhati and recorded the presence of spores of *Cladosporium, Aspergillus, Penicillium curvularia* and *Alternaria*. Similarly, Vishnumittre and Khandelwal (1973) and Singh *et al.* (1980) reported the presence of number of fungal spores of which *Alternaria*, *Helminthosporium* and *Cladosporium* occurred round the year.

Kulkarni and Ramakrishnan (1977) used Burkard volumetric spore trap and recorded diurnal and seasonal variations in spore number in the atmosphere and noticed the influence of favourable humidity and temperature on the sporulation and spore discharge of *Drechslera oryzae*.

Kumar (1982) recorded a total of 52 types of fungal spores in two years of study during January 1979 to December 1980 of the air spora in a pine forest of Dehradun of which *Alternaria alternata* was found in major proportion. Tilak and Rao (1985) conducted the aerobiological experiments to monitor the atmospheric spore load of various plant pathogenic fungi over sunflower field during 1983 and 1984. *Alternaria* in the air was assessed. During 1983 spore concentration was 12,446 per m^3 of air whereas in 1984, it was 12,486 per m^3. The role of the meteorological factors on the decrease or increase of the spore concentration has been studied.

Thakur and Khare (1991) reported that maximum trapping of spores (*C. lindemuthianum* and *C. dematium*) in greengram was recorded when there was moderate temperature between 26 to 29^0C, relative humidity between 91 to 96 per cent, rainfall from 0 to 21.6 mm and wind velocity from 6 to 10 km per ha. Highest spore trap coincided with these conditions prevalent on July 30.

Mesta (2006) opined that the *Alternaria helianthi* spore load in the open air was found maximum during August and September months. Maximum temperature had negative correlation, while rainfall had positive correlation with spore load.

2.7.1.1 Model for prediction of disease

Benagi (1995) developed autoregressive model for the late leaf spot of groundnut by incorporating one variable pertaining to environmental factors at a time *viz.,* RH-I, II, maximum and minimum temperature. It was noticed that RH-I and II of *kharif* 1993 and RH-I and maximum temperature of *kharif*, 1994 were found to have negative relationship with the severity. The logistic model was also developed for understanding the development of late leaf spot during *kharif* 1993 and 1994.

Ashok Kumar *et al.* (1999) studied relationship between bean anthracnose and various weather variables during the course of epidemic and developed prediction models using logistic model and Gompertz model with correlation coefficient 0.99.

Amaresh (2000) developed prediction models for *Alternaria* blight of sunflower using autoregressive and logistic equations. The autoregressive model was given as $Y_{t+1} = 1.233\ Y_t$ with autoregressive coefficient of 0.98, whereas logistic model was given as $Y_t = 100/1+e$ with R = 0.97.

Mesta (2006) studied model for prediction of *Alternaria* blight disease in sunflower and developed prediction models using linear equations. The prediction model developed for $PDI = a + b_1t + b_2t^2$ for *kharif* 2004, where a = 5.72, $b_1 = 2.31$ and $b_2 = 0.3$ and $Y = a + b_1t$ for *kharif* 2005, where a = 0.07 and $b_1 = 5.89$. The R^2 value for both the years was 0.99.

2.7.2 Effect of sowing dates on the severity of disease

Rangaswamy *et al.* (1991) reported that in chilli *Colletotrichum* spp. was found to be very severe on August 28 sown crop compared to the September 18 and September 26 sown crop at GKVK, Bangalore.

Sridhara (1997) indicated that sowing of chilli in nursery in first fortnight of June and subsequent transplanting in first fortnight of July helps in reducing fruit rot disease.

Mittal (1998) found that the late sown crop (30^{th} June) suffered less anthracnose disease incidence consequently got more yield but there was more disease incidence in early sown (15^{th} May) and too late sown (15^{th} July) crop which suffered at later stages of growth from the disease incidence. Considering both the reduced disease incidence and increased

yield, second fortnight of June has been considered as the optimum time for sowing blackgram in the region.

2.7.3 Effect of environmental factors on disease

Chambers (1969) noticed that atleast two consecutive days of rain accompanied by cloudiness and high humidity are necessary for infection by *C. truncatum* in bean. Amount of rain was found to be less important than prolonged wetness of the plants by continued intermittent rain, only slight symptoms resulted when shorter periods of rain occurred.

Thakur (1988) reported that the intensity of the mungbean anthracnose has been found negatively correlated with the temperature. The most favourable weather conditions for the disease development were 26 to 30^{0}C temperature, 90 to 100 per cent relative humidity in the morning and 80 to 93 per cent at noon, rains with a wind velocity of about 13 km per h and the infection of the plants was favoured by overcast 2 and partially cloudy weather 1 but not by clear weather.

Thakur and Khare (1991) found that maximum increase in lesion size of greengram anthracnose was recorded when the relative humidity was 100 per cent followed by temperature, 27^{0}C and exposure to the light for extended period (72 h).

Ashok Kumar *et al.* (1999) studied recurrence and development of anthracnose disease (*C. lindemuthianum*) on kidney bean in relation to weather variables in sub-humid mid hill areas (Zone-II) of Himachal Pradesh. The studies revealed that heavy and frequent rains with moderate temperatures (19-25^{0}C) and high relative humidity (>70%) favoured the progress of disease in terms of vertical and horizontal spread. Correlation and regression analysis of the disease with weather factors further confirmed their role in the disease development.

2.7.4 Survival of *Colletotrichum*

Chona and Nariani (1954) reported that *C. falcatum* causing red rot in sugarcane survived upto five months in unsterilized and upto six months in sterilized soil.

Verma (1974) reported that, conidia of *Colletotrichum* sp. (*C. dematium*, *C. graminicola*, *C. gloeosporioides* and *C. atramentarium*) lost their viability within 90 to 170 days. Yoshida and Ashrita (1999) reported that survival period of *C. dematium* depends on length of incubation temperature. The fungus survived upto 120 days at 25 to 30^{0}C of incubation whereas at 0^{0}C the fungus survived upto 600 days.

Ahmed (1982) found that fungus *C. capsici* causing fruit rot of chilli survived upto eight months in both seed and culture. Sanathkumar (1999) observed that the *C. capsici* could survive upto 225 days on infected seeds stored under room condition, whereas on pedicel and fruit rind it survives for 195 days.

The survival studies carried out by Siddiqui *et al.* (1983) revealed that *C. dematium* survived for more than eight years in capsicum and mungbean, while *C. truncatum* survived more than 10 years in soybean seeds and *C. graminicola* survived for seven years, when the seeds were stored at 5^{0}C in germplasm unit.

Tu (1983) found that longevity of *C. lindemuthianum* varied greatly depending on environmental conditions. Moisture had a profound effect on its longevity. The fungus survived at least five years in infected pods and seeds of white bean that were air dried and kept in storage at 4^{0}C.

According to Rajkumar *et al.* (1989), *Colletotrichum* spp. survived with urdbean seed upto one year and indicated the potentiality of urdbean seed as a carrier of primary inoculum, further, germination percentage gradually increased with the increase in the storage period.

Shirshikar (1995) observed that in soybean five month old seed samples had 13.00 per cent seed infection when compared with 20 months old samples which recorded seed infection of 8.75 per cent, indicating the survival of *C. truncatum* upto 20 months.

Varaprasad (2000) found that the *C. dematium* survived upto 150 days in sterilized soil and upto 125 days in unsterilized soil, whereas under stored conditions (polythene bags), it survived upto 75 days and in exposed conditions, fungus could not survive beyond 25 days.

Deeksha and Tripathi (2002b) reported that the per cent survivability of *C. capsici* in infected crop debris of urdbean was significantly affected by duration of storage as well as placement at different depths in soil. The initial survivability of 100 per cent was completely lost within six months in debris placed at 20 and 25 cm depth and within seven months when debris was placed at 5, 10 and 15 cm depth. In case of infected seeds, the pathogen survived upto the next crop season (9 months), but the survivability decreased with lapse of time in both surface sterilized and non-sterilized seeds. However, the germination percentage of the seeds increased with storage time.

2.7.5 Effect of age of plant in relation to infection of *C. truncatum*

Fuse *et al.* (1981) reported that anthracnose occurred in early growth stage of soybean. Further, Sinclair (1982) also reported that soybean plants are susceptible to *C. truncatum* at each stage of plant development, particularly upto bloom stage.

Hepperly *et al.* (1983) in their studies on anthracnose of soybean in Peurto Rico recorded the appearance of symptoms on all parts at various plant growth stages.

Rahaman and Fakira (1985) reported that inoculation of two weeks old soybean seedlings caused upto 80 per cent mortality but plants were not died when inoculated after one month.

Shirshikar (1995) reported that the soybean plants inoculated at different age showed *C. truncatum* infection. The disease severity on 20 days old seedlings was 3.92, while it was 3.85 on 30 days old seedlings. However, the maximum disease severity of 4.10 was observed on 40 days old plants under 0 to 5 scale.

Varaprasad (2000) studied the effect of age of plant in relation to infection of *C. dematium* and observed that the maximum disease severity was observed on 21 days old plants. However, all the growth stages upto 35 days tested were found to be susceptible to infection.

Madhusudhan (2002) observed the relation of soybean plant age to anthracnose disease severity and reported that, early growth stage was found susceptible to the disease development. Maximum disease severity of 39.25 per cent was recorded on 40 days old seedlings.

2.7.6 Host range

Lenne *et al.* (1984) reported that *Colletotrichum* sp. has wide host range and it affects 24 species of 16 genera in leguminaceae family and 19 species of 17 genera in 17 other families of host plants including caricaceae, graminae, rutaceae and solanaceae.

Bharadwaj and Singh (1986) isolated *C. dematium* f. sp. *truncatum* from greengram, blackgram, soybean and horsegram and studied the pathogenic behaviour of these isolates. They undertook cross inoculation studies on six leguminous crops *viz.,* greengram, blackgram, cowpea, soybean, horsegram and adzukibean. Based on the pathogenicity test of these four isolates, they concluded that all isolates differed from each other in their pathogenic behaviour but blackgram and greengram isolates did not differ much. Whereas, isolates of horsegram and mungbean differed from each other in their pathogenicity on soybean and adzukibean. Mungbean and horsegram were susceptible to isolates from four host species and this differed pattern in pathogenicity of these isolates could be attributed to the existence of different pathogenic variates of *C. truncatum* in nature and their specific adoption to host species.

Sinclair (1988) reported that variability exists among the isolates of *C. truncatum* causing soybean anthracnose. Weidemann *et al.* (1988) in their cross inoculation studies on *C. truncatum* reported the host specificity of the pathogen in leguminous crops. Sinclair and Backman (1989) in their studies on soybean anthracnose indicated that *C. truncatum* varied considerably in pathogenicity.

Shirshikar (1995) studied the reaction of minor pulses *viz.,* greengram, blackgram and horsegram to soybean isolate of *C. truncatum*. It was observed that none of these minor pulses showed infection due to the *C. truncatum* indicating the host specificity of the pathogen.

Rajansharma and Kaushal (1999) opined that, *C. truncatum* possess the large host range and infects the legumes like adjukibean, mungbean, lentil, broadbean, arhar, cowpea, frenchbean and horsegram.

Sharma *et al.* (1999) studied the reaction of different isolates of *C. truncatum* from urdbean on related legumes like adjukibean, mungbean, lentil, fababean, arhar, soybean, cowpea, frenchbean and horsegram. *C. truncatum* had a wide host range among the local edible legumes infecting all the species tested.

Varaprasad (2000) studied that the reaction of six pulse crops *viz.,* pigeonpea, horsegram, cowpea, blackgram, greengram and soybean to chickpea isolates of *C. dematium*. It was observed that all the leguminous hosts tested were found susceptible to *C. dematium*.

Madhusudhan (2002) reported that, *Phaseolus vulgaris* L., *Amaranthus viridis* L., *Physalis minima* L., *Solanum nigrum* L. and *Acalypha indica* L., showed infection by *C. truncatum* indicating lack of host specificity of the pathogen.

2.8 Screening of genotypes for resistance to disease and biochemical factors of resistance

2.8.1 Screening of genotypes for anthracnose resistance

Minussi *et al.* (1975) studied the reaction of 60 bean (*Phaseolus vulgaris* L.) varieties to race BA-1 of *C. lindemuthianum* and reported 5 per cent cultivars as resistant and 88 per cent as susceptible.

Khare and Chacko (1983) screened 26 soybean varieties against anthracnose. The disease index ranged between 0 to 58.2 per cent. Five varieties *viz.,* Kalitur, EC-14437, Lee, N-670 and EC-2586 were completely free from disease under field conditions.

Oladiran and Oso (1983) conducted field trials of nine varieties of *Vigna unguiculata*. The severity of the disease caused by *C. truncatum* and *C. capsici* increased with age of the pods in all the varieties except Vika-1 and Kano 1696 and IAR 339, which were found moderately resistant.

Rahaman and Fakira (1985) reported reaction of soybean cultivars to anthracnose in Bangladesh. They observed that all the cultivars including Bragg, Clark-63 and Lee-74 were susceptible.

Singh and Shukla (1986) reported that most of the commercial varieties of blackgram have been found susceptible against anthracnose.

Manandhar *et al.* (1988) tested 414 soybean germplasm lines under controlled conditions and observed that none of the germplasm lines was completely free from anthracnose disease. It has been noted that the reaction of blackgram and greengram genotypes against the anthracnose pathogens is not consistent (Agarwal, 1989).

Thakur and Khare (1989) evaluated 27 cultivars of *Vigna radiata* against *C. dematium* and *C. lindemuthianum* and found that. varieties *viz*., Pusa 109 and Pusa 115 showed highly resistant and resistant reaction in one season of test but exhibited susceptible reaction next year.

Work carried out at AVRDC, Taiwan for the evaluation of soybean lines against anthracnose revealed that only five lines *viz.,* AGS-18, 128, 138, 139 and 151 recorded severity index below three and hence were classified as resistant (Anon., 1992b).

Shirshikar (1995) screened 42 cultivars of soybean and found none of the cultivars as either immune or highly resistant to the anthracnose disease. Twenty one cultivars showed susceptible reaction and 19 were found highly susceptible. One cultivar *i.e.,* NRC-1 was found moderately resistant and cultivar Durga showed resistant reaction.

Ghawde *et al*. (1996) conducted a field experiment at the College of Agriculture, Nagpur, India to evaluate seven varieties of soybean against *C. truncatum* under artificial epiphytotic conditions. Varieties JS-22 and PKV-1 were found highly resistant and MACS-3 showed resistant reaction.

Madhusudhan (2002) screened 60 genotypes of soybean under greenhouse condition. It was revealed that only three genotypes viz., (PK1129, DSb-2 and Cockerstaurt) were resistant 27 moderately resistant, 19 susceptible and 11 were highly susceptible to *C. truncatum*.

Rathaiah and Sharma (2004) reported that two greengram cultivars *viz.,* MLTG-2 and TRM-18 were highly resistant to *C. truncatum*.

Laxman (2006) observed that among different greengram genotypes screened through artificial inoculation, none were found resistant. All the screened genotypes showed the highly susceptible reaction except Pusa baisaki, which showed the susceptible reaction against *C. truncatum*.

2.8.2 Biochemical factors of disease resistance

2.8.2.1 Chlorophyll

Bhaskaran and Kandaswamy (1978) observed reduction of photosynthetic pigment in the necrotic halo and dead tissues of sunflower leaves infected by *Alternaria* leaf blight. The reduction of chlorophyll content may be due to reduction in synthesis of pigment or loss of chlorophyll.

Benagi (1995) noticed that reduction of chlorophyll was less in resistant varieties due to infection of late leaf spot in groundnut. Chlorophyll 'a' and 'b' were reduced as the disease progressed in leaf tissue of susceptible varieties.

Rajivkumar and Singh (1996) observed that there was decrease in chlorophyll 'a' and chlorophyll 'b' content in infected tissue due to the *Alternaria* leaf blight in sunflower as compared to healthy tissue. Chlorophyll 'a', 'b' and chlorophyll a/b ratio were also low in infected leaves as compared to healthy leaves of sunflower (Bhavani *et al*., 1998).

Amaresh (2000) noticed higher amount of chlorophyll 'a', chlorophyll 'b' and total chlorophyll in resistant sunflower genotypes for *Alternaria* leaf blight. Further, he noticed that there was decrease in all the above three biochemical constituents as a result of disease.

Mesta (2006) also reported that chlorophyll reduction was less in resistant varieties due to infection of *Alternaria* blight in sunflower.

2.8.2.2 Sugars

Benagi (1995) observed the increase in total sugar content in resistant genotypes of groundnut cultivars against late leaf spot, but increased in susceptible genotypes as the infection advanced. Higher amount of reducing sugars was observed in resistant genotypes and lower amount in susceptible genotypes at later stages of disease development.

Decrease in reducing and non-reducing sugars in the *Alternaria* sp. inoculated leaves when compared to uninoculated leaves of sunflower was noticed by Rajivkumar and Singh (1996).

Sindhan and Parashar (1996) reported that total, reducing and non-reducing sugars were low in early and late leaf spot resistant groundnut cultivars in comparison to susceptible ones. Further, they observed decrease in carbohydrates in all the cultivars after infection by early and late leaf spot pathogens due to rapid hydrolysis of sugars during pathogenesis through the enzymes secreted by pathogens.

Sindhan *et al.* (1999) noticed the higher contents of total sugar, reducing sugar and non-reducing sugar in healthy leaves of susceptible genotypes than of resistant ones. In diseased leaves their amount decreased in both resistant and susceptible genotypes due to infection by *Cercospora* leaf spot in mungbean.

Amaresh (2000) observed decrease in reducing and non-reducing sugar content in both resistant and susceptible genotypes of sunflower.

2.8.2.3 Phenols

Phenols are known to impart resistance against pathogens, because of their antimicrobial activity and it is often assumed that, their main role in plants is to act as protective compounds against disease causing agents such as fungi, bacteria and viruses

(Rohringer and Samborski, 1967). Concentration of phenolic compounds was usually higher in resistant than in susceptible genotypes of different crop plants (Arora and Wagle, 1985 and Saini *et al*., 1988).

Sharma *et al.* (1983) noticed that the level of phenolic compounds was slightly higher in the resistant maize inbred CM-104 as compared to the susceptible CM-600. Total phenols increased in both inbreds after inoculation of *Cochliobolus heterostrophus*. The increase in phenolics in susceptible inbred ranged from 1.3 to 6.1 per cent and that in the resistant from 1.6 to 11.6 per cent.

Sindhan and Parashar (1996) noticed changes in phenolic compounds in resistant and susceptible cultivars of groundnut. Total phenols and ortho-dihydroxy (O.D.) phenols were higher in healthy and infected leaves of resistant cultivars as compared to susceptible. After infection by early and late leaf spot pathogens, the concentration of phenolic compounds increased in all the cultivars.

Sindhan *et al.* (1999) reported that healthy leaves of resistant genotypes contained higher amount of total phenol and O.D. phenol than susceptible one. In diseased leaves, their amount increased in both the genotypes due to infection by *Cercospora* leaf spot in mungbean. The higher amount of phenolic compounds in diseased leaves may be due to either enhancement of synthesis or translocation of phenolics to the site of infection or hydrolysis of phenolic glycosides by fungal glycosides to yield free phenols, which helped in arresting the spread of the pathogen.

2.9 Disease management

2.9.1 *In vitro* evaluation of fungicides

Almeida and Yamashita (1977) reported the toxicity of fungicides to soybean pathogen *in vitro*. Benomyl, thiophanate methyl, chlorothalonil and mancozeb; all used at 1, 10 and 100 ppm were toxic to *C. truncatum* and other fungi. Maximum inhibition of mycelial growth was found with benomyl and thiophanate methyl.

Singh *et al.* (1993) conducted *in vitro* evaluation of 12 fungicides belonging to different chemical groups. Growth of *C. truncatum* was inhibited by carbendazim, benomyl, thiophanate methyl and carboxin.

Virendra Singh *et al.* (1993) evaluated 12 different fungicides against *C. truncatum* which was completely inhibited by carbendazim, benomyl, thiophanate methyl and carboxin. *In vitro* tests conducted by Vitti *et al.* (1993) revealed that benomyl and thiabendazole were highly effective against *C. truncatum*.

Among the non-systemic fungicides tested against *C. capsici* under *in vitro* conditions, mancozeb and captan were the best in inhibiting the growth of *C. capsici*. But, mancozeb, captan, chlorothalonil at 3000 ppm completely inhibited the fungal growth (Mesta, 1996).

Hegde (1998) noticed that, among non-systemic fungicides mancozeb at 3000 ppm showed maximum inhibition of *C. capsici* causing fruit rot of chilli followed by copper-oxychloride. Out of four systemic fungicides tested, carbendazim was found most effective than rest of the fungicides in inhibiting the mycelial growth of *C. capsici*.

While studying *in vitro* evaluation of fungicides against *C, dematium* (Varaprasad, 2000) found that carbendazim (0.1%), kitazin (0.3%) were found effective among systematic fungicide in inhibition the growth of fungus, while mancozeb (0.2 and 0.3%) was found to be superior among the non-systemic fungicides.

Madhusudhan (2002) tested ten fungicides *in vitro* against *C. truncatum.* Benomyl, carbendazim and prochloraz were found superior among systemic fungicides in inhibiting the growth of the fungus. Benomyl and carbendazim inhibited cent per cent mycelial growth of the fungus at all the three concentrations tested (0.025, 0.05 and 0.1%). SAAF was found to be superior among the non-systemic fungicides by inhibiting 99.22 and 85.92 per cent at 0.25 and 0.2 per cent concentrations, respectively.

Laxman (2006) found that among eight different systemic fungicides tested *in vitro* against *C. truncatum*, most of them showed complete inhibition of mycelial growth at the

concentration of 0.05 and 0.1 per cent except tricyclazole and hexaconazole. However, tricyclazole and hexaconazole were found most effective at 0.15 per cent. Among non-systemic fungicides, wettable sulphur was very effective at all concentrations followed by mancozeb.

2.9.2 *In vitro* evaluation of plant extracts

Gupta *et al.* (1981) reported that conidial germination of *C. capsici* was inhibited by Phytonoids of *Allium cepa* L., *Allium sativum* L., *Azardiracta indica* L.*, Ocimum basilicum* L. and *Leucas* spp.

The essential oil extracted from *Nigella sativa* L. exhibited strong antimicrobial activity against *C. capsici* (Rathee *et al*., 1982). Annapurna Jetti *et al.* (1987) reported that leaf extracts of *Polyalthia longifolia* L. inhibited the growth of *C. gloeosporioides*.

Mesta (1996) showed the superiority of neem leaf extract and onion bulb extract in inhibition of *C. capsici* growth causing fruit rot of chilli. Further, Shivapuri *et al.* (1997) noticed that among the plant extracts tested against five fungal pathogens including *C. capsici*, neem, *Datura stromonium* L., *Oscimum sanctum* L., *P. longifolia* and *Vinca rosea* L. were found more fungitoxic.

The extracts of ginger, garlic and neem gave excellent control of seed borne *C. truncatum* when soybean seeds were dipped for 30 min in these extracts (Hossain *et al*., 1999).

Gomathi and Kannabiran (2000) screened aqueous leaf extracts of 23 wild plants against the anthracnose fungi, *C. capsici* and *Gleosporium piperatum* Ell. and E8 infecting *Capsicum annuum* L. The leaf extracts of *Solanum tocum* SW., *Datura metel* L. and *Prosopis juliflora* (SW.) DC were found effective in reducing conidial germination and mycelial growth of these fungi. Further, Varaprasad (2000) found among six botanicals tested *in vitro* that leaf extract of *P. longifolia* @ 10 per cent concentration inhibited the growth of *C. dematium*.

Maximum inhibition of spore germination of *C. truncatum* by parthenium leaf extract was observed by Madhusudhan (2002) while evaluating 15 plant extracts *in vitro*.

Garlic (60.13%), neem (57.14%) and eucalyptus oil (61.93%) were found most promising botanicals against *C. truncatum*, which showed higher inhibition of mycelial growth at 10 per cent concentration (Laxman, 2006).

2.9.3 *In vitro* evaluation of biocontrol agents

Gupta *et al.* (1991) observed the growth inhibition of *C. lindemuthianum* on frenchbean by *Gliocladium virens, Trichoderma harzianum* and *T. viride* to 31.66, 44.66 and 88.33 per cent, respectively under *in vitro* condition.

Medereiros and Menezas (1994) studied the antagonistic potential of *T. harzianum* and *T. pseudokoningi* against *Colletotrichum gloeosporioides*, which showed a high degree of sensitivity for the antagonists.

Seven *Trichoderma* spp., seven isolates of *Pseudomonas fluorescens*, two isolates of *Pseudomonas fluorescens*, *Bacillus subtilis* and a yeast (*Saccharomyces cerveisiae*) were screened against *C. capsici,* both under *in vitro* and on chilli plants. Among the fungal antagonists, *S. cerevisiae* exhibited the maximum reduction of mycelial growth followed by *T. viride*. Among the 10 bacterial antagonists, *B. subtilis* showed the maximum growth reduction, followed by *P. fluorescens* isolate 27 (Jeyalakshmi *et al*., 1998).

Varaprasad (2000) tested six biocontrol agents for the control of *C. dematium*. Out of them, *T. koningii* (TNAU) inhibited maximum growth followed by *T. harzianum* (UASD).

The effect of antagonist alone and in combination of plant extract and chemicals was studied by Chandrasekaran and Rajappan (2002). In individual *Trichoderma viride* at 0.4 per cent showed 50 and 52 per cent disease index of leaf anthracnose and pod blight, respectively in combination with *Lawsonia inermis* at 1 per cent alum at 0.1 per cent and *Trichoderma* at 0.4 per cent through seed treatment and foliar spray recorded 11 and 6 per cent PDI of leaf anthracnose and pod blight, respectively.

Trichoderma harzianum, *T. viride*, *T. hematum, Gliocladium virens*, *Bacillus* sp. and *Ralstonia fluorescens* were screened for their efficacy against *C. capsici* causing anthracnose of bell pepper. *T. hematum* was found better biological control agent followed by *T. viride*, *Bacillus* sp. and *P. fluorescens* (Pathania *et al.,* 2004).

Laxman (2006) opined that among fungal bioagents tested, *T. harzianum* was found to be most effective in *C. truncatum* growth suppression followed by *T. viride*, whereas *Bacillus subtilis* (TNAU) isolate showed maximum mycelial growth suppression among bacterial bioagent.

2.9.4 *In vitro* evaluation of indigenous technology knowledge (ITK's)

Because of the inherent hazardous effects involved in conventional chemical management, the alternate plant protection measures like organic farming, use of FYM, green manuring, neem oil, botanicals and animal byproducts such as cow urine, butter milk as descried in Vedas, Arthashastra, Agnipuran, Surapala's (Wojtilla, 1985; Sadhale, 1996 and Nene, 2003) *etc.* are gaining importance.

Padmodaya (1996) reported the inhibitory effect of mahapanchagavya (MPG) on tomato wilt pathogen under *in vitro* condition and showed its superiority over carbendazim in reducing *Fusarium* wilt of tomato.

Seed treatment with skimmed milk powder, quick lime and thorough seed washing gave a level of control of common bunt of wheat (*Tilletia carries*) which was similar to that obtained with copper oxychloride control (Sollinger *et al.,* 1997).

According to Szczech (1999), addition of vermicompost to various container media significantly inhibited the infection of tomato plant by *Fusarium oxysporum* f. sp. *lycopersici*.

Bergen and Krishtensen (2001) examined that seed treatment of wheat with milk powder had fully controlled the *Tilletia tritici*.

Sridhar *et al.* (2002) reported that application of 50 ml of cow urine in 500 ml of water spraying on plants in early morning reduced the virus, fungus and bacterial disease incidence in vegetable crops. Manikandan (2005) studied that spraying of 200 ml of cow urine mixed with 2 litres of water was found effective in controlling the brinjal damping off in nursery.

Traditional methods form the basis of management of plant diseases in low input situations. Modified panchagavya mixture (mixture of cow milk, curd, ghee, dung and urine supplemented with yeast and common salts) was found most effective for the management of panama disease of banana (Jahagirdar *et al.,* 2003).

According to Kannan *et al.* (2005), combined application of soil drenching and foliar spray of sheep urine at 10 per cent was found most effective in reducing the incidence of groundnut stem rot to 9 per cent. With regard to yield, the same treatment registered the maximum yield of 1655 kg ha^{-1}, whereas controlled plot recorded 1053 kg ha^{-1}.

Priya and Kurucheve (2005) studied the effect of animal excrements on the conidial germination of *Cercospora personata.* Among the animal excrements tested, cow urine at 10 per cent, cow dung at 20 per cent and cow urine + cow dung (1:1) at 2.5 per cent concentration recorded complete inhibition of conidial germination.

Antifungal potential of panchagavya against *Rhizoctonia solani* causing damping off of cauliflower seedlings was evaluated by Sugha (2005). It inhibited 40 to 100 per cent mycelial growth and suppressed the disease by 78 to 82 per cent in nursery.

Vijayalakshmi *et al.* (2005) reported the effective control of chilli leaf spot by 10 per cent cow urine spray once in 10 days thrice followed by half-litre cow urine along with half litre sour butter milk mixed with nine litres of water once in seven days twice.

Banu and Rohini (2006) tested different dilutions of vermiwash against nematodes under *in vitro* conditions and found to be deleterious to varying extent. It greatly affected the juvenile hatching of *Meloidogyne incognita.* Undiluted vermiwash caused maximum nematode mortality and inhibition in hatching.

Raja *et al.* (2006) noticed that animal urine containing high nitrogen significantly reduced the *Macrophomina phaseolina, Fusarium oxysporum* f. sp. *lycopersici* and *Rhizoctonia solani*.

Sapre and Verma (2006) reported that cow urine and butter milk reduced the mycelial growth, number and size of sclerotia of *Rhizoctonia bataticola*. The mycelial growth was completely inhibited by butter milk at 500 and 1000 ppm whereas at 100 ppm, it was the least. Cow urine and butter milk not only reduced the mycelial growth of *Sclerotium rolfsii* but also affected the viability of sclerotia formed and reduced number of sclerotia per plate. Smallest sclerotia were recorded in butter milk followed by cow urine, slight reduction in mycelial growth of *Fusarium solani* f. sp. *glycine* by cow urine and butter milk. Conidial production decreased with increasing concentration of cow urine.

Kannan *et al.* (2007) reported that in pot and field studies, foliar spray of combined application of buffalo urine and sheep urine (1:1 v/v) at 5 per cent concentration on peanut completely inhibited the mycelial growth, production and germination of *Sclerotium rolfsii*.

Panchagavya - an organic formulation evaluated *in vitro* for its antifungal activity against *Curvularia lunata* in rice was found to be the dominant pathogen in causing grain discolouration (Sumangala and Patil, 2007).

2.9.5 Integrated disease management

Bharadwaj and Thakur (1991) conducted field trials, where they used carbendazim (0.1%), captafol (0.25%) and mancozeb (0.25%) applied alone as single sprays at 60 days, two sprays at 45 and 60 days or three sprays at 45, 60 and 75 days or in sequence one after the other on a three sprays schedule at 45, 60 and 75 days after sowing for the control of leaf spot and pod blight of urdbean, caused by *C. dematium* f. sp. *truncatum*. All three fungicides reduced the disease severity on foliage, however, three sprays schedule was found more effective than the single or two spray carbendazim than mancozeb or captan.

Shirshikar (1995) found that either benomyl or carbendazim seed treatment at the rate of 3 g per kg of seed along with one foliar spray at the rate of 0.1 per cent on 30th day after sowing was effective in controlling the soybean anthracnose.

Chandrasekaran *et al.* (2000) studied the effect of a plant extract, an antagonist and a fungicide treatment both individually and in combination. Seed treatment with alum (0.1%) recorded the highest seed germination of 90 per cent, compared to 68.0 per cent in the untreated control. Seed treatment followed by a foliar spray with *Lawsonia inermis* leaf extract (1%) and alum (0.1%) recorded leaf anthracnose and pod blight incidence of 7.0 and 4.2 per cent, respectively with a grain yield of 2191 kg ha^{-1}. Seed treatment with *L. inermis* (1%) + *Trichoderma viride* (0.4%) + alum (0.1%) registered 7.4 per cent leaf anthracnose, 5.6 per cent pod blight incidence and yield of 2186 kg ha^{-1}.

Varaprasad (2000) studied on integrated approach for the management of chickpea blight disease. Out of six treatments tested, seed treatment with carbendazim @ 2 g per kg + two foliar spray of SAAF (0.05%) at 15 days interval gave maximum reduction in disease incidence followed by seed treatment with carbendazim @ 2 g per kg + foliar spray with SAAF 0.05% and *Polyalthia longifolia* (10%) extract at 15 days interval.

Deeksha and Tripathi (2002c) studied on management of blackgram anthracnose and found that seed treatment followed by two prophylactic sprays of bavistin or tilt @ 0.1% each at 15 days interval showed minimum disease severity and maximum grain yield followed by contaf (0.1%) and Indofil M-45 (0.2%) sprayed plots. Among the biocontrol agents, *Gliocladium virens* gave better results than *Trichoderma harzianum*.

Madhusudhan (2002) observed that either benomyl or carbendazim seed treatment at the rate of 2 g per kg of seed along with two foliar applications at 0.1 per cent on 30 and 45th day of sowing was found effective in controlling the soybean anthracnose.

Laxman (2006) studied the efficacy of different fungicides and biorationals under field condition against greengram anthracnose and found that among different treatment, the least disease incidence was observed in propiconazole followed by hexaconazole and carbendazim. But, in case of biorationals, the least disease incidence was noticed in azadirachtin.

3. MATERIAL AND METHODS

The present investigations were carried out in the laboratory and field during 2006-07 and 2007-08. All the field experiments were conducted at the Agricultural Research Station, Bidar, University of Agricultural Sciences, Dharwad, Karnataka, while laboratory experiments were undertaken at Department of Plant Pathology, College of Agriculture, University of Agricultural Sciences, Dharwad, Karnataka.

Bidar is situated in Northeastern Transitional Zone of Karnataka at $70^0$45' North latitude and $76^0$45' East longitude and at an altitude of 699 m above mean sea level. This zone receives a fairly well distributed rainfall of about 870 mm from June to October. On an average, the mean daily maximum and minimum temperatures are 36.4^0C and 20.8^0C, respectively. The relative humidity on an average, varies between 48 and 90 per cent. The meteorological data recorded during the experimental period at Bidar are presented in Appendix I and II. The materials used and methods followed in conducting the experiments are described in this chapter.

3.1 General laboratory procedures

3.1.1 Glasswares cleaning

For all laboratory experimental studies, Corning and Borosil glasswares were used. Wherever required, they were kept in the cleaning solution containing 60 g potassium dichromate ($K_2Cr_2O_7$) and 60 ml of concentrated sulphuric acid (H_2SO_4) in one litre of water for a day. Then, they were cleaned by washing with detergent followed by rinsing several times in tap water and finally in distilled water.

3.1.2 Sterilization

All glasswares, solid and liquid media were subjected to sterilization by autoclaving at 1.1 kg per cm^2 (121^0C) for 20 minutes. The plant tissues were surface sterilized in 1:1000 mercuric chloride solution followed by three changes in sterile water. All cultural studies were conducted in aseptic condition under laminar flow. The tips of inoculation needle, forceps and cork borers were sterilized under flame.

3.2 Survey and surveillance for anthracnose severity

Roving method of survey was conducted during *kharif* 2006-07 and 2007-08 in three taluks of Bagalkot, one taluk of Belgaum, two taluks of Bellary, five taluks of Bidar, two taluks of Bijapur, four taluks of Dharwad, three taluks of Gadag, four taluks of Gulbarga, three taluks of Haveri, two taluks of Koppal and two taluks of Raichur districts. In each taluka, minimum of five villages were selected and in each village, minimum of five fields were selected to assess the severity of greengram anrhracnose.

The anthracnose severity was recorded by following 0 – 9 scale of Mayee and Datar (1986) (Plate 1a and 1b).

Scale

Category	Description
0	No symptoms on leaves
1	Small pin-head size lesions covering 1% or less leaf area
3	Small pin-head size lesions covering 1-10% of leaf area
5	Lesions big but not coalescing, covering 11-25% of the leaf area
7	Lesions on leaves covering 26-50% of leaf area. Cankers on stem and pod infection
9	Lesions on leaves covering 51% or more of leaf area. Defoliation of leaves, deep cankers on stem and pods, blighting of plant occurs

Plate 1a: Disease scoring scale of anthracnose of greengram using 0-9scale (upper surface)

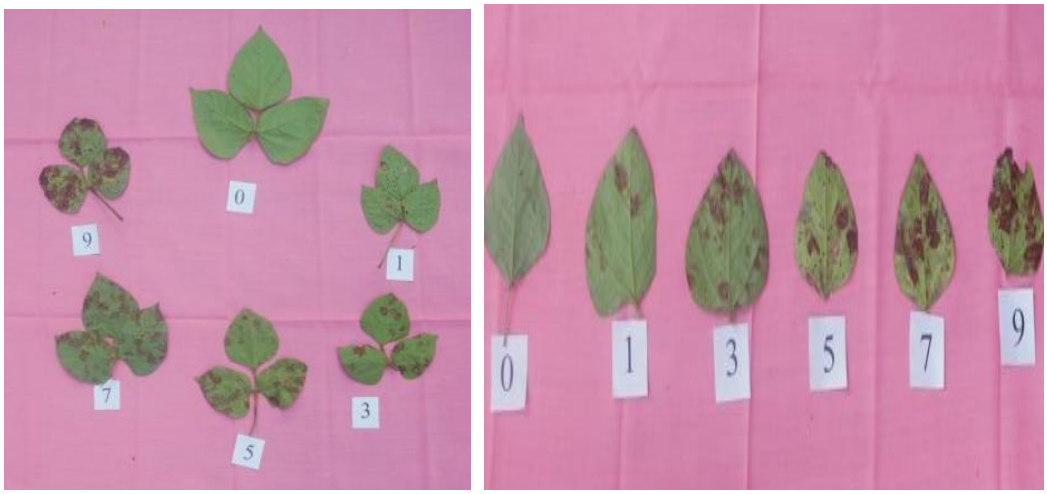


Plate 1b: Disease scoring scale of anthracnose of greengram using 0-9 (lower surface)

Further, these scales were converted to per cent disease index (PDI) using the formula given by Wheeler (1969).

$$PDI = \frac{\text{Sum of individual disease ratings}}{\text{No. of observations assessed}} \times \frac{100}{\text{Maximum disease rating}}$$

3.3 Collection of diseased specimens, isolation and maintenance of culture

During the field survey carried out in *kharif* 2006, a large number of anthracnose infected greengram leaf samples were collected from different places. Standard tissue isolation procedure was followed to isolate the causal fungal pathogen. The infected tissues of the leaves and twigs were cut into small bits of 1 to 2 mm size and surface sterilized in 1:1000 mercuric chloride ($HgCl_2$) solution for one minute and washed repeatedly twice in sterile distilled water to remove the traces of mercuric chloride before transferring them to sterile potato dextrose agar (PDA) slants under aseptic conditions. The slants were incubated at a temperature of 27 $\pm$ 1^0C and observed for fungal growth. Further, the pure culture of the fungus was obtained by single spore isolation method.

3.3.1 Single spore isolation

Ten ml of clear, filtered two per cent water agar was poured into sterile petriplates and allowed to solidify. Dilute spore suspension was prepared in sterilized distilled water from 15 days old culture. One ml of such suspension was spread uniformly on agar plate. These plates were incubated at 27 $\pm$ 1^0C for 12 hr. Then such plates were examined under microscope to locate single isolated and germinated conidium and marked with ink on the surface of the plates.

The growing hyphal tip portion was transferred to PDA slants with the help of cork borer under aseptic conditions and incubated at 27 $\pm$ 1^0C. Such culture tubes were used for further studies.

3.3.2 Proving the pathogenicity

Artificial inoculation of the fungus was carried out to prove the pathogenicity. The greengram seedlings were raised in pots and inoculated with fungal suspension (1×10^{10} suspension) by spraying the foliage and incubated in growth chamber to maintain high relative humidity (90%) for expression of symptoms. The inoculated plants were monitored for expression of symptoms which appeared one week after incubation. Anthracnose appeared as brown spots on lower surface of the leaf. The fungus was re-isolated from infected leaf of the plant and the culture obtained was compared with original culture for confirmation.

3.3.3 Maintenance of the cultures

The fungus was sub-cultured on potato dextrose agar (PDA) slants and allowed to grow at 28 $\pm$ 1^0C for 12 days, such slants were preserved in refrigerator at 5^0C and maintained. Sub-culturing was done once in a month, such cultures were used throughout the study, virulence of the fungus was maintained by passing through the host after every three months.

3.4 Assessment of loss due to anthracnose of greengram

Field experiments were conducted during *kharif* 2006 and 2007 at Agricultural Research Station, Bidar to assess the losses due to anthracnose of greengram using a susceptible variety Chinamung. Three treatments were imposed with 0.1 per cent carbendazim besides a treatment with unsprayed control. The details of the treatments are furnished below.

Sl. No.	Treatment	Spray schedule
1.	T_1	One spray (20 days after sowing (DAS))
2.	T_2	Two sprays (20 and 30 DAS)
3.	T_3	Three sprays (20, 30 and 40 DAS)
4.	T_4	No spray (control)

Experimental details

Design	: Randomized Block Design (RBD)
Replications	: Five
Plot size	: 3.0 × 5.0 m
Variety	: Chinamung
Season	: *Kharif* 2006 and 2007

The disease severity was recorded at different stages of crop growth. Necessary insecticides were sprayed at suitable concentrations to control insect pests.

Observations recorded

a) Per cent disease index (PDI)

The intensity of the disease was recorded by scoring all the tagged ten plants in each treatment using 0 to 9 scale of Mayee and Datar (1986). Further, the PDI was calculated with the above scales using the formula of Wheeler (1969) at weekly interval.

b) Area under disease progress curve (AUDPC)

Further, the area under disease progress curve (AUDPC) was calculated by using the formula as suggested by Wilcoxson *et al*. (1975).

$$AUDPC = \sum_{i=1}^{k} \tfrac{1}{2} (S_i + S_{i-1}) (T_i - T_{i-1})$$

Where,

S_i = Severity of anthracnose at the end of time i

k = Number of successive evaluation of anthracnose of greengram

$T_i - T_{i-1}$ = Time interval between two evaluations i and i-1 of the disease

c) Yield

Crop was harvested after maturity of pods and grain and stalk yields of net plot were recorded as kg per ha and later expressed in quintals per ha.

d) Per cent reduction over control

$$\text{Disease reduction (\%)} = \frac{\text{Disease severity in control} - \text{Disease severity in treatment}}{\text{Disease severity in control}} \times 100$$

e) Per cent yield increase over control

$$\text{Yield increase (\%)} = \frac{\text{Yield in treatment plot} - \text{Yield in control plot}}{\text{Yield in control plot}} \times 100$$

f) Per cent loss in grain and stalk yield

The per cent loss in grain and stalk yield was calculated by using following formula.

$$\text{Per cent yield loss} = \frac{Yp - Yx}{Yp} \times 100$$

Where,

Yp : Potential yield

Yx : Yield when per cent disease severity is x

g) Benefit:Cost Ratio (BCR)

Total cost incurred for application of fungicides including cost of fungicides and labours were calculated. Additional benefit due to increased yield in each treatment over

control was worked out and benefit cost ratio was calculated using additional benefits and total costs.

3.4.1 Crop loss model

An attempt was made to identify the relationship between yield and severity of disease in the form of PDI calculated at weekly intervals starting from 25 to 60 DAS of the crop.

Crop loss models for anthracnose of greengram were developed using simple linear regression functions in the form of Y = a + bx with Y as yield in quintals and x as per cent disease index. The models were developed for two consecutive years 2006 and 2007. The coefficient of determination (R^2) was calculated to know the extent to which the model is capable of explaining the variation.

3.5 Cultural and physiological studies of *Colletotrichum truncatum*

3.5.1 Cultural studies

3.5.1.1 Growth phase of *Colletotrichum truncatum* on liquid media

Thirty ml of potato dextrose broth (PDB) was added into each of 150 ml conical flasks and sterilized. The growth of the fungus was studied at 3, 5, 7, 9, 11, 13, 15, 17, 19 and 21 days of inoculation. The flasks were then inoculated with 5 mm disc of *Colletotrichum truncatum* from actively growing culture and incubated at 27 $\pm$ 1^0C. Each treatment was replicated three times. Three flasks were harvested separately at a time, starting from the third day onwards upto 19^{th} day by leaving a gap of 48 h between the two successive harvests. The cultures were filtered through previously weighed Whatman No. 42 filter paper of 12.5 cm diameter, which were dried to a constant weight at 60^0C in an electric oven prior to filtration. The mycelial mat on the filter paper was thoroughly washed with sterile distilled water to get rid of the salts likely to be associated with the mycelial mass.

The filter paper along with the mycelial mat were dried to a constant weight at 60^0C for 48 h, cooled in a desiccator and weighed immediately on an analytical balance. The difference between final and initial weight of filter discs were taken as the weight of the mycelia. The data were analysed statistically.

3.5.1.2 Growth of *Colletotrichum truncatum* on different solid media

The cultural characters of the *C. truncatum* were studied on the following ten different solid media and the best media for the fungus growth was identified.

1. Potato dextrose agar
2. Oat meal agar
3. Host extract agar
4. Czapek's agar
5. Malt extract agar
6. Sabouraud's agar
7. Yeast extract agar
8. Richard's agar
9. Potato carrot agar
10. Corn meal agar

The composition and preparation of the above mentioned synthetic and non-synthetic media were obtained from Ainsworth and Bisby's Dictionary of the fungi (Hawksworth *et al.*, 1983). The composition of the media is given below.

1) Potato dextrose agar (PDA)

In most of the experimental studies, the potato dextrose agar (PDA) was used. The composition of PDA is as follows.

Potato peeled	200 g
Dextrose	20 g
Agar-agar	20 g
Distilled water	1000 ml (volume to make up)

Two hundred gram of peeled potatoes were cut into small bits and boiled in distilled water and the extract was collected by filtering through muslin cloth.

Dextrose 20.0 g and agar 20.0 g each were dissolved in the potato extract and the final volume was made upto 1000 ml with distilled water and sterilized as described earlier and preserved for further use.

2) Oat meal agar (OMA)

Oat flakes	30 g
Agar-agar	20 g
Distilled water	1000 ml (volume to make up)

Oat flakes were boiled in 500 ml distilled water for 30 min and filtered through muslin cloth. Agar was melted in 500 ml distilled water separately. Both the solutions were mixed thoroughly and the volume was made upto 1000 ml and was sterilized.

3) Host extract agar (HEA)

Healthy greengram plant shoots	200 g
Agar-agar	20 g
Distilled water	1000 ml (volume to make up)

Greengram shoots were boiled in 500 ml water for 30 min. Extract was collected by filtering through muslin cloth. The agar was melted in 500 ml water, both the solutions were mixed and the volume was made upto 1000 ml and was sterilized.

4) Czapek's agar

Sucrose ($C_6H_{12}O_6$)	30 g
Sodium nitrate ($NaNO_3$)	20 g
Potassium dihydrogen phosphate (K_2PO_4)	1.0 g
Magnesium sulphate ($MgSO_4$-$2H_2O$)	0.5 g
Potassium chloride (KCl)	0.5 g
Ferric chloride	0.01 g
Agar-agar	20 g
Distilled water	1000 ml (volume to make up)

Agar-agar was melted in 500 ml distilled water. Two solutions were mixed thoroughly and the volume was made upto 1000 ml and was sterilized.

5) Malt extract agar

Malt extract	20 g
Agar-agar	20 g
Distilled water	1000 ml (volume to make up)

All the ingredients were dissolved in 400 ml distilled water and agar was dissolved separately in 500 ml of distilled water and mixed with the above solution and the volume was made upto one litre. The medium was sterilized at 1.1 kg per cm^2 pressure for 15 minutes.

6) Sabouraud's agar

Dextrose	40 g
Peptone	10 g
Agar-agar	20 g
Distilled water	1000 ml (volume to make up)

All the ingredients were dissolved one by one in 400 ml distilled water and agar was dissolved separately in 500 ml distilled water and mixed with the above solution and the volume was made upto one litre before sterilization.

7) Yeast extract agar

Yeast	20 g
Agar-agar	20 g
Distilled water	1000 ml (volume to make up)

Twenty gram of yeast extract was dissolved in 400 ml distilled water and in other 400 ml distilled water dissolved 20 gram agar-agar and both the solutions were mixed together. The volume was made upto 1000 ml and then subjected for sterilization.

8) Richard's agar

Sucrose	50 g
Potassium dihydrogen phosphate	5 g
Potassium nitrate	10 g
Magnesium sulphate	2.5 g
Ferric chloride	0.02 g
Agar agar	20 g
Distilled water	1000 ml (volume to make up)

All the above ingredients, except potassium dihydrogen phosphate and agar were dissolved in 450 ml of distilled water. Agar was melted separately in 500 ml of distilled water and was mixed with the above solution. The volume was made upto 950 ml. Potassium dihydrogen phosphate was dissolved in 50 ml of distilled water. The two solutions were autoclaved and subsequently mixed together.

9) Potato carrot agar

Grated potato	20 g
Grated carrot	20 g
Agar agar	20 g
Distilled water	1000 ml (volume to make up)

Boil grated vegetables for 1 hr. in the tap water. Strain through fine sieve, add agar. Boil over water bath till agar dissolves, sterilize at 15 p.s.i. for 20 min.

10) Corn meal agar

Corn flakes	60 g
Agar agar	20 g
Distilled water	1000 ml (volume to make up)

Sixty grams of dehydrated corn flakes were boiled for 15 min in 500 ml of distilled water and filtered. Twenty g of agar was melted separately and both the solutions were mixed. The volume was made upto 1000 ml.

Twenty ml of each medium listed above was poured into 90 mm diameter Petri plates. After solidification, 5 mm discs of *Colletotrichum truncatum* from actively growing culture were cut using a cork borer and a single disc was placed upside down at the centre of Petri dish. Each set of experiment was replicated thrice and the plates were incubated at 27 $\pm$ 1^0C. The measurements of the colony diameter was taken when the maximum growth was attained in any one of the media tested. Then, cultural characters such as colony diameter, colony colour, type of margin and sporulation were also recorded. The sporulation was graded as follows.

Sporulation grade

Sl. No.	Score	Grade	Description (conidia/ microscopic filed [100 X])
1.	++++	Excellent	>150
2.	+++	Good	101 – 150
3.	++	Fair	51 – 100
4.	+	Poor	1 – 50
5.	-	No sporulation	-

3.5.1.3 Growth of *C. truncatum* on different liquid media

The composition and preparation of different liquid media used were the same as that of solid media except that agar was not added. Twenty ml of different liquid media were added into each of 100 ml conical flasks. These flasks were then sterilized at 1.1 kg per cm^2 pressure for 20 min. The flasks were inoculated with 5 mm mycelial discs obtained from periphery of 10 days old culture and incubated at 28 $\pm$ 1^0C for 15 days. Each treatment was replicated thrice. Dry mycelial weight and sporulation (under high power) in each treatment were recorded as described earlier.

3.5.2 Physiological studies

3.5.2.1 Effect of temperature on the growth and sporulation of *C. truncatum*

The growth of *C. truncatum* was tested at 10, 15, 20, 25, 30, 35 and 40^0C. Potato dextrose agar was poured into 90 mm diameter petriplates. After solidification, 5 mm disc from actively growing cultures were cut and inoculated to solidified petriplates and incubated for 15 days in the incubators adjusted to required temperature levels. Each treatment was replicated thrice. After incubation period, radial growth and sporulation from solid media were recorded as described earlier.

3.5.2.2 Effect of relative humidity (RH) on growth and sporulation of *C. truncatum*

Five mm discs of ten day old culture of *C. truncatum* of greengram were placed at the centre of petridish containing PDA media under aseptic condition and petridish were exposed to 65, 75, 85, 95 and 100 per cent relative humidity levels maintained in desicators. Different levels of relative humidity were created by using different concentration solutions of H_2SO_4. The desicators were kept at 27 $\pm$ 1^0C with four replications. Observations of colony diameter and sporulation were recorded 13 days after incubation.

3.5.2.3 Effect of different pH on growth and sporulation of *C. truncatum*

Colletotrichum truncatum was grown on 30 ml potato dextrose broth with selected pH range of 4.0, 4.5, 5.0, 5.5, 6.0, 6.5, 7.0, 7.5 and 8.0. The pH levels were adjusted by adding 1 N alkali (NaOH) or acid (HCl). A seven day old five mm mycelial disc from actively growing culture were inoculated separately into conical flasks containing 30 ml medium at different pH levels. Three replications were maintained for each pH level. These flasks were incubated at 27 $\pm$ 1^0C for 10 days. The mycelial growth was harvested and dried in hot air oven and the dry weights were recorded by using electronic digital balance and sporulation was recorded as described earlier.

3.5.2.4 Effect of light intensities on growth and sporulation of *C. truncatum*

The effect of light intensities on the growth and sporulation of *C. truncatum* was studied by exposing the culture to following treatments.

1) Alternate period of 12 hr light under day light tubes and 12 hr darkness.
2) Continuous light under day light tubes.
3) Continuous darkness.

Twenty one petridishes were prepared with 20 ml of potato dextrose agar medium in each. The petridishes were inoculated aseptically with 5 mm mycelial disc from ten days old culture. Seven replications were maintained for each treatment. The petridishes of each treatment were exposed to alternate period of 12 hr light under day light tubes and 12 hr of darkness, continuous light under day light tubes and continuous darkness for eight days. Observations on fungal growth and sporulation were recorded.

3.6 Epidemiological studies

3.6.1 Aerobiology

Aerobiological studies were carried out to trap the conidia of *Colletotrichum truncatum* present in the air current during *kharif* 2006 and 2007. For this, aeroscope exposure of stationary slide was done by mounting it on a wind wane and placed inside greengram field at ARS, Bidar. In both the years, the crop was sown on first day of 24^{th} standard week and harvested during 32^{nd} standard week. The observations were made till the harvest of the crop.

A slide, which was smeared with a thin layer of vaseline was used for trapping spores, by keeping smeared slide in the slot inside the box. The slide was removed every day at 08.30 hr. Average number of conidia per microscopic field was recorded under low power taking count of ten microscopic fields on a slide. Appearance of anthracnose disease on greengram crop in the aerospore installed field was recorded. Observations were made daily to record the first appearance of the disease in the field. In addition, the severity of disease was also recorded at weekly intervals starting from first appearance. Meanwhile, the weather data *viz.,* maximum and minimum temperature, morning and evening relative humidity and rainfall received during the period of aerobiological studies were recorded (Appendix I and II).

The information obtained from these observations were studied in relation to weather factors *viz.,* minimum and maximum temperature, rainfall and relative humidity (morning and evening) prevailed during the crop period by following standard statistical methods. The multiple regression equation was developed for estimation of spore load and PDI by taking weather parameters as input variables.

3.6.1.1 Effect of weather parameters in relation to spore load and severity of the disease

An attempt was made to study the effect of weather factors in relation to spore load and disease severity by subjecting the data to regression analysis. The weather parameters were correlated to weekly spore load and weekly per cent disease index by calculating the Karl Pearson's correlation coefficient (r) as given below.

$$r = \frac{\sum_{i=1}^{n} (x_i - \bar{x})\,(y_i - \bar{y})}{\sqrt{\sum_{i=1}^{n} (xi - \bar{x})^2 \sum_{i=1}^{n} (yi - \bar{y})^2}}$$

Where,

x and y are two variables

$\bar{x}$: Mean of x

$\bar{y}$: Mean of y

r : Karl Pearson's correlation coefficient

Further, the data were subjected to multiple linear regression analysis to find out the linearity of the independent variables for prediction.

3.6.1.2 Disease prediction models

The weekly disease severity was graphically analysed for estimation of disease development and to predict the intensity. The 2nd degree polynomial model was found suitable to estimate the disease progression.

$$Yx = a + b_1x + b_2x^2$$

Where,

Yx : Expected disease severity at time x
x : Time interval is seven days
a : Intercept
bi : Coefficient

Where, a and bi are intercept and regression coefficients, respectively for Yx indicated expected disease severity at time x of seven days interval (Cox and Hinkley, 1979 and Snedecor and Cochran, 1994).

3.6.2 Effect of date of sowing on the severity of anthracnose and it's correlation with weather factors

A field experiment was conducted during *kharif* 2006 and 2007 at ARS, Bidar to assess the progress of anthracnose at different time interval in different dates of sowing. A replicated field trial was carried out to explore the possibility of disease escape.

The experiment was conducted in randomized block design with four replications. The first date of sowing was done with highly susceptible variety Chinamung on 4th June and subsequent sowings were done at weekly interval. Totally six different dates of sowings were undertaken. The severity of anthracnose was recorded on five randomly selected plants using a disease rating scale 0 to 9. Further, these ratings were converted to per cent disease index (PDI) as explained earlier. The meteorological data for the experimental period was collected and correlated with anthracnose severity.

3.6.3 Viability and survival of conidia of *C. truncatum*

3.6.3.1 Survival on infected leaves

The present investigation on viability and survival of *C. truncatum* was undertaken as a part of epidemiological study during 2006-07 at Agricultural College, Dharwad (Karnataka) to obtain information about the perpetuation of the pathogen during the off-season. The freshly anthracnose infected leaves of greengram plant were collected and stored under different storage conditions *viz.,* freeze (4 - 5^0C), under tree shade (18 – 22^0C), room temperature (20 – 25^0C), glasshouse (25 – 28^0C) and field condition (28 – 30^0C) in separate lots.

Per cent germination of conidia on each type of stored leaf was recorded before their preservation. The viability of conidia on leaf under different storage conditions were regularly examined by checking germination under microscope.

3.6.3.2 Survival in infected seeds

Survival study of *C. truncatum* on greengram seeds was undertaken for a period of 12 months. For this purpose, freshly harvested anthracnose infected seeds were used. The seed samples were drawn at regular monthly interval and each time 400 seeds were subjected to standard blotter test technique to evaluate *C. truncatum* seed infection. Similarly, viability of greengram seeds during each observation was also recorded by subjecting 100 seeds from each seed sample to standard germination test and per cent germination was worked out (Anon., 1996).

3.6.4 Effect of plant age in relation to infection of *C. truncatum*

An experiment was planned during *kharif* 2006-07 to study the effect of age of the crop on anthracnose severity. The healthy seeds of greengram cultivar Chinamung were used in the experimentation.

Staggered sowing at an interval of five days was done in earthen pots containing sterilized soil kept in the glasshouse so as to get 5, 10, 15, 20, 25, 30, 35, 40, 45 and 50 days old plants for simultaneous inoculations. Three pots for each plant age group were maintained. Spray inoculation with conidial suspension (10^{10} conidia/ml) was done to all the plants of different age groups. The inoculated seedlings were covered with polythene hood for 48 hr to create high humidity. Observations on disease severity were recorded on the 10th day after inoculation using 0 to 9 scale and per cent disease index was calculated.

3.6.5 Host range studies

The objective of this study was to identify the collateral hosts of *C. truncatum* other than greengram. Different pulses raised in pots were inoculated artificially by spraying conidial suspension of greengram isolates of *C. truncatum*. Inoculated plants were covered with polythene bags overnight. Observations were recorded 10 days after inoculation on the development of symptoms on different pulses.

3.6.6 Cross inoculation studies

Isolates from different diverse pulses were tested for their ability to infect greengram crop. Isolates were collected from several diverse pulses infected by anthracnose in the field and conidial suspension were prepared and then spraying conidial suspension on to the healthy greengram leaves. Similarly, conidial suspension from greengram were inoculated onto the different pulses. Observations for infection were then recorded to know the cross inoculable nature of the pathogen.

Different pulses used for host range and cross inoculation studies

Sl. No.	Common name	Botanical name
1.	Blackgram	*Vigna mungo*
2.	Soybean	*Glycine max*
3.	Horsegram	*Macrotyloma uniflorum*
4.	Cowpea	*Vigna unguiculata*
5.	Redgram	*Cajanus cajan*
6.	Bengalgram	*Cicer aerietinum*
7.	Frenchbean	*Phaseolus vulgaris*
8.	Clusterbean	*Cyamopsis tetragonoloba*

3.7 Screening of genotypes for anthracnose resistance and biochemical factors of resistance

3.7.1 Screening of greengram genotypes under artificial conditions against *C. truncatum*

Thirty greengram genotypes obtained from All India Co-ordinated Research Project on MULLARP, Dharwad were screened against *Colletotrichum truncatum* under greenhouse conditions.

To carryout the screening work, 60 earthen pots were taken with sterilized soil and kept in greenhouse. Twenty seeds of each cultivar were sown in each pot and two such pots were maintained for each genotype. Later, five seedlings per pot were maintained for screening purpose. Conidial suspension of *C. truncatum* (10^5 conidia/ml) was prepared from ten days old culture grown on potato dextrose broth medium and seedlings were inoculated with the spore suspension after 20 days of sowing. After inoculation, the plants were kept in growth chamber maintained at 25^0C temperature and 90 per cent relative humidity. Observations were recorded on tenth days of inoculation by following 0 to 9 scale given by Mayee and Datar (1986).

Genotypes categorization

Category	Reactions
0	Immune
1	Resistant
3	Moderately resistant
5	Moderately susceptible
7	Susceptible
9	Highly susceptible

3.7.2 Screening of promising greengram varieties under field conditions

A field experiment was conducted to know the resistance levels in the promising varieties which were developed at Agriculture Research Station, Bidar during *kharif* 2006-07 and 2007-08. A total of six promising varieties which were agronomically superior were evaluated. The experiment was conducted with randomized block design with four replications. The severity of anthracnose was recorded using a disease rating scale 0 to 9 given by Mayee and Datar (1986) as described earlier.

3.7.3 Biochemical studies

Effect of anthracnose of greengram on biochemical constituents *viz.,* chlorophyll, sugars and phenols was estimated both in healthy and diseased leaves. For this, two resistant (TM-96-2 and TARM-18), two moderately resistant (BGS-9 and TM-97-55) and three

susceptible genotypes (Sel-4, Chinamung and Yellow mung) were selected and grown in glasshouse condition. At 30 DAS, the plants were inoculated with spore suspension of *C. truncatum* at concentration of 10^5 conidia per ml. The plants were tested for the above said biochemical constituents after ten days of inoculation (at 40 DAS). Similarly, healthy leaves were also used for estimation.

3.7.3.1 Chlorophyll content

Total chlorophyll, chlorophyll 'a' and chlorophyll 'b' content were determined (Arnon, 1949). Leaf tissue from the middle portion of the plant was cut into small pieces. One hundred mg of sample was homogenized with 80 per cent acetone. The final volume of the extract was made to 25 ml. The absorbance of the extract was measured at 645, 652 and 663 nm in spectrophotometer. The total chlorophyll, chlorophyll 'a' and chlorophyll 'b' contents were calculated using the following formula.

$$\text{Total Chlorophyll} = 20.2\,(A_{645}) + 8.02\,(A_{663}) \times \frac{V}{1000 \times W \times a}\ (\text{mg g}^{-1}\ \text{fr.wt.})$$

$$\text{Chlorophyll (a)} = 12.7\,(A_{663}) - 2.69\,(A_{645}) \times \frac{V}{1000 \times W \times a}\ (\text{mg g}^{-1}\ \text{fr.wt.})$$

$$\text{Chlorophyll (b)} = 22.9\,(A_{645}) - 14.65\,(A_{652}) \times \frac{V}{1000 \times W \times a}\ (\text{mg g}^{-1}\ \text{fr.wt.})$$

Where,

V : Volume of the extract (25 ml)
W : Fresh weight of the sample (100 mg)
a : Path length of light (1 cm)

3.7.3.2 Estimation of sugars and phenols

Five grams of leaf material was extracted in ethanol as per the procedure followed by Jaypal and Mahadevan (1968) and clarified with saturated solution of lead acetate. The excess lead aceate was precipitated by the addition of sufficient quantity of standard solution of di-sodium hydrogen orthophosphate.

The precipitate was removed by filtering the alcohol extract through Whatman No. 1 filter paper and the filtrate was made up to 25 ml volume with 80 per cent alcohol. Reducing sugars, non-reducing sugars, total sugars, phenols and ortho dihydroxy phenols were estimated in alcohol extract of fresh leaves.

3.7.3.3 Sugars

Reducing sugars for leaf samples were estimated by Nelson's modification of Somogyi's method (Nelson, 1944). Non-reducing sugars were hydrolyzed by using 1 ml of 1 N H_2SO_4 and then estimated as in case of reducing sugars to get the total sugars. Non-reducing sugars were calculated by subtracting the reducing sugar from that of total sugars.

3.7.3.4 Total phenols

Estimation of total phenols present in plant samples was carried out following Folin-Ciocalteau reagent method. One ml of the alcohol extract was taken in a test tube to which 1.0 ml of Folin-Ciocalteau reagent was added followed by 2.0 ml of sodium carbonate solution. The tubes were shaken well and heated in boiling hot water bath for exactly one minute and then cooled under running tap water. The blue solution was diluted to 25 ml with water and its absorbance was read at 650 nm in spectrophotometer. The amount of phenols present in the sample was calculated from a standard curve prepared from catechol.

3.7.3.5 Ortho dihydroxy phenols

Arnow's reagent specially reacts with ortho dihydroxy phenols by producing a pink coloured complex, the intensity of which is measured in a colorimeter. One ml of the alcohol

extract was pipetted into a test tube to which 1.0 ml of 0.05 N HCl, 1.0 ml of Arnow's reagent, 10.0 g sodium nitrate ($NaNO_2$) and 10 g of sodium molybdate (Na_2MoO_4) were dissolved in 100 ml distilled water, 10.0 ml of distilled water and 2.0 ml of 1 N NaOH were added.

Soon after the addition of NaOH the contents of test tube turned to pink colour. The intensity was read at 515 nm in spectrophotometer. The ortho dihydroxy phenol content in the unknown samples was determined from the standard curve of catechol.

3.8 Disease management

3.8.1 *In vitro* evaluation of fungicides against *Colletotrichum truncatum*

Eight systemic and five non-systemic fungicides were tested against *C. truncatum* on the potato dextrose agar media using poison food technique under *in vitro* condition. The systemic fungicides were tried at 0.05, 0.1 and 0.15 per cent concentrations, whereas non-systemic fungicides were evaluated at 0.1, 0.2 and 0.25 per cent concentrations. The list of fungicides used along with their chemical and trade names are given below.

Systemic fungicides

Sl. No.	Common Name	Chemical Name	Trade Name
1.	Carbendazim	Methyl 2 Benzimidazole carbomate	Bavistin 50 WP
2.	Propiconazole	1-(2-(2, 4-D)-4-Propyl-1,3 diozolan- 2yl methyl) IH-1, 2, 4 Triazole	Tilt 25% EC
3.	Hexaconazole	RS-2-(2, 4-D)-1-(1H-1, 2, 4 Trizole-1-yl) hezan 2-ol	Contaf 5% EC
4.	Tricyclazole	5-methyl-1, 2, 4-triazole (3, 4b) benzothiazole	Beam 75% WP
5.	Benomyl	Methyl-N (1 Butyl carbonyl)2- Benzimidazole carbonate	Benlate 50 WP
6.	Thiophanate methyl	1, 2, bis (3-metoxy carboxyl-1-2-thiouredo) benzene	Roko 70 WP
7.	Difenconazole	Cis, trans-3-chloro-4(4-methyl-2-(1H-1, 2, 4-Traizole-1-y1, methyl)-1, 3-dioxolan-2-y1) phenyl 4-chlorophenyl ether	Score 25 EC
8.	Penconazole	1-[2-(2,4-dichlorophenyl) pentyl]-1H-1, 2, 4-triazole	Topaz 10 EC

Non-systemic fungicides

Sl. No.	Common Name	Chemical Name	Trade Name
1.	Mancozeb	Manganese ethylene bis dithiocarbonate plus zinc	Indofil M-45 75 WP
2.	Propineb	Zinc propylenebis dithiocartamate	Antracol 75 WP
3.	Copper oxychloride	Copper oxychloride	Fytolon 50 WP
4.	Chlorothalonil	Tetrachloro isopthalo nitrate	Kavach 75 WP
5.	Carbendazim 12% + Mancozeb 63%	Methyl 1H-benzimidazole-2yl-carbomate + manganesethyl lene bis – dithiocarbmate plus zinc	Saaf 75 WP

Poison food technique

The poison food technique (Shravelle, 1961) was followed to evaluate the efficacy of fungicides in inhibiting the mycelial growth of *C. truncatum*. The fungus was grown on PDA medium for eight days prior to setting up the experiment. The PDA medium was prepared and

melted. The fungicidal suspension was added to the melted media to obtain the required concentrations. About 20 ml of poisoned medium was poured in each sterilized petriplates. Suitable check was maintained without addition of fungicides. Five mm mycelial disc was taken from the periphery of eight days old colony was placed in the centre of petriplate and incubated at 28 $\pm$ 1^{0}C for 15 days. Three replications were maintained for each treatment. The diameter of the colony was measured when maximum growth in control plates were occurred. The per cent inhibition was calculated by using the formula of Vincent (1947).

$$I = \frac{(C - T)}{C} \times 100$$

Where,

I : Per cent inhibition
C : Mycelial growth in control
T : Mycelial growth in treatment

3.8.2 *In vitro* evaluation of botanicals

The present investigation was carried out to evaluate the extracts of different plant species to know the possible presence of fungi toxicant properties against *C. truncatum*.

Preparation of plant extracts

Fifty grams of fresh healthy plant parts (leaves/root/bulbs) collected from field were washed with distilled water and air-dried and crushed in 50 ml of sterile water. The crushed product was filtered through muslin cloth and collected the filtrate. The prepared solution gave 100 per cent, which was further diluted to required concentrations of 5.0, 7.5 and 10.0 per cent. The extracts were tested against *C. truncatum* on the cultural media using poison food technique under *in vitro* condition. Details about the botanicals and part used are given below.

Botanicals

Sl. No.	Plant (common name)	Scientific name	Plant part used
1.	Neem	*Azadirachta indica*	Kernel
2.	Eucalyptus	*Eucalyptus citridora*	Oil
3.	Cynodon	*Cynodon dactylon*	Plant
4.	Bellary jali	*Prosophis juliflora*	Leaf
5.	Parthenium	*Parthenium historophorus*	Plant
6.	Garlic	*Allium sativum*	Bulb
7.	Neem	*Azadirachta indica*	Readymade herbal product
8.	Onion	*Allium cepa*	Bulb
9.	Turmeric	*Curcuma longa*	Rhizome
10.	Ginger	*Zingiber officinale*	Rhizome

3.8.3 *In vitro* evaluation of bioagents

Antagonistic microorganisms like *Bacillus subtilis*, *Pseudomonas fluorescens*, *Trichoderma harzianum*, *T. viride*, *T. koningii* and *Gliocladium virens* were evaluated for their antagonistic properties against *C. truncatum* by dual culture technique.

Dual culture test

Bioagents were evaluated for their efficacy through dual culture technique. The bioagents and the test fungus were inoculated side by side on a single petridish containing solidified PDA medium. Three replications were maintained for each treatment with one control by maintaining only pathogen and bioagent separately. Inoculated plates were incubated at 27 $\pm$ 1^{0}C for eight days. The diameter of the colony of both bioagents and the pathogen was measured in two directions and average was recorded. Per cent inhibition of growth of the test fungus was calculated by using the formula of Vincent (1947).

3.8.4 *In vitro* evaluation of ITK's

Five ITK's *viz.,* cow urine, cow milk, vermiwash, panchagavya and butter milk were evaluated *in vitro* against spore germination by cavity slide method. Different ITKs were tried at 1:2 (50%); 1:5 (20%); 1:10 (10%); 1:15 (6.67%) and 1:20 (5%) concentrations. Required concentrations of each product were prepared in distilled water and four replications were maintained for each concentration in the treatments. One hundred spores were observed 12 hours after incubation in moist chamber. A control treatment was maintained with distilled water. Per cent inhibition of spore germination over control was calculated using the formula given by Vincent (1947).

$$\text{Per cent inhibition} = \frac{C - T}{C} \times 100$$

Where,

C : Per cent germination of spore in control

T : Per cent germination of spore in treatment

3.8.5 Integrated management

The field trials were conducted for two consecutive years during *kharif* 2006 and 2007 at Agricultural Research Station, Bidar. The experiments were laid out in Randomized Block Design with two replications and seventeen treatments including chemicals, bioagent, botanicals and untreated control. The details of treatment combinations are given hereunder.

Experimental details

Variety : Chinamung; Plot size : 3.0 × 5.0 m

T_1	: *Trichoderma harzianum* at 4 g/kg of seed (SD)
T_2	: Carbendazium 50 WP at 2 g/kg of seed (SD)
T_3	: Benomyl 50 WP at 2 g/kg of seed (SD)
T_4	: Thiophanate methyl 70 WP at 2 g/kg of seed (SD)
T_5	: Carbendazim 12% + mancozeb 63% at 2 g/kg of seed (SD)
T_6	: Tricyclozole 75 WP at 2 g/kg of seed (SD)
T_7	: T_1 + One spray of Eucalyptus oil at 10%
T_8	: T_1 + One spray of cow urine at 10 %
T_9	: T_1 + One spray of Azadirachtin (1500 ppm) at 0.2%
T_{10}	: T_2 + One spray of carbendazim 50 WP at 0.1%
T_{11}	: T_3 + One spray of Benomyl 50 WP at 0.1%
T_{12}	: T_4 + One spray of Thiophanate methyl 70 WP at 0.1%
T_{13}	: T_5 + One spray of carbendazim 12% + Mancozeb 63% at 0.2%
T_{14}	: T_6 + One spray of Tricyclozole 75 WP at 0.1%
T_{15}	: One spray of Hexaconazole 5% EC at 0.1%
T_{16}	: One spray of Propiconazole 25% EC at 0.1%
T_{17}	: Control

The spraying of fungicides were undertaken immediately after the appearance of the disease.

Ten plants in each plot were scored for disease severity and data were converted into per cent disease index (PDI) as explained earlier. Each treatment was harvested separately and yield per plot was recorded further benefit:cost ratio was calculated.

3.9 Statistical analysis and interpretation

The data obtained from the laboratory and field experiments were statistically analyzed by following the standard procedures (Panse and Sukhatme, 1967). The percentage values were converted to arcsine values wherever required.

4. EXPERIMENTAL RESULTS

Among the different foliar diseases of greengram, anthracnose caused by *Colletotrichum truncatum* (Schw.) Andrus and Moore is one of the major diseases, causing an yield loss upto 24 to 67 per cent. Hence, the present investigations on epidemiology and integrated management of anthracnose of greengram were carried out in the laboratory as well as in the field during *kharif* 2006-07 and 2007-08, both at Agricultural Research Station, Bidar and Department of Plant Pathology, College of Agriculture, UAS, Dharwad. The results thus obtained are presented hereunder.

4.1 Symptomatology

Under natural conditions, spots appeared first on the lower surface of leaf later on they appeared on leaf petiole, stem and also on pod. The spots were brown with reddish centers on lower surface. At later stage, they appeared on upper surface with reddish brown ring like spot of 8 – 10 mm diameter and subsequently they turned to dark brown chlorotic spots later turned to 'shot holes'. On lower surface, the symptom looked large (10 – 12 mm diameter) patches of bright blood red stains. Similarly, on petiole and stem, reddish brown coloured streaks appeared. The same symptoms appeared on pods also with discoloured and small sized production of seeds. In severe cases, the whole leaf was covered with brown patches. The spots coalesced to form large sized patches and in severe cases premature defoliation occurred (Plate 2).

4.2 Morphological studies

The colonies of *Colletotrichum truncatum* were dark brown to black with septate, branched, brown and 3 to 4 μ thick mycelium. The acervuli were black in colour, oval to conical in shape and measuring 171.5 $\times$ 248.0 μ. The acervuli were embedded with very light pinkish coloured mucilaginous mass containing numerous conidia. Setae were longer than conidiophore, erect, hairlike, broader at base and tapering at apex, black coloured and arising through the mucilaginous mass of conidia and were measuring 78.0 to 201.0 $\times$ 5.0 – 7.1 μ. Conidia were single celled, smooth walled, hyaline, curved and measured 20.0 to 23.3 $\times$ 3.5 to 4.0 μ in size, and germinate by germtubes. The appressoria were sparse, pale to dark brown, clavate or circular in outline and 8 $\times$ 6.7 μ in size (Plate 3).

4.3 Survey for the severity of disease

A roving survey was conducted during *kharif* 2006 and 2007 in eleven major greengram growing districts of Northern Karnataka *viz.*, Bagalkot, Belgaum, Bellary, Bidar, Bijapur, Dharwad, Gadag, Gulbarga, Haveri, Koppal and Raichur. The village-wise disease severity has been presented in Table 1 and 2 for *kharif* 2006 and *kharif* 2007, respectively.

Data pertaining to survey conducted during *kharif* 2006 as presented in Table 1 revealed that, anthracnose of greengram was noticed with a range of 21.36 to 58.97 per cent. The anthracnose severity in Bagalkot district ranged from 25.15 per cent (Ballur) to 37.25 per cent (Bevoor). The disease index ranged between 27.22 (Inchal) to 35.20 per cent (Belavadi) in Belgaum district. In Bellary district, the disease ranged between 20.15 per cent (Sonna) to 33.16 per cent (Itagi). In Bidar, the disease index varied from 35.62 (Kamthan) to 60.54 per cent (Dubalgundi). In Bijapur district, the disease index ranged between 19.71 per cent (Halsangi) to 25.36 per cent (Almel). The disease index ranged between 27.81 (Gambyapur) to 50.17 per cent (Hirenarti) in Dharwad district. In Gadag district, the disease index ranged between 35.16 per cent (Advisomapur) to 52.64 per cent (Kalkeri). In Gulbarga, the disease index varied from 35.29 (Tengali) to 60.19 per cent (Pechanpalli). In Haveri district, the disease index ranged between 32.18 per cent (Basapur) to 41.08 per cent (Antarvalli). In Koppal, the disease index varied from 27.33 (Chikkenkoppa) to 38.11 per cent (Talkal). The disease index ranged between 26.75 (Guragunta) to 35.22 per cent (Heerapur) in Raichur district.

During *kharif* 2007, the disease severity was noticed with a range of 24.67 to 60.07 per cent (Table 2). In Bagalkot district, the anthracnose severity ranged from 28.52 per cent (Sunag) to 41.27 per cent (Bevoor). In Belgaum district, the disease index was in the range of 28.66 per cent (Inachal) to 40.16 per cent (Nesargi). The disease index in Bellary was in the

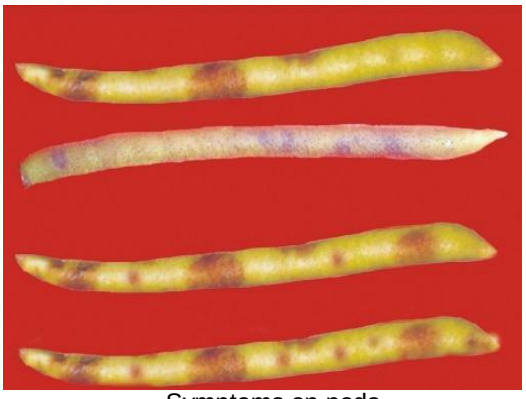
Symptoms on upper surface

Symptoms on lower surface

Shot hole Symptoms

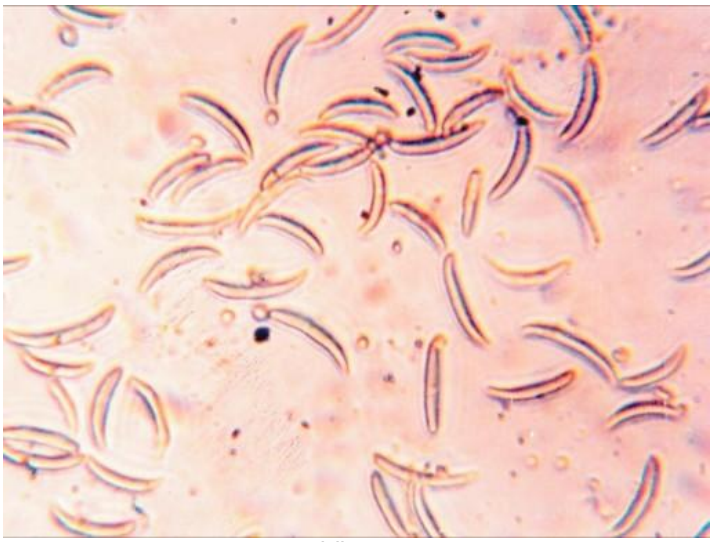
Symptoms on branches

Symptoms on stem

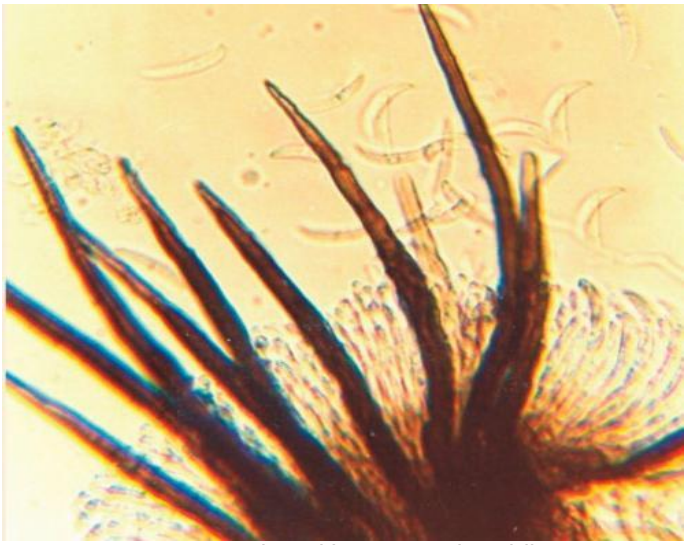


Symptoms on pods

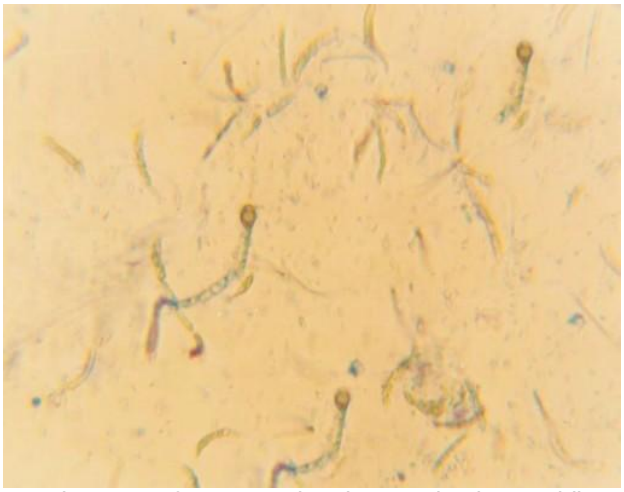
Plate 2: Symptoms of anthracnose of greengram caused by colletotrichum truncatum



Conidia



Acervulus with setae and conidia



Development of appressorium by germinating conidia

Plate 3: Microphotograph showing morphological characters of C.truncatum

Table 1: Survey for the severity of anthracnose of greengram caused by *Colletotrichum truncatum* during *kharif* 2006 in different villages of northern Karnataka

Sl No.	District	Taluk	Village	No. of fields	Stage of the crop (DAS)	Percent disease index (PDI)
1.	Bagalkot	Bagalkot	Anagawadi	5	35	28.56
			Bevoor	6	45	37.25
			Halloor	5	35	30.16
			Kamatagi	7	35	29.22
			Sannapur	5	35	27.75
		Mean				30.59
		Bilagi	Amalzari	5	35	27.55
			Ballur	5	35	25.15
			Sunag	6	40	27.70
			Tumarmatti	5	38	26.15
			Yadahalli	6	40	28.25
		Mean				26.96
		Hungund	Amingad	6	40	28.75
			Ilakal	5	42	29.78
			Kandagallu	5	35	27.63
			Karadi	6	40	28.21
			Kesarwadi	5	32	25.55
		Mean				27.98
2.	Belgaum	Bailhongal	Belavadi	6	45	35.20
			Budarakatti	5	40	30.16
			Inchal	5	40	27.22
			Kenganur	5	41	32.15
			Nayanagar	5	45	34.60
		Mean				31.87
3.	Bellary	Hadagali	Hagarnoor	5	32	28.75
			Hirehadagali	6	35	30.25
			Holgundi	5	35	31.40
			Itagi	5	45	33.16
			Uattangi	6	30	25.81
		Mean				29.87
		Hagaribommanalli	Maratagi	6	35	21.85
			Morigeri	5	40	27.22
			Ramnagar	5	42	30.02
			Shivanandnagar	5	35	22.65
			Sonna	6	30	20.15
		Mean				24.38

Contd……

Sl. No.	District	Taluk	Village	No. of fields	Stage of the crop (DAS)	Percent disease index (PDI)
4	Bidar	Aurad	Kandagul	5	45	41.55
			Khanapur	5	40	40.61
			Kouta(B)	6	45	49.28
			Kouta (K)	5	45	45.16
			Santapur	6	48	50.23
		Mean				45.37
		Basavakalyan	Dhanuur	5	50	50.12
			Kherda	5	50	60.11
			Rajeshwar	6	45	48.31
			Rajola	6	45	45.21
			Yarabag	5	50	49.75
		Mean				50.70
		Bhalki	Byalhalli	5	50	49.98
			Chalkapur	6	45	43.11
			Halbarga	6	45	47.52
			Mehekar	5	45	44.67
			Nittur	5	48	45.29
		Mean				46.11
		Bidar	Chikpeth	6	45	41.34
			Janawad	5	40	39.75
			Kamthan	5	40	35.62
			Markhal	5	40	45.23
			Noubad	5	45	41.08
		Mean				40.60
		Humnabad	Alur	6	50	56.71
			Dhummansur	5	50	59.72
			Dubalgundi	5	52	60.54
			Hallikhed (B)	5	50	60.21
			Nimbur	6	45	57.65
		Mean				58.97
5	Bijapur	Indi	Chadchan	5	35	20.15
			Halsangi	6	35	19.71
			Rodagi	5	40	22.10
			Salotagi	5	35	23.78
			Tamba	5	35	21.06
		Mean				21.36
		Sindagi	Almel	6	45	25.36
			Koralli	5	35	21.41
			Malaghan	5	35	19.75
			Shivanagi	5	30	23.18
			Somajal	6	40	24.53
		Mean				22.85

Contd……

Sl. No.	District	Taluk	Village	No. of fields	Stage of the crop (DAS)	Percent disease index (PDI)
6.	Dharwad	Dharwad	Garag	6	45	41.30
			Lokur	6	50	48.21
			Narendra	5	50	46.53
			Neeralkatti	5	55	49.62
			Tadkod	5	45	39.28
		Mean				44.98
		Hubli	Byahatti	5	35	38.61
			Ingalahalli	6	45	46.33
			Manakod	5	45	40.15
			Sattur	5	48	39.75
			Unakal	6	35	35.82
		Mean				40.13
		Khalaghatgi	Bgudihal	5	35	30.28
			Dumwad	6	40	34.26
			Emmatti	5	30	29.75
			Gambyapur	5	30	27.81
			Jammihal	6	35	38.15
		Mean				32.05
		Kundagol	Gudageri	5	45	50.15
			Hirenarti	6	50	50.17
			Kalasa	5	50	48.35
			Ramankoppa	6	45	43.28
			Saunshi	5	45	48.70
		Mean				48.13
7	Gadag	Gadag	Advisomapur	6	40	35.16
			Beladadi	6	45	41.28
			Hirehandigol	5	35	39.11
			Hulkoti	5	40	37.39
			Papanashi	5	45	42.70
		Mean				39.13
		Naragund	Biranatti	5	45	45.16
			Chikknaragund	6	50	50.10
			Kalkeri	5	52	52.64
			Konnur	5	45	49.81
			Yaragal	6	40	48.78
		Mean				49.30
		Ron	Abbigeri	6	40	38.75
			Chikkamannur	5	45	41.65
			Doudi	5	50	45.18
			Mallapur	6	50	43.51
			Nidagundi	5	45	39.80
		Mean				41.78

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Sl. No.	District	Taluk	Village	No. of fields	Stage of the crop (DAS)	Percent disease index (PDI)
8	Gulbarga	Chincholli	Kodli	6	48	50.15
			Nidagunda	5	50	57.41
			Pechanpalli	5	55	60.19
			Ratakal	5	45	49.28
			Sulepeth	6	50	54.33
		Mean				54.27
		Chittapur	Hirur	5	35	38.81
			Kandagul	5	40	42.38
			Sugareddy	5	45	45.15
			Tengali	6	35	35.29
			Vatavatti	6	50	49.11
		Mean				42.15
		Gulbarga	Firozabad	5	52	40.81
			Mahagaon	5	40	36.89
			Partabad	5	45	41.28
			Pattan	6	50	46.36
			Sannu	5	45	38.75
		Mean				40.82
		Sedam	Gounalli	5	55	59.42
			Handaraki	6	45	42.38
			Kodla	5	48	45.60
			Namar	5	52	56.23
			Surawar	6	50	48.91
		Mean				50.51
9	Haveri	Byadagi	Budapanhalli	6	45	38.51
			Chikkhanji	5	40	37.90
			Hirehanji	5	35	36.63
			Kagenhalli	5	48	39.18
			Motebennur	6	45	40.34
		Mean				38.51
		Haveri	Basapur	6	35	32.18
			Guttal	5	45	40.75
			Hosaritti	5	45	39.63
			Kurugunda	5	40	35.24
			Neglur	6	40	38.06
		Mean				37.17
		Ranebennur	Antarvallli	5	50	41.08
			Hullatti	5	45	39.75
			Itagi	6	42	42.33
			Kamadod	6	45	38.65
			Khunbevu	5	45	40.91
		Mean				40.54

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Sl. No.	District	Taluk	Village	No. of fields	Stage of the crop (DAS)	Percent disease index (PDI)
10	Koppal	Kushtagi	Aralhalli	6	40	33.10
			Gotagi	5	40	35.21
			Hanumsagar	6	35	28.56
			Kyadaguppi	5	35	30.16
			Yellogal	5	40	30.05
		Mean				31.42
		Yalburga	Binnal	6	35	31.50
			Chikkenkoppa	5	40	27.33
			Karmudi	5	40	35.42
			Talkal	6	40	38.11
			Yerihanchinal	5	38	29.28
		Mean				32.33
11.	Raichur	Lingasugur	Guragunta	5	40	26.75
			Madkihal	5	45	30.23
			Maski	6	50	32.50
			Mudgal	6	40	29.78
			Santekallur	5	40	31.56
		Mean				30.16
		Raichur	Ashapur	5	40	32.36
			Chandrabanda	5	35	29.62
			Heerapur	6	45	35.22
			Kudlur	5	38	27.40
			Yaragera	5	40	30.78
		Mean				31.08

range of 22.51 per cent (Hampasagar) to 35.21 per cent (Masalwad). In Bidar, the disease index varied from 40.31 per cent (Janawad) to 61.80 per cent (Chitaguppa). In Bijapur district, the disease index ranged between 20.19 per cent (Golasar) to 30.06 per cent (Atharga). The disease index ranged between 30.56 per cent (Kannyanayakanakoppa) to 52.41 per cent (Hirenarti) in Dharwad district. In Gadag district, the disease index ranged between 38.43 per cent (Hulkoti) to 54.17 per cent (Chikkanaragund). In Gulbarga, the disease index varied from 38.91 per cent (Kerur) to 60.10 per cent (Nidagunda). In Haveri district, the disease index ranged between 36.75 per cent (Hosalli) to 46.19 per cent (Kodihalli), while in Koppal district the range was 29.27 per cent (Hanumasagar) to 40.10 per cent (Talkal). Similarly, in Raichur district, the disease index was in the range of 29.78 per cent (Gudanal) to 38.61 per cent (Matmari).

The consolidated district-wise observation on severity is given in Table 3 and depicted in Fig. 1a and 1b. The data indicated that, the disease appeared to be in severe form during both the years (*kharif* 2006 and 2007). But, the severity during *kharif* 2007 was more (38.34%) than *kharif* 2006 (35.53%).

During *kharif* 2006, highest disease severity was observed in Bidar district (48.35%) followed by Gulbarga (46.94%), Gadag (43.40%), Dharwad (41.32%), Haveri (38.74%), Koppal (31.88%), Belgaum (31.87%), Raichur (30.62%), Bagalkot (28.51%), Bellary (27.13%) and Bijapur (22.11%). Among the taluks, Humnabad of Bidar district recorded maximum severity (58.97%) followed by Chincholli (54.27%) of Gulbarga district. All other talukas recorded less than 51 per cent. Indi taluk of Bijapur district recorded the least severity (21.36%).

Table 2: Survey for the severity of anthracnose of greengram caused by *Colletotrichum truncatum* during *kharif* 2007 in different villages of northern Karnataka

Sl No.	District	Taluk	Village	No. of field	Stage of the crop (DAS)	Percent disease index
1.	Bagalkot	Bagalkot	Anagawadi	5	35	30.15
			Bevoor	5	45	41.27
			Dodwad	6	45	40.35
			Kamatagi	5	35	31.23
			Lokapur	5	40	38.55
			Mannakatti	6	45	35.23
		Mean				36.13
		Bilagi	Amalzari	5	35	31.06
			Ballur	5	40	30.15
			Sunag	5	40	28.52
			Tumarmatti	6	45	30.23
			Yadahalli	5	35	29.75
		Mean				29.94
		Hungund	Amadihal	6	40	31.78
			Amingad	5	42	31.20
			Ilakal	5	35	32.11
			Kandagallu	5	40	29.23
			Karadi	5	32	30.62
			Nandawadgi	6	35	30.15
		Mean				30.85
2	Belgaum	Bailhongal	Belavadi	5	45	38.20
			Budarkatti	5	40	32.75
			Inchal	5	40	28.66
			Kenganur	5	41	37.25
			Nayanagar	6	45	39.10
			Nesargi	5	45	40.16
		Mean				36.02
3.	Bellary	Hadagali	Hirehadgali	6	32	31.50
			Hirekolati	5	35	30.65
			Holgundi	5	35	33.25
			Itagi	5	45	30.23
			Masalwad	6	30	35.21
			Uttangi	5	32	29.45
		Mean				31.72

Contd……

Sl. No.	District	Taluk	Village	No. of field	Stage of the crop (DAS)	Percent disease index
		Hagaribommanalli	Gaddikeri	5	35	32.50
			Hampasagar	5	40	22.51
			Maratagi	6	42	23.65
			Morigeri	5	35	26.05
			Ramnagar	6	30	28.75
			Shivanand nagar	5		24.15
		Mean				26.27
4.	Bidar	Aurad	Ganeshpur	6	45	52.72
			Kamalnagar	5	40	48.51
			Kandagul	7	45	45.06
			Khanapur	6	45	47.15
			Kouta (K)	7	48	47.21
			Thanakushnoor	5		43.63
		Mean				47.38
		Basavakalyan	Dhannur	6	50	58.18
			Islampur	6	50	42.33
			Kherda	7	45	55.37
			Pandergera	5	45	49.76
			Rejeshwar	6	50	56.25
			Sadlapur	5		48.16
		Mean				51.68
		Bhalki	Byalhalli	5	50	50.15
			Halabarga	5	45	46.05
			Khatakchincholli	6	45	49.11
			Korur	7	45	43.91
			Mehakar	6	48	48.45
			Nidaban	6		48.50
		Mean				47.70
		Bidar	Basantpur	7	45	41.83
			Chickpeth	6	40	42.21
			Godmpalli	5	40	49.15
			Hokran	6	40	50.46
			Janawad	7	45	40.31
			Markhal	7	45	47.50
			Sultanpur	6		48.75
		Mean				45.74
		Humnabad	Alur	5	50	59.21
			Chitaguppa	6	50	61.80
			Dhummansur	5	52	60.05
			Dubalgundi	5	50	61.11
			Kabirabadwadi	5	45	59.30
			Kallur	6	50	58.75
			Kodambal	5		60.27
		Mean				60.07

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Sl. No.	District	Taluk	Village	No. of field	Stage of the crop (DAS)	Percent disease index
5.	Bijapur	Indi	Atharga	5	35	30.06
			Benakanhalli	5	35	23.28
			Chadachan	6	40	21.23
			Golasar	6	35	20.19
			Roogi	5	35	26.73
			Salotagi	6	35	25.11
			Tadawalaga	5	35	28.18
			Tamba	5	35	22.59
		Mean				24.67
		Sindagi	Almel	6	45	27.31
			Devaranavadgi	5	35	30.05
			Golageri	5	35	26.42
			Kannolli	5	30	29.10
			Koralli	5	40	23.15
			Rampur	6	35	28.38
			Somajal	6	38	25.27
			Yenkanchi	6	35	22.56
		Mean				26.53
6	Dharwad	Dharwad	Garag	5	45	42.17
			Kardigudda	5	50	40.28
			Lokur	5	50	50.17
			Mangalgatti	6	55	48.05
			Narendra	6	45	48.85
			Tadkod	5	52	45.51
		Mean				45.84
		Hubli	Agadi	5	35	45.83
			Byahatti	6	45	40.15
			Ingalhalli	5	45	48.21
			Manakod	6	48	41.33
			Palikoppa	5	35	39.75
			Sattur	5	40	40.10
		Mean				42.56
		Kalghatgi	Bgudihal	5	35	40.70
			Dumwad	5	40	35.62
			Emmatti	6	30	39.25
			Gambyapur	5	30	36.83
			Jammihal	5	35	38.11
			Kannyanayakankoppa	5	40	30.56
		Mean				36.85

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Sl. No.	District	Taluk	Village	No. of field	Stage of the crop (DAS)	Percent disease index
		Kundagol	Hirenarti	5	45	52.41
			Hosanagar	6	50	51.22
			Jigalur	5	50	48.33
			Kalsa	5	45	49.78
			Kamadolli	5	45	50.10
			Ramanakoppa	5	50	48.65
		Mean				50.08
7.	Gadag	Gadag	Beladadi	5	40	42.16
			Hirehandigol	5	45	40.18
			Hulkoti	6	35	38.43
			Kurkoti	5	40	44.52
			Mulgund	5	45	43.20
			Papnashi	5	45	44.23
		Mean				42.12
		Naragund	Biranatti	5	45	48.11
			Chikkanargund	5	50	54.17
			Kalken	6	52	53.34
			Konnur	5	45	49.76
			Yaragal	5	40	52.08
		Mean				51.49
		Ron	Abbigeri	5	40	40.51
			Chikkamannur	5	45	45.10
			Doudi	5	50	48.05
			Mallapur	6	50	44.23
			Nidagundi	5	45	40.75
		Mean				43.73
8	Gulbarga	Chincholli	Inapur	5	48	51.16
			Kodli	6	50	57.28
			Nidagunda	6	55	60.10
			Pechanpalli	5	45	59.42
			Ratakal	6	50	54.51
			Sulepeth	6	50	58.33
		Mean				56.80
		Chittapur	Bimanhalli	5	35	49.23
			Kandagul	6	40	45.70
			Margol	5	45	44.61
			Ramtirth	6	35	40.32
			Satanur	5	50	39.75
			Vatavatti	5	52	50.15
		Mean				44.96

Contd……..

Sl. No.	District	Taluk	Village	No. of field	Stage of the crop (DAS)	Percent disease index
		Gulbarga	Firozabad	5	52	41.75
			Kadbur	6	40	45.32
			Kerur	5	45	38.91
			Partabad	6	50	46.15
			Pattan	5	45	47.50
			Sannur	5	40	39.81
		Mean				43.24
		Sedam	Handaraki	5	55	48.78
			Kodla	6	45	54.35
			Madana	5	48	44.52
			Namar	5	52	59.15
			Satapatanhalli	5	50	57.21
			Surawar	6	50	49.10
		Mean				52.19
9.	Haveri	Byadagi	Chikkahanji	5	45	39.23
			Hirehanji	6	40	38.75
			Kagenhalli	5	35	37.82
			Kodihalli	5	48	46.19
			Mallur	5	45	40.05
			Motebennur	6	45	42.50
		Mean				40.76
		Haveri	Guttal	6	35	41.08
			Hosalli	5	45	36.75
			Hosaritti	6	45	41.70
			Kabbur	5	40	42.19
			Kurugunda	5	40	37.52
			Negalur	6	45	40.10
		Mean				39.89
		Ranebennur	Antaravalli	5	50	43.27
			Asundi	5	45	41.70
			Hullatti	6	42	40.23
			Itagi	6	45	45.62
			Kamadod	5	45	44.38
			Yallapur	5	40	39.81
		Mean				42.50
10.	Koppal	Kushtagi	Aralhalli	5	40	35.61
			Gotagi	5	40	36.17
			Hanumnal	6	35	30.75
			Hanumasagar	5	35	29.27
			Kyadaguppi	5	40	31.18
			Yellogal	5	40	32.05
		Mean				32.51

Contd…….

Sl. No.	District	Taluk	Village	No. of field	Stage of the crop (DAS)	Percent disease index
		Ylaburga	Bannikoppa	5	35	37.05
			Binnal	5	40	33.28
			Halligudi	5	40	38.46
			Hallikeri	5	40	29.87
			Karmudi	5	38	36.18
			Kuknur	5	40	30.75
			Talkal	6	45	40.10
		Mean				35.10
11.	Raichur	Lingasugur	Gudanal	5	40	29.78
			Hatti	5	45	35.61
			Kota	5	50	33.45
			Maski	6	40	36.70
			Mudagl	5	40	30.86
			Santekallur	5	40	36.10
		Mean				33.75
		Raichur	Ashapur	6	40	35.28
			Chandrabanda	5	35	32.54
			Kapagal	5	45	37.32.
			Kudlur	5	38	36.70
			Matmari	5	40	38.61
			Yargerra	6	35	31.85
		Mean				35.38

During *kharif* 2007, the severity of anthracnose was more compared to *kharif* 2006 in all surveyed talukas. Highest severity was observed in Bidar district (50.51%), followed by Gulbarga (49.30%), Gadag (45.78%), Dharwad (43.83%), Haveri (41.05%), Belgaum (36.02%), Raichur (34.57%), Koppal (33.81%), Bagalkot (32.31%), Bellary (29.00%) and Bijapur (25.60%) districts.

Humnabad taluk of Bidar district recorded highest severity (60.07%) followed by Chincholli (56.80%) and Sedam (52.19%) talukas in Gulbarga district, Basavakalyan (51.68%), taluk of Bidar district and Naragund (51.49%) of Gadag district. All other taluks recorded less than 51 per cent severity. Indi taluka of Bijapur district recorded the least severity (24.67%).

4.4 Crop loss assessment

Field experiments were conducted during *kharif* 2006 and *kharif* 2007 to estimate the losses due to anthracnose in susceptible cultivar Chinamung using carbendazim fungicide. Spray schedule was implemented irrespective of disease appearance from 20 days after sowing (DAS) and imposed at 10 days interval in respective treatments. The observations on anthracnose severity were recorded at weekly interval after onset of disease. The detailed results are presented in Table 4a, 4b, 5, 6, 7, 8, 9, and Fig. 2 and 3.

Table 3: Percent disease index of anthracnose of greengram caused by *Colletotrichum truncatum* during *kharif* 2006 and 2007 in northern Karnataka

District	Percent disease index (PDI)			
	Taluk	2006	2007	Average
Bagalkot	Bagalkot	30.59	36.13	33.36
	Bilagi	26.96	29.94	28.45
	Hungund	27.98	30.85	29.42
Mean		28.51	32.31	30.41
Belgaum	Bailhongal	31.87	36.02	33.95
Mean		31.87	36.02	33.95
Bellary	Hadagali	29.87	31.72	30.80
	Hagaribommnalli	24.38	26.27	25.33
Mean		27.13	29.00	28.07
Bidar	Aurad	45.37	47.38	46.38
	Basavakalyan	50.70	51.68	51.19
	Bhalki	46.11	47.70	46.91
	Bidar	40.60	45.74	43.17
	Humnabad	58.97	60.07	59.52
Mean		48.35	50.51	49.43
Bijapur	Indi	21.36	24.67	23.02
	Sindgi	22.85	26.53	24.69
Mean		22.11	25.60	23.86
Dharwad	Dharwad	44.98	45.84	45.41
	Hubli	40.13	42.56	41.35
	Kalagatagi	32.05	36.85	34.45
	Kundagol	48.13	50.08	49.11
Mean		41.32	43.83	42.58
Gadag	Gadag	39.13	42.12	40.63
	Naragund	49.30	51.49	50.40
	Ron	41.78	43.73	42.76
Mean		43.40	45.78	44.59
Gulbarga	Chincholli	54.27	56.80	55.54
	Chittapur	42.15	44.96	43.56
	Gulbarga	40.82	43.24	42.03
	Sedam	50.51	52.19	51.35
Mean		46.94	49.30	48.12
Haveri	Byadagi	38.51	40.76	39.64
	Haveri	37.17	39.89	38.53
	Ranebennur	40.54	42.50	41.52
Mean		38.74	41.05	39.90
Koppal	Kushtagi	31.42	32.51	31.97
	Yalburga	32.33	35.10	33.72
Mean		31.88	33.81	32.85
Raichur	Lingasugur	30.16	33.75	31.96
	Raichur	31.08	35.38	33.23
Mean		30.62	34.57	32.60
Grand mean		35.53	38.34	36.94

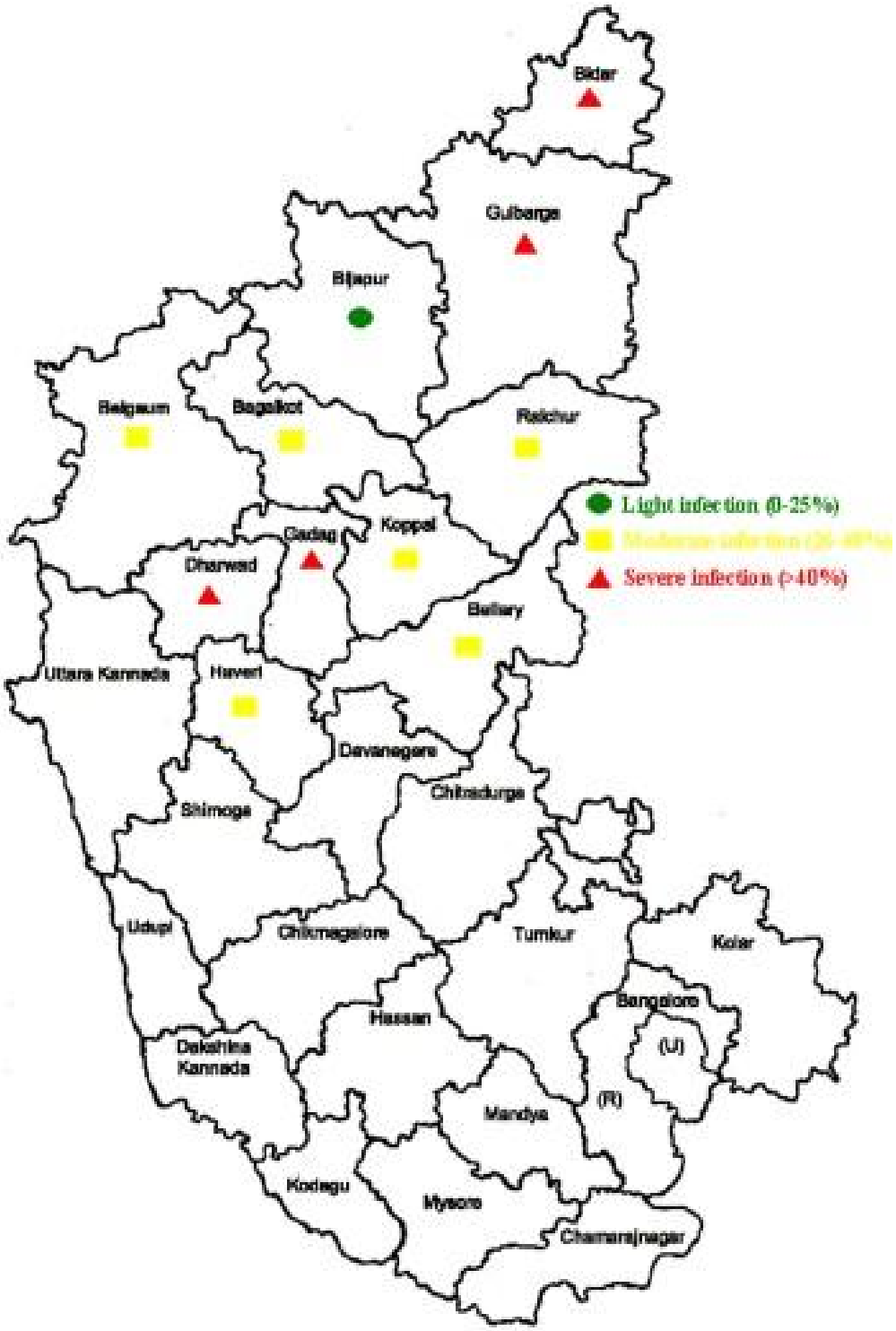


Fig.1a: Severity of anthracnose of greengram caused by Colletotrichum truncatum in different districts Northern Karnataka during Kharif2006

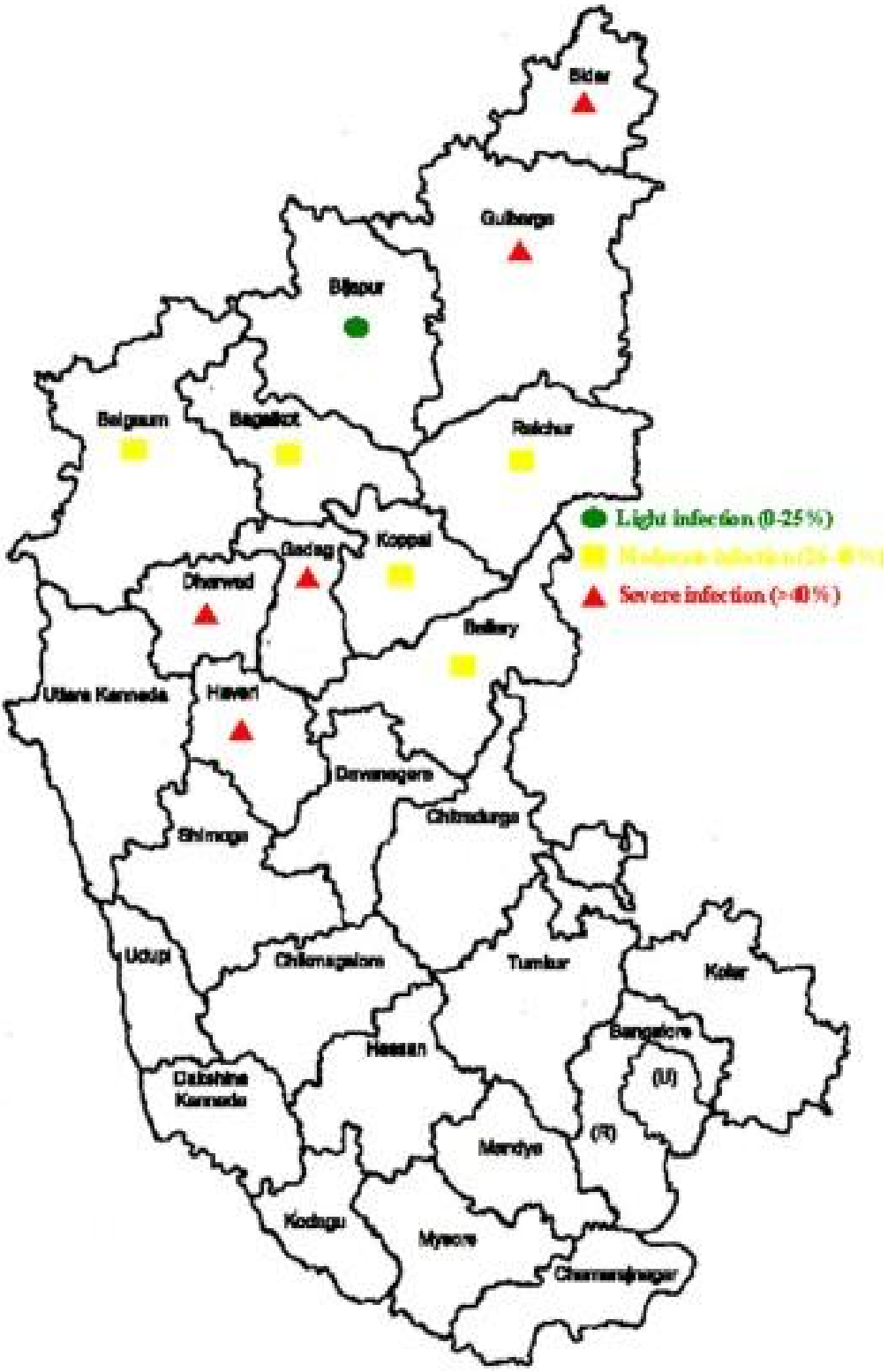


Fig.1bSeverity of anthracnose of greengram caused by Colletotrichum truncatum in different districts of Northern Karnataka during kharif 2007

4.4.1 Per cent disease index (PDI)

Table 4a revealed that at 25 DAS, PDI was significantly reduced in all treatment compared to untreated control. Disease advancement also considerably minimized on greengram crop receiving subsequent fungicide sprays, which recorded 9.90 per cent at 25 DAS and 25.11 per cent at 60 DAS, while disease progression was highest and reached peak value of 58.34 per cent at 60 DAS in unsprayed crop.

Similarly, plots received two sprays recorded PDI of 13.98 per cent at 32 DAS and at 60 DAS it was 20.19 per cent. Per cent disease index in plots received three sprays recorded 17.89 per cent at 60 DAS. The reduction in the disease was to the extent of 57.87 per cent in treatment with three sprays followed by 54.03 per cent and 46.76 per cent with two and one spray, respectively.

Table 4b indicated that, the PDI was significantly less in treated plots with fungicide at 25 DAS compared to control. The disease progress from 25 to 60 DAS was very fast in control treatments (16.07 to 60.08%) compared to one time fungicide sprayed plots (10.36% to 26.52%).

Further, the PDI in treatment received two and three applications of fungicides have not shown significant difference at 32 and 39 DAS. Disease was reduced to an extent 54.01 to 58.05 per cent when sprayed two and three times, respectively.

Pooled data of two years (Table 5) revealed that, the per cent disease index was observed with 18.59, 21.06 and 25.96 per cent at three, two and one spray plot at 60 DAS, respectively. Whereas, highest PDI of 59.38 was observed in control plot, where no spray was undertaken (Plate 4).

4.4.2 AUDPC value

Highest AUDPC value was obtained in control plots with value of 1179.50 and 1261.23 during 2006 and 2007, respectively. The lowest A value was obtained in plots with three and two applications of fungicides. Similarly, the highest per cent decrease of A value over control were noticed in three sprays (56.87%) followed by two sprays (52.97%) during 2006 (Table 4a), whereas 57.02 and 53.08 per cent in three and two sprayed plots, respectively during 2007 (Table 4b). Similar trend was also noticed with respect to A value and per cent decrease of A value over control in pooled data (Table 5).

4.4.3 Grain yield

The grain yield of greengram differed significantly among the treatments with different spray schedules (Table 4a). During 2006, it was recorded lowest in untreated control (7.57 q/ha), whereas it was 9.20, 12.59 and 12.70 q per ha at one, two and three sprayed plots, respectively. The reduction in the disease due to fungicide resulted in increased grain yield. It was observed that maximum per cent increase in grain yield was recorded in plots with three sprays (67.77%), two (66.31%) and one spray (21.53%).

During the year 2007, the lowest grain yield was recorded in untreated control (7.28 q/ha), whereas, it was 9.09, 12.01 and 12.13 q per ha at one, two and three times sprayed plots, respectively. The highest increase in grain yield of 66.62 per cent was recorded in three sprays followed by two (64.97%) and one (24.86%) spray (Table 4b). Similar trend was observed with respect to grain yield and per cent increase over control in pooled data (Table 5).

4.4.4 Stalk yield

During 2006, maximum stalk yield was recorded in plots received three sprays (15.08 q/ha) and was found on par with two sprays (14.95 q/ha). The increase in stalk yield ranged from 31.09 to 87.56 per cent in one to three sprays (Table 4a). Similar trend was observed with respect to stalk yield and per cent increase over control during 2007 and pooled data also (Table 4b and 5).

4.4.5 Loss in grain and stalk yield

The loss in grain yield due to anthracnose was to the tune of 40.18 per cent in control plot, whereas it was least (0.97%) in plots received two sprays. The highest loss in stalk yield

Table 4a: Crop loss assessment due to anthracnose of greengram caused by *Colletotrichum truncatum* during *Kharif*-2006

Sl. No	Treatment (Carbendazim at 0.1%)	Percent Disease Index at DAS						Percent reduction over control	AUDPC (A) value	Per cent decrease of A value over control	Grain yield (q/ha)	Percent grain yield increase over control	Stalk yield (q/ha)	Percent stalk yield increase over control
		25	32	39	46	53	60							
1	T_1 - One spray	9.90 (18.33)*	14.11 (22.06)	17.15 (24.46)	21.00 (27.26)	21.68 (27.75)	25.11 (30.07)	46.76	640.12	45.73	9.20	21.53	10.54	31.09
2.	T_2 - Two sprays	9.51 (17.95)	13.98 (21.96)	14.04 (22.01)	17.90 (25.03)	18.48 (25.46)	20.19 (26.70)	54.03	554.75	52.97	12.59	66.31	14.95	85.95
3.	T_3 - Three sprays	9.23 (17.68)	13.86 (21.86)	13.96 (21.94)	14.91 (22.71)	16.39 (23.88)	17.89 (25.02)	57.87	508.76	56.87	12.70	67.77	15.08	87.56
4.	T_4 - No spray	14.02 (21.99)	21.07 (27.32)	25.09 (30.06)	38.03 (38.07)	48.13 (43.93)	58.34 (49.80)	-	1179.50	-	7.57	-	8.04	-
	S.E m ±	0.27	0.16	0.14	0.24	0.17	0.13				0.13		0.08	
	CD at 5 %	0.84	0.51	0.44	0.75	0.51	0.41				0.40		0.24	

* Values in parenthesis are arc sine transformed values.

DAS : Days After Sowing

Table 4b: Crop loss assessment due to anthracnose of greengram caused by *Colletotrichum truncatum* during *Kharif*-2007

Sl. No	Treatment (Carbendazim at 0.1%)	Percent Disease Index at DAS						Percent reduction over control	AUDPC (A) value	Per cent decrease of A value over control	Grain yield (q/ha)	Percent grain yield increase over control	Stalk yield (q/ha)	Percent stalk yield increase over control
		25	32	39	46	53	60							
1	T_1 - One spray	10.36 (18.90)*	15.38 (23.09)	18.42 (25.41)	22.03 (27.99)	22.95 (28.62)	26.52 (30.99)	46.95	680.54	46.04	9.09	24.86	9.22	22.44
2.	T_2 - Two sprays	9.75 (18.19)	14.45 (22.34)	15.47 (23.33)	18.86 (25.73)	19.60 (26.28)	22.00 (27.97)	54.01	589.79	53.24	12.01	64.97	14.10	87.25
3.	T_3 - Three sprays	9.53 (17.97)	14.24 (22.17)	14.64 (22.49)	16.13 (23.67)	17.94 (25.06)	19.08 (25.90)	58.05	540.79	57.12	12.13	66.62	14.26	89.38
4.	T_4 - No spray	16.07 (23.62)	22.05 (28.00)	28.09 (32.00)	41.03 (39.83)	50.93 (45.53)	60.08 (50.82)	-	1261.23	-	7.28	-	7.53	-
	S.E m ±	0.27	0.15	0.23	0.21	0.14	0.21				0.10		0.12	
	CD at 5 %	0.84	0.45	0.83	0.66	0.43	0.64				0.32		0.38	

* Values in parenthesis are arc sine transformed values.

DAS : Days After Sowing

Table 5: Crop loss assessment due to anthracnose of greengram caused by *Colletotrichum truncatum* during *Kharif*-2006 and 2007 (Pooled data)

Sl. No	Treatment (Carbendazim at 0.1%)	Percent Disease Index at DAS						Percent reduction over control	AUDPC (A) value	Per cent decrease of A value over control	Grain yield (q/ha)	Percent grain yield increase over control	Stalk yield (q/ha)	Percent stalk yield increase over control
		25	32	39	46	53	60							
1	T_1 - One spray	9.95 (18.62)*	14.74 (22.58)	17.82 (24.97)	21.51 (27.63)	22.32 (28.19)	25.96 (30.63)	46.81	661.29	45.84	9.47	27.46	9.88	26.83
2.	T_2 - Two sprays	9.63 (18.07)	14.22 (22.15)	14.82 (22.67)	18.38 (25.38)	19.04 (25.87)	21.06 (27.32)	54.07	572.85	53.08	12.30	65.55	14.53	86.52
3.	T_3 - Three sprays	9.38 (17.83)	14.05 (22.01)	14.26 (22.19)	15.52 (23.20)	17.17 (24.47)	18.59 (25.54)	57.95	524.83	57.02	12.42	67.16	14.67	88.32
4.	T_4 - No spray	15.04 (22.82)	21.56 (27.66)	26.60 (31.05)	39.53 (38.95)	49.53 (44.73)	59.38 (50.40)	-	1221.01	-	7.43	-	7.79	
	S.E m ±	0.19	0.11	0.16	0.18	0.10	0.15				0.18		0.06	
	CD at 5 %	0.58	0.32	0.57	0.56	0.32	0.45				0.57		0.19	

* Values in parenthesis are arc sine transformed values.

DAS : Days After Sowing

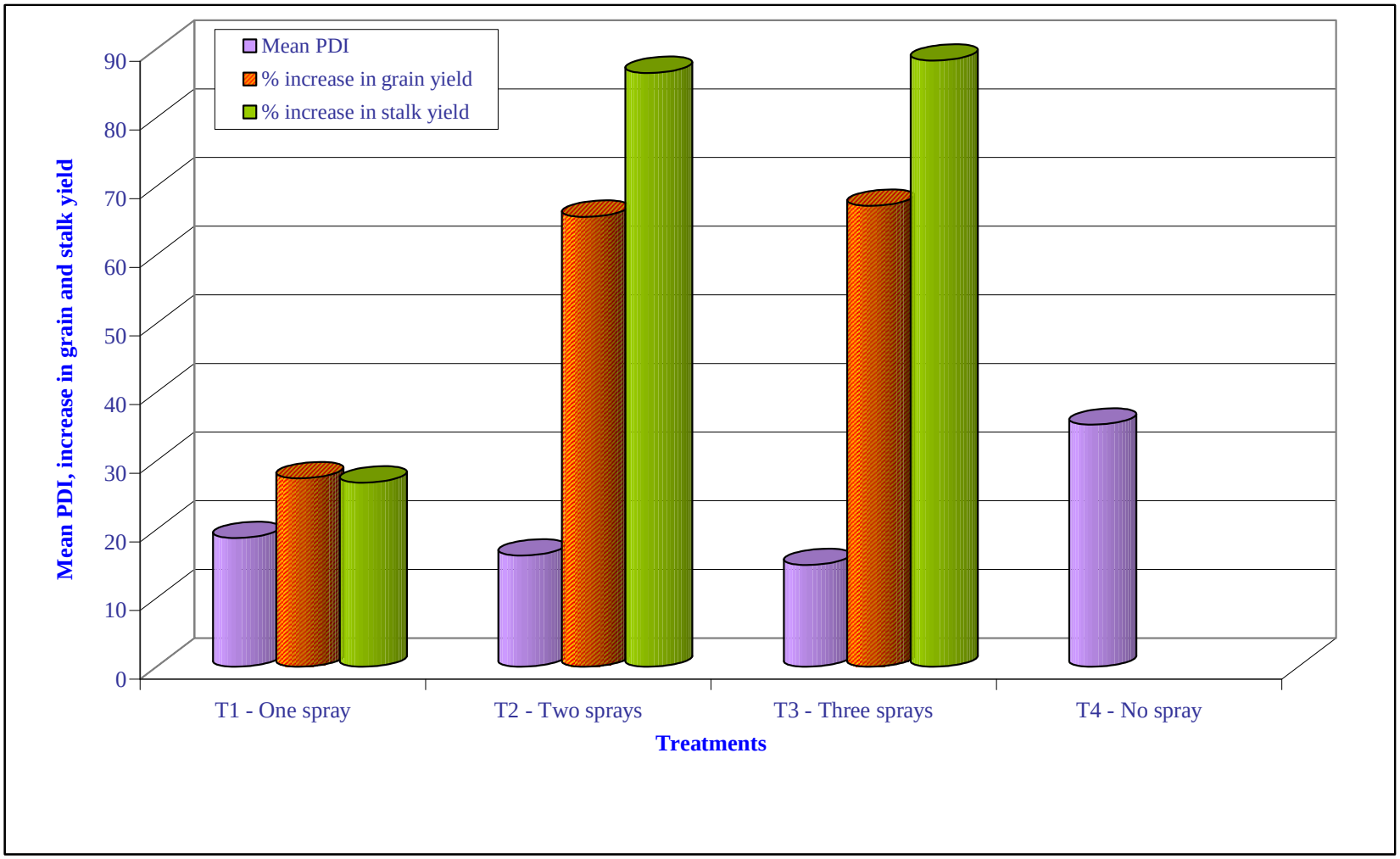
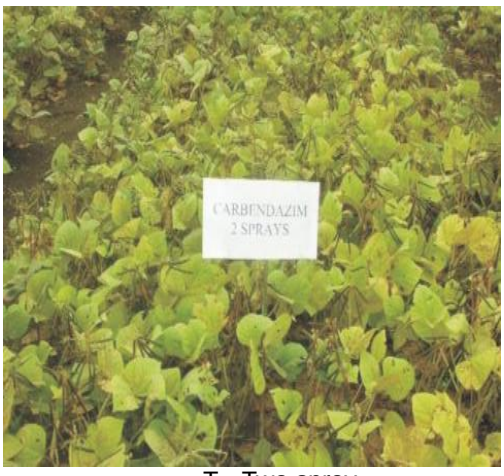


Fig. 2: Effect of number of carbendazim (0.1%) sprays on mean PDI, increase in grain and stalk yield of greengram during 2006 and 2007

Experimental field view

T_1- One spray



T_2- Two spray

T_3-Three spray

T_4- Four spray

Plate4: Effect of different sprays of carbendazim (0.1%) on severity of anthracnose on susceptible greengram variety, Chinamung

was also recorded in control plot (46.90%) followed by plots received one (32.65%) and two (0.95%), sprays of carbendazim (Table 7).

4.4.6 Benefit:Cost ratio (BCR)

Maximum BCR of 17.44 and 16.43 was obtained from plots which have received two sprays of fungicides during 2006 and 2007, respectively (Table 6). The mean BCR of both the years revealed the similar benefit from the treatments received two sprays (16.94) followed by one spray (12.35) and three sprays (11.18) (Table 7).

4.4.7 Crop loss model

The investigation on crop loss due to anthracnose of greengram was carried out and also an attempt was made to identify the relationship between grain and stalk yield and level of per cent disease index at weekly interval from 25 to 60 DAS in susceptible variety Chinamung. Simple linear regression models were developed and presented in Table 8.

During *kharif* 2006, maximum correlation coefficient of 0.84 was obtained between the PDI and the grain yield recorded at 60 DAS, however during *kharif* 2007, it was found to be 0.89. In both years, the coefficient of determination (R^2) of 0.75 and 0.79 were obtained. In case of pooled linear regression model, the correlation coefficient of disease with the yield was found to be 0.87 and coefficient of determination (R^2) was 0.76. Further, the observed and predicted grain yield values were compared and found that there was no much deviation. During 2006 and 2007 difference in yield values were reported to be in the range of 0.30 to -1.90 and 0.25 to -1.60, respectively (Table 9 and Fig. 4).

4.5 Cultural and physiological studies

4.5.1 Cultural studies

4.5.1.1 Growth of *C. truncatum* on potato dextrose broth at different incubation periods

The experiment was conducted to ascertain the number of days required for maximum growth of the fungus by monitoring the dry mycelial weight. The results are presented in Table 10 and Fig. 5.

There was significant difference among the incubation periods. The dry mycelial weight of *C. truncatum* gradually increased from third day (94.41 mg) and reached maximum on 15^{th} day (214.13 mg) and was significantly superior over all other treatments. It showed declining trend from 17^{th} day onwards.

4.5.1.2 Growth and sporulation of *C. truncatum* on different solid media

The diversity in cultural and morphological characters of *C. truncatum* was studied on ten solid media at room temperature (28 $\pm$ 1^0C). The radial growth of the fungus and sporulation were recorded when it attained the maximum growth in all the media tested. Observations on various colony characters were recorded. The data were presented in Table 11 and Fig. 6. The fungus recorded maximum growth on potato dextrose agar (90.00 mm) and was found superior over other media. Next to potato dextrose agar, oat meal agar (85.20 mm) and Czapek's agar (84.23 mm), which were on par with each other. The minimum growth was observed in potato carrot agar (41.29 mm) (Plate 5).

The maximum sporulation was found in potato dextrose agar and oat meal agar. Poor sporulation was noticed in yeast extract agar and potato carrot agar. With respect to the mycelial colour, it varied from dull white to grey. The growth varied from flat to fluffy with smooth to irregular margins. The fungus showed light grey coloured with smooth margin mycelia on potato dextrose agar with excellent sporulation. Mycelial growth on Czapek's dox agar was light grey in colour and rough margin having good sporulation. Mycelial growth on host extract agar showed dark grey colour and margin was smooth with fair sporulation. Yeast extract and Sabouraud's agar media produced dull white colour mycelia with slight flat to fluffy growth having poor to fair sporulation. In case of potato carrot agar, malt extract agar and corn meal agar, the fungus showed dirty white coloured with slight flat to irregular margin mycelium with poor to fair sporulation.

4.5.1.3 Growth and sporulation of *C. truncatum* in different liquid media

Average dry mycelial weight and sporulation of *C. truncatum* grown on ten different liquid media after 15 days of incubation was recorded as descried in "Material and Methods" and data are presented in Table 12 and depicted in Fig. 7.

Table 6: Benefit:Cost ratio in loss estimation study due to anthracnose as influenced by the number of fungicidal sprays

Spray No	Cost of fungicides (Rs.)	Labour (Rs.)	Total Cost (Rs)	Additional increase in yield over control (q/ha)				Additional benefit (Rs.)						Benefit:Cost ratio	
				2006		2007		2006			2007			2006	2007
				Grain	Stalk	Grain	Stalk	Grain	Stalk	Total	Grain	Stalk	Total		
1.	225	90	315	1.63	2.50	1.81	1.69	3586	125	3711	3982	85	4067	11.78	12.91
2.	473	180	653	5.02	6.91	4.73	6.57	11044	346	11390	10406	329	10735	17.44	16.43
3.	743	270	1013	5.13	7.04	4.85	6.73	11286	352	11638	10670	337	11007	11.49	10.87

Note: Cost of fungicide (Carbendazim) : Rs. 450/kg
Cot of labour/spray /ha : Rs. 90/- (Male Rs. 50/- + Female Rs. 40/-)
Grain value : Rs. 2200/q
Stalk value : Rs. 50/q

Table 7: Effect of number of sprays of carbendazim on severity of anthracnose, yield parameters and benefit:cost ratio in greengram during 2006 and 2007

Sl. No.	Treatment	PDI	Grain yield		Stalk yield		BCR
			Yield (q/ha)	Loss over T_3 (%)	Yield (q/ha)	Loss over T_3 (%)	
1	T_1 - One spray	18.76	9.47	23.75	9.88	32.65	12.35
2	T_2 - Two sprays	16.20	12.30	0.97	14.53	0.95	16.94
3	T_3 - Three sprays	14.83	12.42	0	14.67	0	11.18
4	T_4 - No spray	35.27	7.43	40.18	7.79	46.90	-

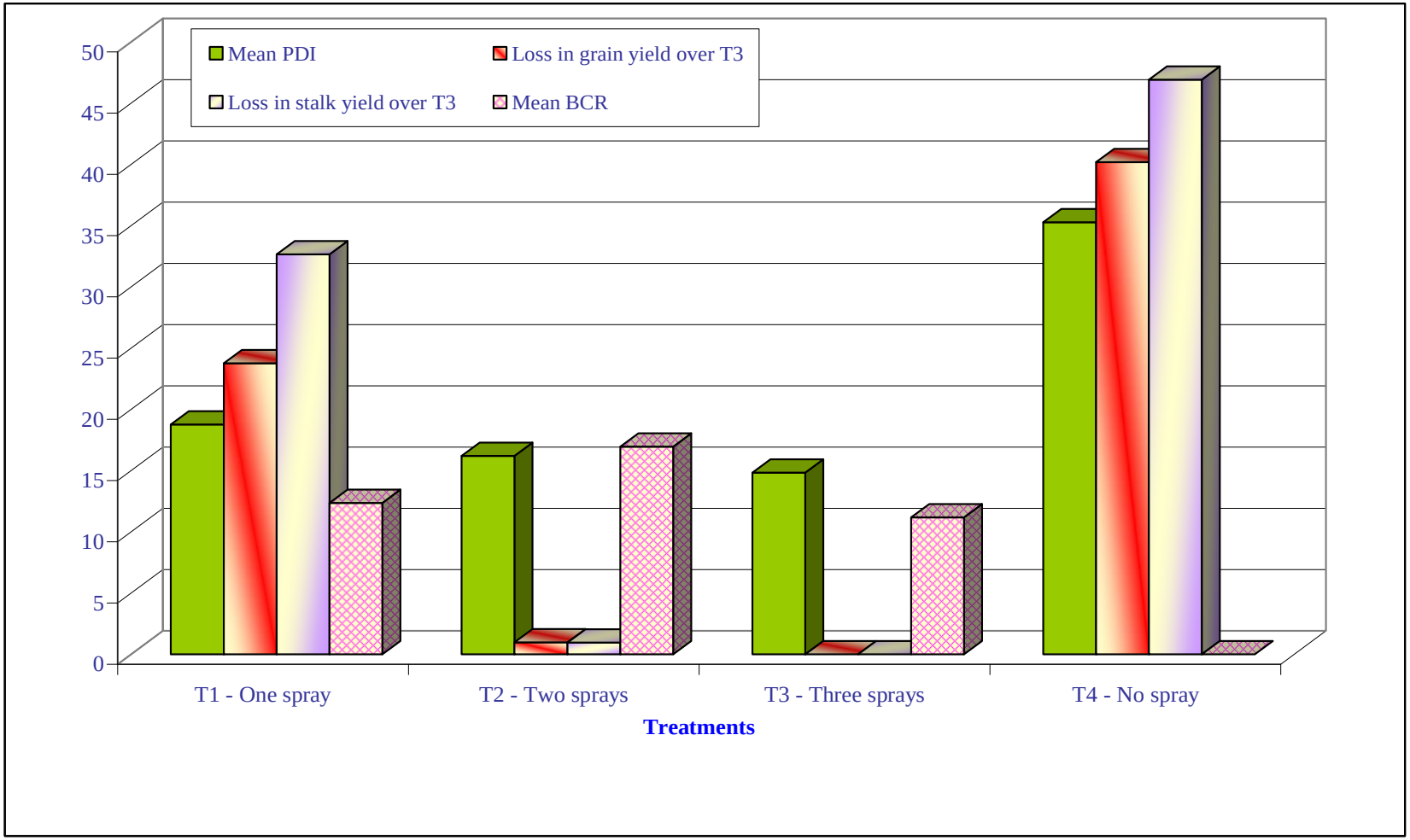


Fig. 3: Effect of number of sprays of carbendazim (0.1%) on mean PDI, loss in grain and stalk yield (q/ha) and mean benefit:cost ratio (BCR) of greengram during 2006 and 2007

Table 8: Crop loss models showing the relationship between anthracnose PDI and yield (q/ha) of greengram

Yield	Model for yield	r	R^2
2006	Y=15.458 – 0.240 PDI	-0.84	0.75
2007	Y=14.825 – 0.214 PDI	-0.89	0.79
Pooled	Y = 15.141 – 0.227 PDI	-0.87	0.76

Table 9: Observed and predicted grain yield (q/ha) due to anthracnose of greengram during *kharif* 2006 and 2007

Sl. No.	Treatment (carbendazim at 0.1%)	*Kharif* 2006			*Kharif* 2007		
		Yield (q/ha)		Difference	Yield (q/ha)		Difference
		Observed	Predicted		Observed	Predicted	
1	T_1 - One spray	9.20	11.10	-1.90	9.09	10.69	-1.60
2	T_2 - Two sprays	12.59	11.69	0.90	12.01	11.24	0.77
3	T_3 - Three sprays	12.70	12.01	0.69	12.13	11.55	0.58
4	T_4 - No spray	7.57	7.27	0.30	7.28	7.03	0.25

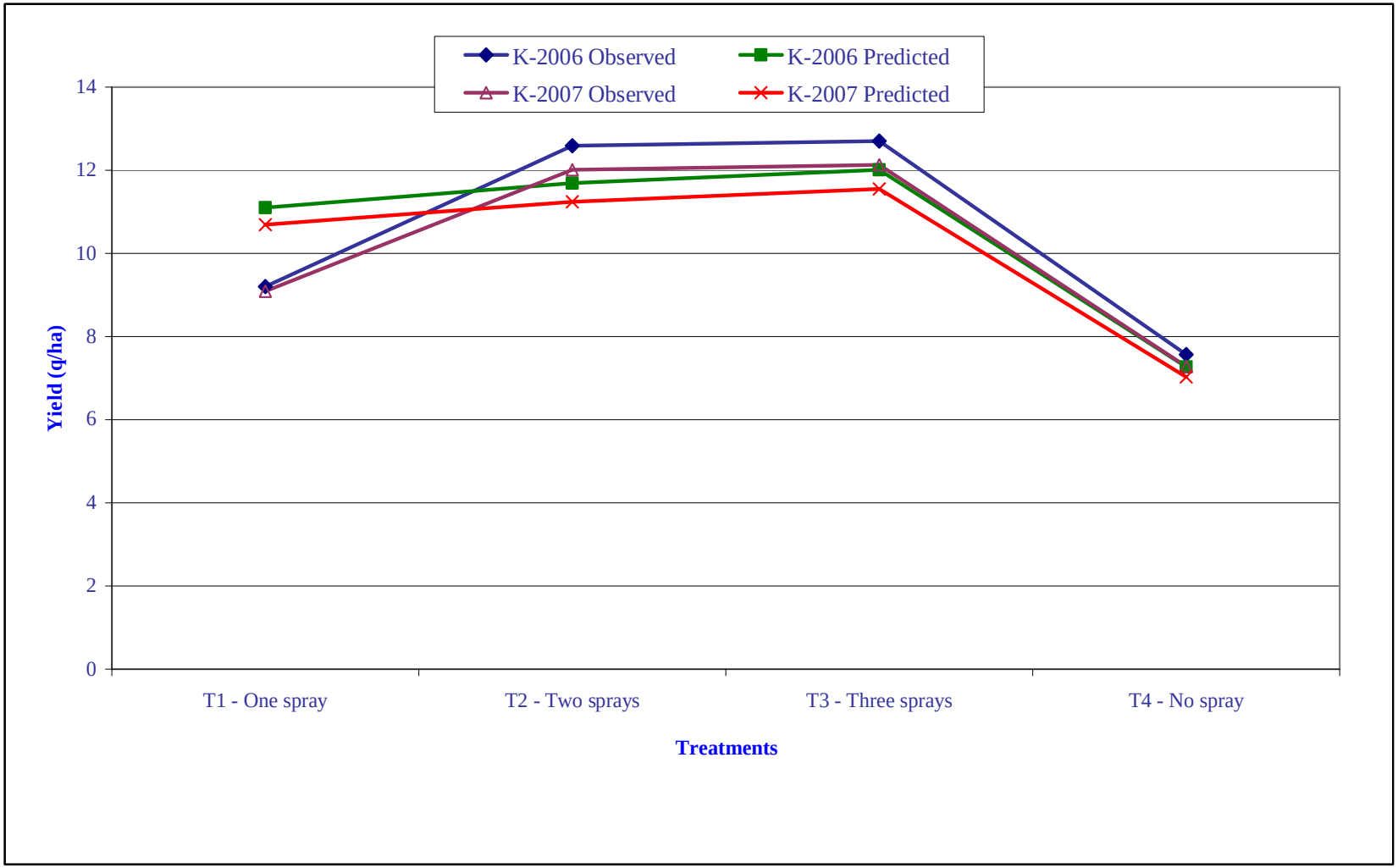


Fig. 4: Observed and predicted grain yield (q/ha) values

Table 10: Effect of incubation period on growth of *Colletotrichum truncatum* on potato dextrose broth

Sl. No.	Days after inoculation	Dry mycelial weight (mg)
1.	3	94.41
2.	5	122.40
3.	7	126.47
4.	9	146.37
5.	11	161.22
6.	13	184.38
7.	15	214.13
8.	17	197.18
9.	19	173.38
10.	21	122.37
S.E m ±		0.58
CD at 1 %		2.35

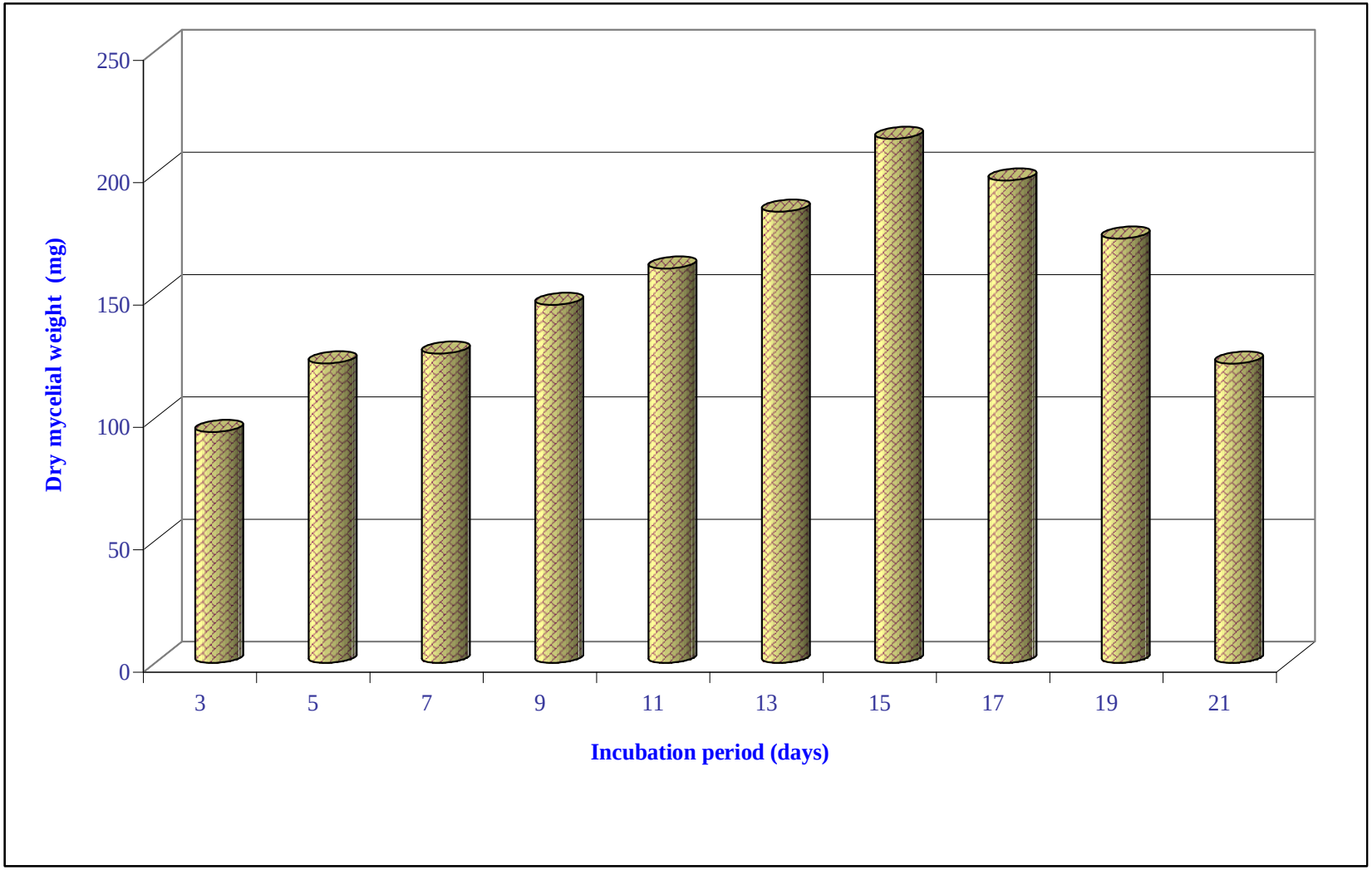


Fig. 5: Growth of C. truncatum on potato dextrose broth at different incubation periods

Data from the Table 12 indicated that, there was significant difference between the liquid media. Richard's medium supported the maximum growth (203.17 mg) and was significantly superior over other media. Next to Richard's medium Czapek's medium (168.30 mg), potato dextrose broth (162.30 mg) and Sabouraud's media (159.17 mg) were found best and differed significantly with each other. The minimum growth was observed in potato carrot medium (34.43 mg) (Plate 6).

Excellent sporulation was observed in Richard's and potato dextrose medium, while fair sporulation was noticed in Czapek's, host extract, Sabouraud's, malt extract and corn meal medium. Poor sporulation was seen in oat meal, yeast extract and potato carrot medium.

4.5.2 Physiological studies

4.5.2.1 Effect of temperature on the growth and sporulation of *C. truncatum*

The fungus *C. truncatum* was grown on potato dextrose agar medium at different temperatures *viz.*, 10, 15, 20, 25, 30, 35 and 40^0C to know the suitable temperature requirement for their maximum radial growth and sporulation (Table 13 and Fig. 8).

Data from Table 13 revealed that, maximum mean colony diameter of fungus at temperatures of 30^0C (80.17 mm) and 25^0C (79.77 mm) was significantly superior over all other temperatures and these were on par with each other. Lowest mean colony diameter was obtained at temperatures of 10^0C (23.47 mm) and 40^0C (21.30 mm) which were on par with each other (Plate 7).

Excellent sporulation was observed at both 25 and 30^0C. Further, good sporulation was at 20^0C and fair at 35^0C, while in 10, 15 and 40^0C temperatures poor sporulation was seen.

4.5.2.3 Effect of relative humidity (RH) on the growth and sporulation of *C. truncatum*

The data pertaining to effects of relative humidity levels for the growth and sporulation are presented in Table 14 and Fig. 9.

The radial growth and sporulation of the fungus was observed maximum at 95.00 per cent relative humidity (85.05 mm), which was on par with 85.00 per cent relative humidity (83.68 mm). Lowest colony diameter was obtained at RH level of 65.00 per cent. Relative humidity levels of 85 and 95 per cent were found to be favourable for the excellent sporulation of fungus followed by good sporulation at 75 per cent RH (Plate 8).

4.5.2.2 Effect of hydrogen ion concentration (pH) on the growth and sporulation of *C. truncatum*

The fungus was grown in Richard's medium at different pH levels and observations on dry mycelial weight and sporulation was taken as described in "Material and Methods" and data are presented in Table 15 and depicted in Fig. 10.

Data from Table 15 revealed that, *C. truncatum* grew well at 6.5 pH with maximum dry mycelial weight of 103.27 mg and was significantly superior over other pH levels followed by pH level of 6.0 (95.53 mg). At pH 5.5 and 7.0, it was on par with each other. The least mycelial weight was obtained at pH of 8.0 (49.50 mg). The fungus responded with excellent sporulation at 6.0 and 6.5 pH levels. Whereas, at 5.5 and 7.0 pH levels, it showed good sporulation. Poor sporulation was noticed at pH levels of 4.0, 4.5 and 8.0 (Plate 9).

4.5.2.4 Effect of light intensities on the growth and sporulation of *C. truncatum*

The effect of light on the fungal growth and sporulation was studied. The results are presented in Table 16 and Fig. 11.

The data presented in Table 16 indicated that exposure of petridishes to alternate cycle of 12 hr light under day light tubes and 12 hr darkness resulted in maximum mycelial growth of the fungus to the tune of 82.50 mm as well as excellent sporulation. Similarly, exposure of petridishes to continuous darkness resulted in production of mean colony diameter of 70.27 mm with excellent sporulation. The least mean colony diameter of 61.97 mm was observed in the petridishes which was exposed to continuous light with good sporulation. All the treatments differed significantly (Plate 10).

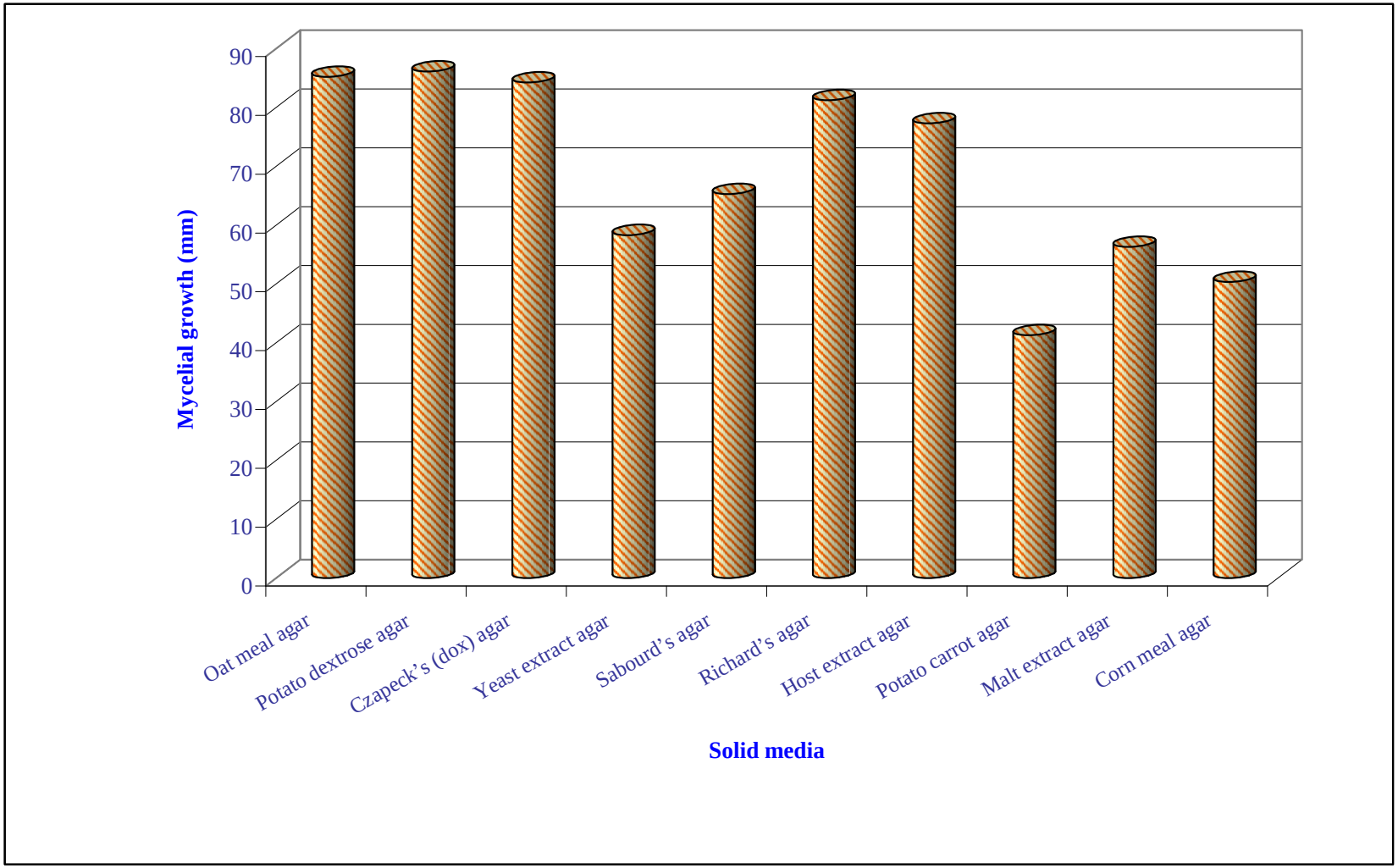


Fig. 6: Effect of different solid media on growth and development of C. truncatum

Table 12: Effect of different liquid media on dry mycelial weight and sporulation of Colletotrichum truncatum

Sl. No.	Media	Dry mycelial weight (mg)	Sporulation
1	Czapek`s medium	168.30	+ +
2	Richard`s medium	203.17	+ + + +
3	Potato dextrose broth	162.30	+ + + +
4	Host extract medium	144.23	+ +
5	Sabouraud`s medium	159.17	+ +
6	Oat meal medium	131.23	+
7	Yeast extract medium	105.24	+
8	Potato carrot medium	34.43	+
9	Malt extract medium	88.30	+ +
10	Corn meal medium	64.22	+ +
S.Em±		0.66	
CD at 1%		2.70	

Sporulation : + + + + = Excellent, + + + = Good, + + = Fair, + = Poor, - = No sporulation

4.6 Epidemiological studies

4.6.1 Aerobiology

As a part of epidemiology of the disease, aerobiological studies such as trapping of anthracnose conidia, first appearance of spores and disease and progression of spore load and development of disease were carried out during *kharif* 2006 and *kharif* 2007 and data are presented in Table 17, 18 and depicted in Fig. 12, 13a and 13b.

The investigation on sampling of air borne conidia of *C. truncatum* in greengram field was carried out throughout the cropping season under different environmental conditions, which was aimed primarily to understand the epidemiology of the disease. The pathogen was releasing the inoculum continuously in the form of conidia throughout the crop season. Hence, air sampling for presence of conidia was carried out by mounting spore trap in greengram experimental field during 2006-07 and 2007-08 (Plate 11a and 11b). The data on intensity of anthracnose in relation to atmospheric spore load and weather parameters were presented in Table 17 and 18.

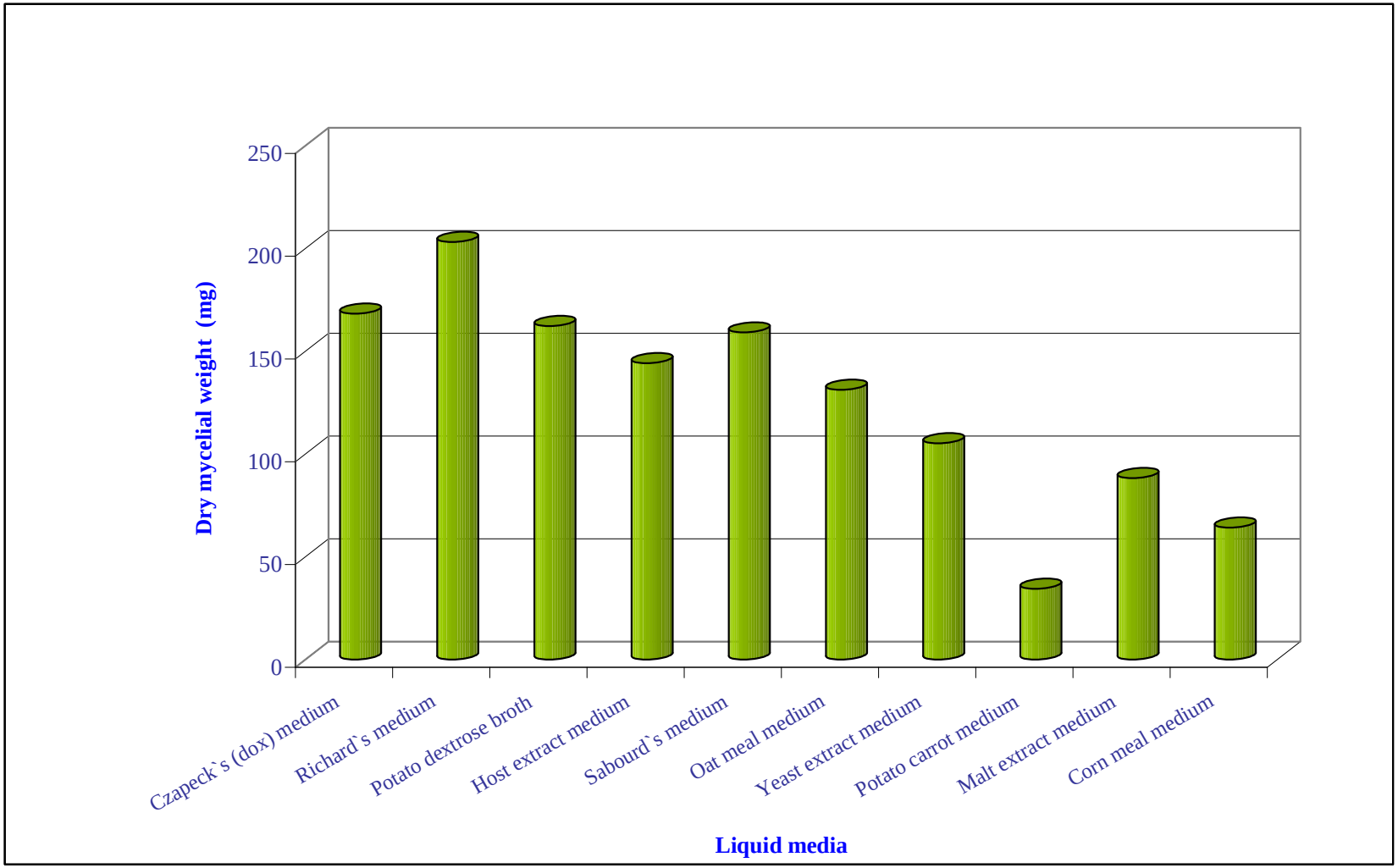


Fig. 7: Effect of different liquid media on dry mycelial weight of C. truncatum

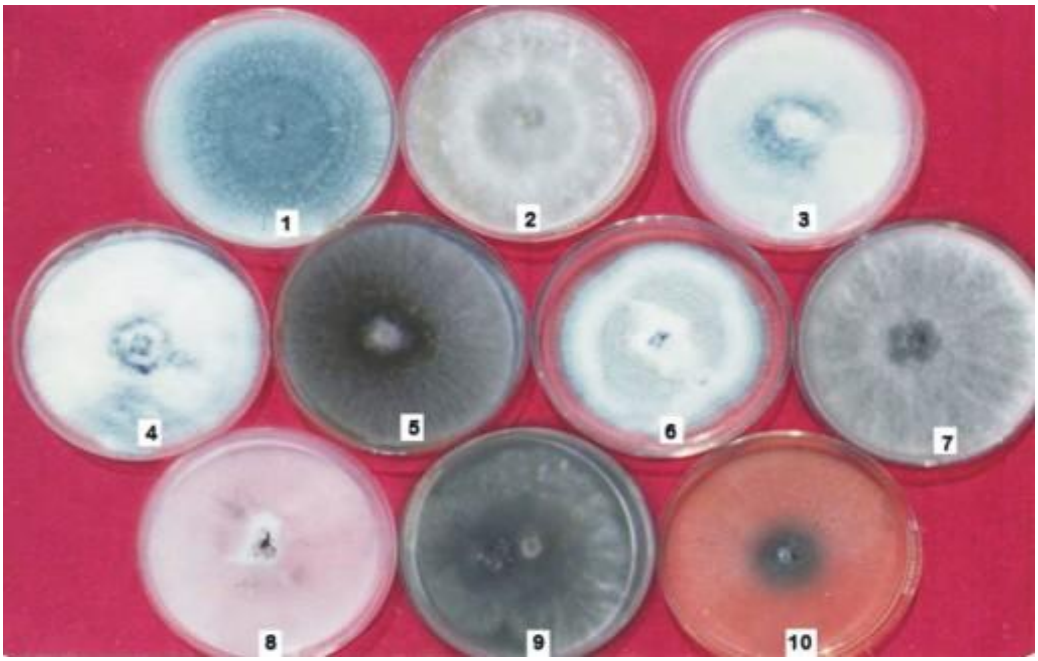


Plate 5: Effect of different solid media on growth and development of C. truncatum

1. Potato dextrose agar
2. oat meal agar
3. yeast extract agar
4. Sabouraud's agar
5. Host extract agar
6. Corn meal agar
7. Czapek's agar
8. Malt extract agar
9. Richard's agar
10. Potato carrot agar

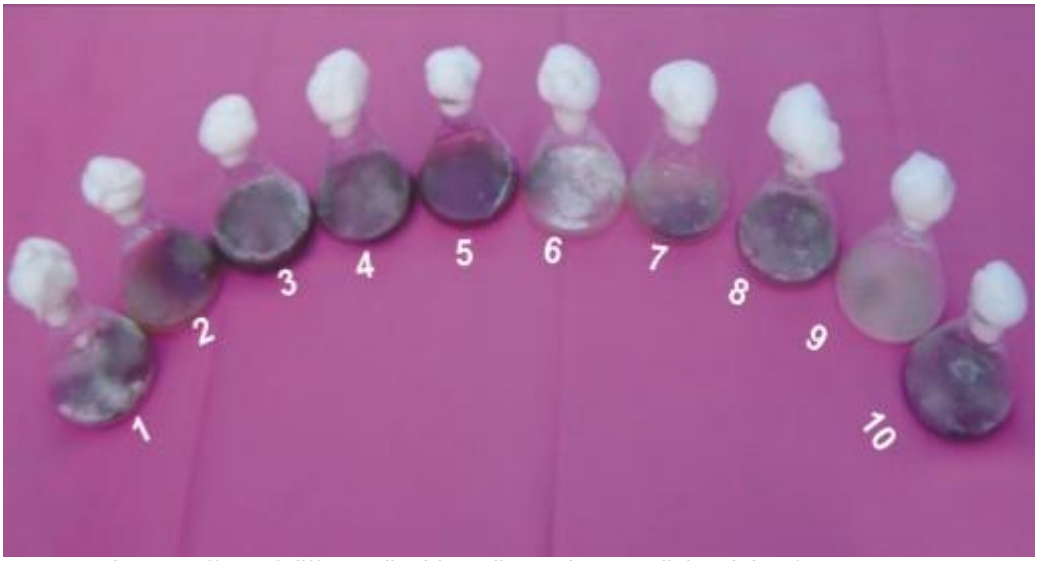


Plate 6. Effect of different liquid media on dry mycelial weight of C.truncatum

1.Richard's medium
2. Host extract medium
3. Potato dextrose broth
4. Oat meal medium
5. Malt extract medium
6. Yeast extract medium
7. Potato carrot medium
8. Sabourad's medium
9. Corn meal medium
10. Czapek,s medium

Table 13: Effect of temperature on growth and sporulation of *Colletotrichum truncatum*

Sl. No.	Temperature (ºc)	Mean colony diameter (mm)	Sporulation
1	10	23.47	+
2	15	35.30	+
3	20	50.33	+ + +
4	25	79.77	+ + + +
5	30	80.17	+ + + +
6	35	60.23	+ +
7	40	21.30	+
S.Em±		0.53	
CD at 1%		2.29	

Sporulation : + + + + = Excellent, + + + = Good, + + = Fair, + = Poor, - = No sporulation

The data presented in Table 17 revealed that during *kharif* 2006, the number of spores trapped daily varied as the disease severity progressed in the field. Air sampling carried out during 2006 indicated that, the first appearance of spores in the atmosphere was recorded after 13 days after sowing. The spore load gradually increased and reached peak of 22.14 during 31st standard meteorological week (July 30 – August 05), while it was least during 25th week (4.70). Similarly, PDI was also recorded lowest during 25th standard week (9.26) and increased throughout the cropping period. It was peak during last stage *i.e.,* 32nd standard week (SW) (68.50). During the cropping period maximum temperature ranged from 26.56^{0}C (31st SW) to 34.51^{0}C (24th SW), minimum temperature from 20.57^{0}C (31st SW) to 23.60^{0}C (24th SW), relative humidity (morning) from 77.43 per cent (24th SW) to 96.00 per cent (31st SW) and relative humidity (evening) from 37.14 per cent (24th SW) to 79.14 per cent (31st SW). However, rainfall was very erratic. It ranged from 1.5 mm (28th SW) to 173.2 mm (31st SW).

Table 18 revealed that during *kharif* 2007, the first appearance of spore load in the atmosphere was recorded after 12 days of sowing. The spore load gradually increased from 5.10 to maximum of 23.05 during 32nd SW. The PDI trend followed similar to that of previous year. It was least (10.40) in the beginning and increased continuously every week and reached the peak during the last stage *i.e.,* 32nd SW (70.12). The maximum temperature ranged from 27.77^{0}C (26th SW) to 34.69^{0}C (24th SW), minimum temperature from 20.24^{0}C (26th SW) to 22.97^{0}C (24th SW), relative humidity morning from 86.29 (24th SW) to 95.00 per cent (26th SW) and relative humidity evening from 41.29 per cent (24th SW) to 70.14 per cent (26th SW). Rainfall was highly erratic like previous year, which ranged from 0 mm (27th SW) to 129.7 mm (25th SW) (Fig. 13a and 13b). In general, the spore concentration was remarkably higher during 2007 when compared to 2006.

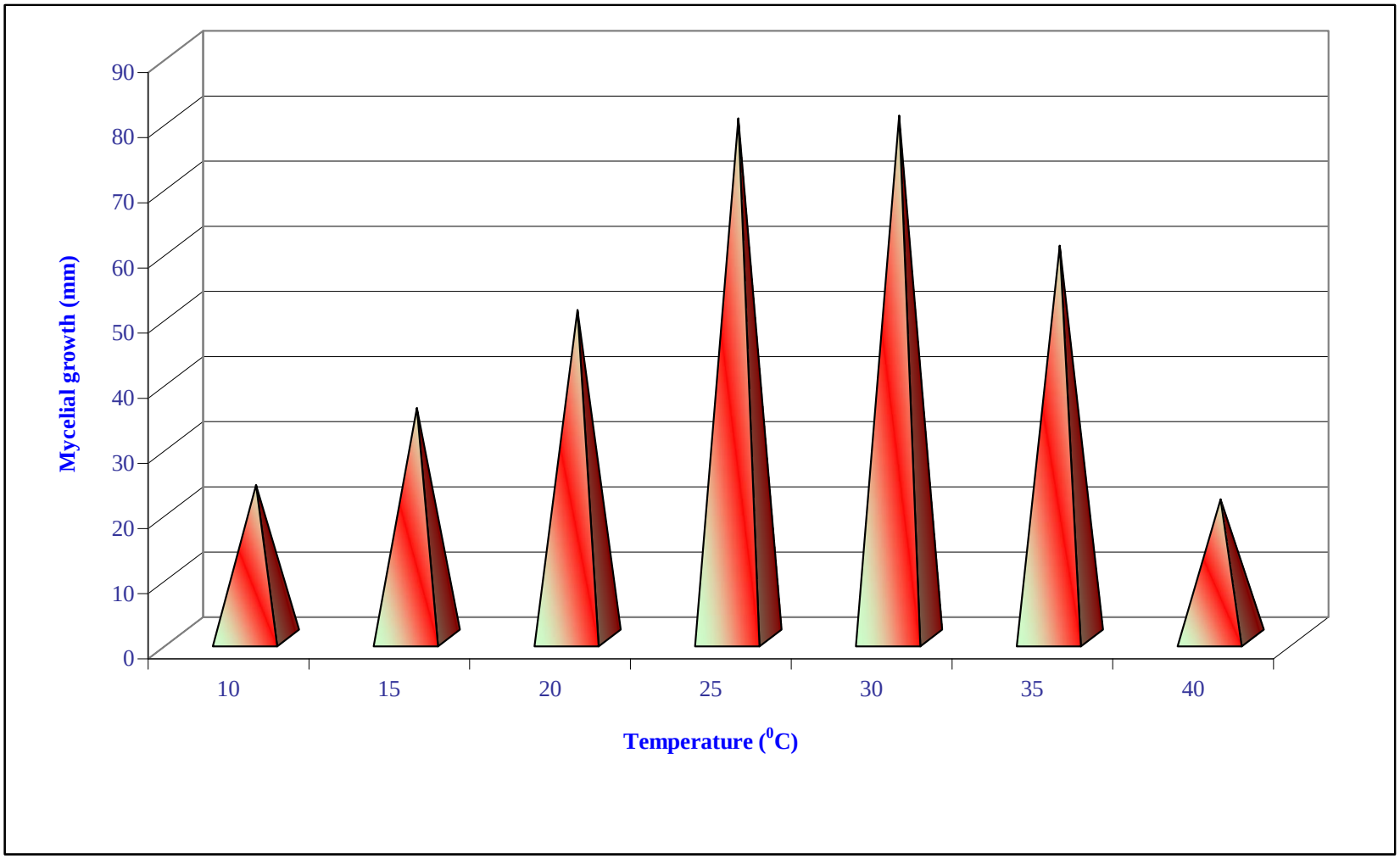


Fig. 8: Effect of temperature on mycelial growth of C. truncatum

Table 14: Effect of relative humidity on growth and sporulation of *Colletotrichum truncatum*

Sl. No.	Relative Humidity (%)	Mean colony diameter (mm)	Sporulation
1	65	60.78	+ +
2	75	74.13	+ + +
3	85	83.68	+ + + +
4	95	85.05	+ + + +
5	100	64.15	+ +
S.Em±		0.59	
CD at 1%		2.56	

Sporulation : + + + + = Excellent, + + + = Good, + + = Fair, + = Poor, - = No sporulation

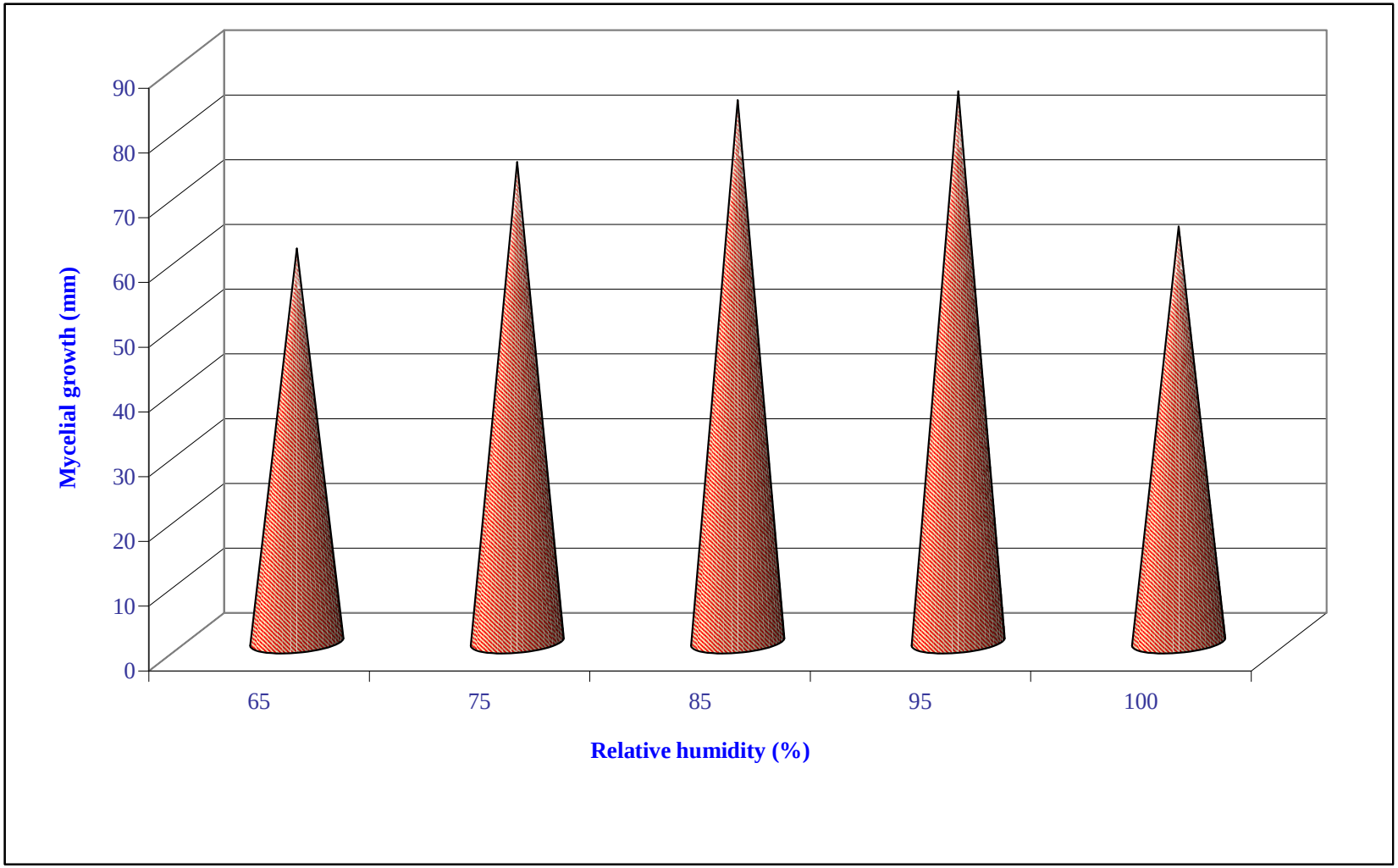


Fig. 9: Effect of relative humidity (RH) levels on mycelial growth of C. truncatum

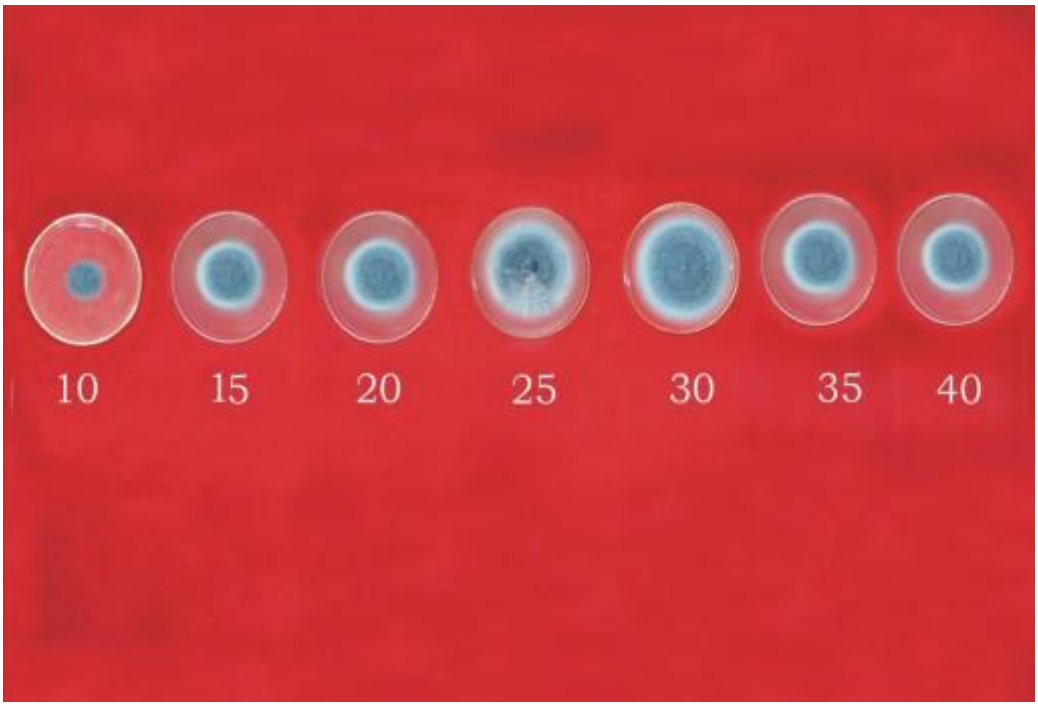


Plate 7: Effect of temperature (^{0}C) on mycelial growth of C. truncatum

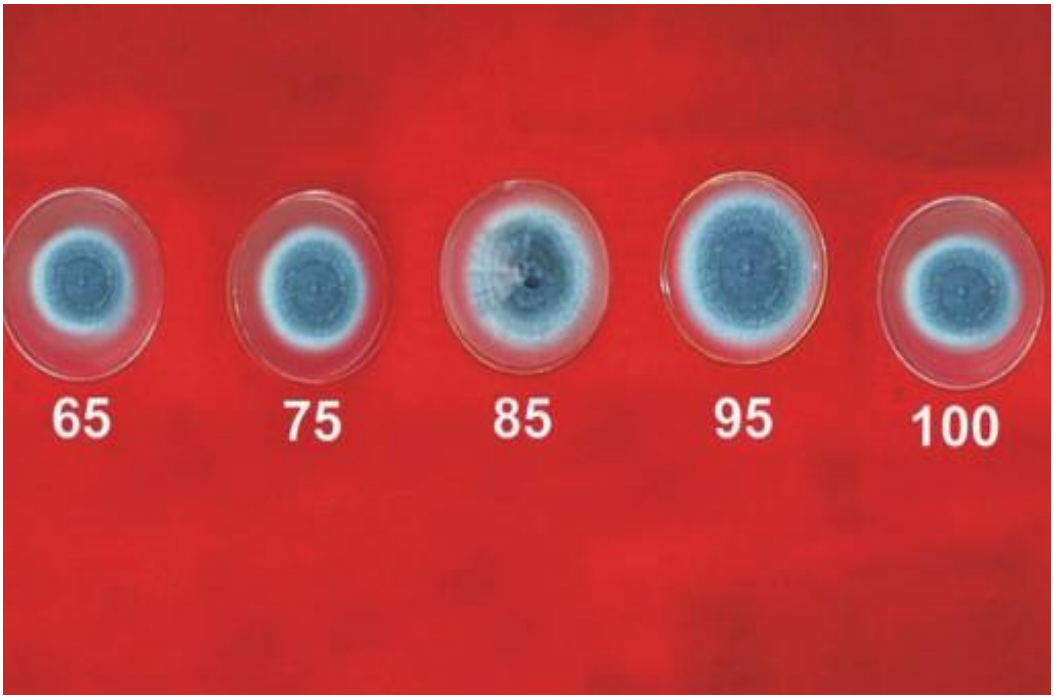


Plate 8: Effect of relative humidity (%) on mycelial growth of C. truncatum

Table 15: Effect of pH on dry mycelial weight and sporulation of *Colletotrichum truncatum*

Sl. No.	pH level	Mean mycelial dry weight (mg)	Sporulation
1	4.0	52.47	+
2	4.5	73.33	+
3	5.0	80.37	+ +
4	5.5	92.38	+ + +
5	6.0	95.53	+ + + +
6	6.5	103.27	+ + + +
7	7.0	91.30	+ + +
8	7.5	78.40	+ +
9	8.0	49.50	+
S.Em±		0.56	
CD at 1%		2.33	

Sporulation : + + + + = Excellent, + + + = Good, + + = Fair, + = Poor, - = No sporulation

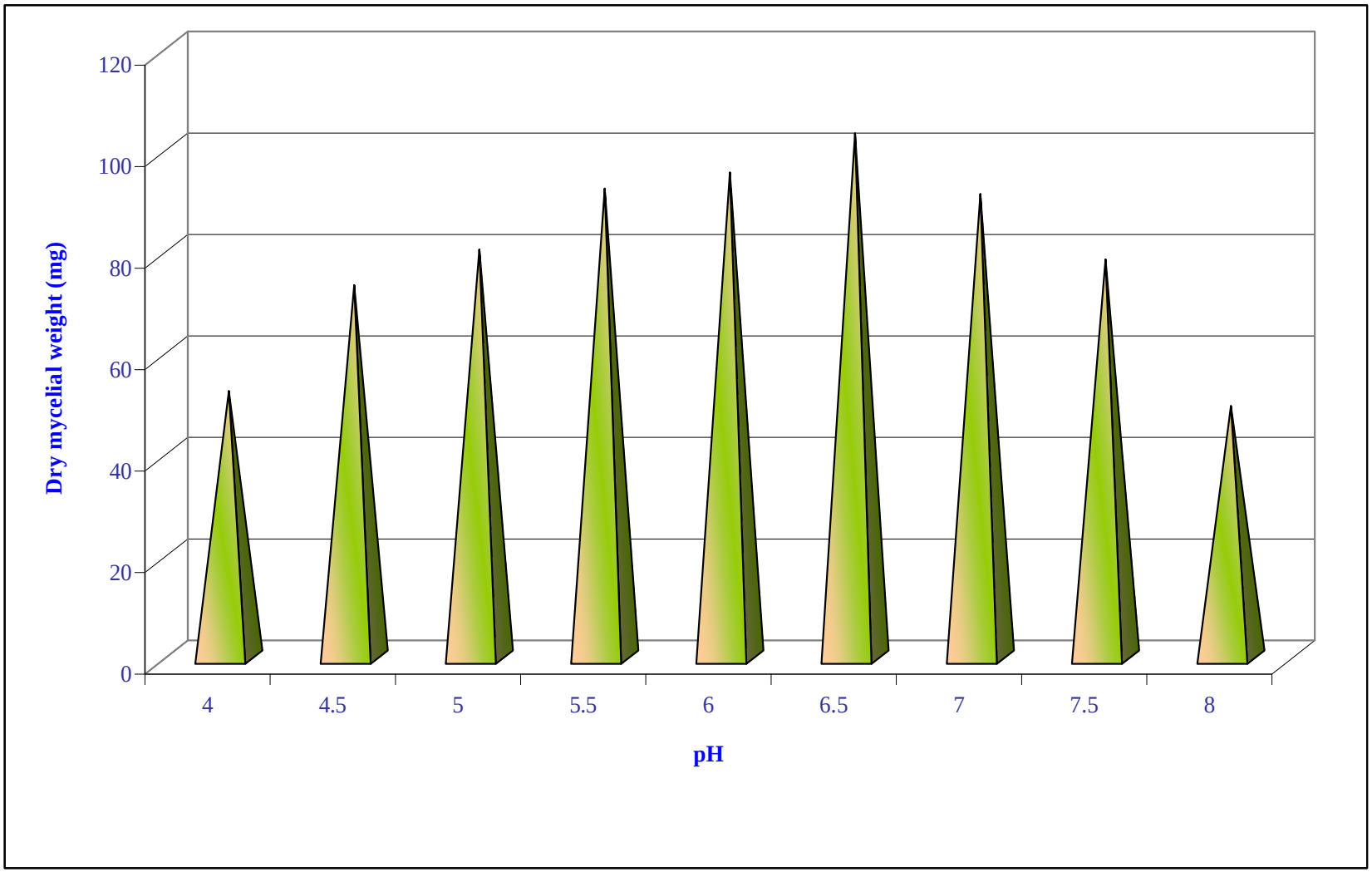


Fig. 10: Effect of pH on dry mycelial weight of C. truncatum

Table 16: Effect of light on the growth and sporulation of *Colletotrichum truncatum*

Sl. No.	Light duration	Mean colony diameter (mm)	Sporulation
1	Continuous dark	70.27	+ + + +
2	Continuous light	61.97	+ + +
3	Alternate cycles of light & dark	82.50	+ + + +
S.Em±		0.40	
CD at 1%		1.73	

Sporulation : + + + + = Excellent, + + + = Good, + + = Fair, + = Poor, - = No sporulation

4.6.1.1 Correlation and multiple linear regression analysis between spore load of *C. truncatum* in relation to weather parameters

An attempt was made to establish the relationship between weather factors *viz.*, minimum and maximum temperature, morning and evening relative humidity and rainfall with spore load of *C. truncatum* through correlation and multiple linear regression analysis. The correlation coefficients for both the years (2006 and 2007) are presented in Table 19.

The relationship between spore load of *C. truncatum* and weather factors during 2006 indicated a negatively higher correlation between maximum and minimum temperature with a correlation coefficient of -0.821 and -0.891, respectively. While, relative humidity morning (0.762) and relative humidity evening (0.728) were significantly positively correlated with weekly spore load but rainfall was non-significantly positively correlated.

During 2007 also, spore load was significantly negatively correlated with temperature and positively with relative humidity. However, the pooled analysis of both the years revealed that maximum temperature (-0.830) and minimum temperature (-0.806) were significantly negatively correlated while relative humidity morning (0.657), relative humidity evening (0.645) and rainfall (0.047) were positively correlated with weekly spore load.

The multiple linear regression of spore load of *C. truncatum* in relation to weather parameters, during 2006 indicated that regression coefficients for maximum temperature, minimum temperature, morning relative humidity, evening relative humidity and rainfall were -2.42, -8.32, -0.83, 0.07 and -0.07, respectively. Whereas during 2007, the regression coefficients for these weather parameters observed were of 3.89, -12.39, 0.52, 0.36 and -0.15, respectively. The pooled regression coefficients for maximum temperature, minimum temperature, morning RH, evening RH and rainfall were -2.55, -2.18, 0.09, -0.04 and -0.05, respectively (Table 20).

During 2006, the multiple linear regression equation was fitted to the data and the equation arrived for the weather parameters was $Y = 344.10 – 2.42\ X_1 – 8.32\ X_2 – 0.83\ X_3 + 0.07\ X_4 – 0.07\ X_5$.

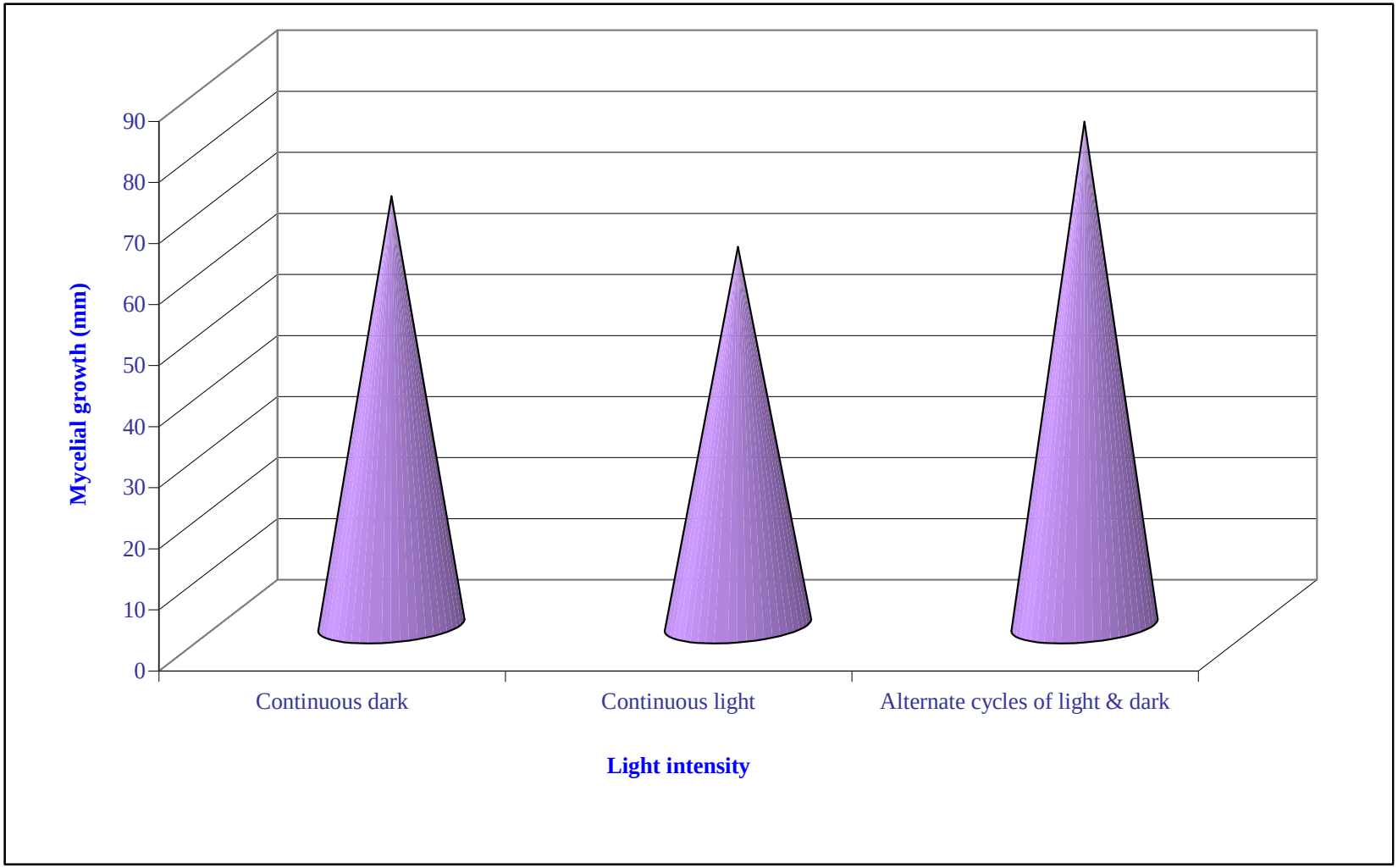


Fig. 11: Effect of light on mycelial growth of C. truncatum

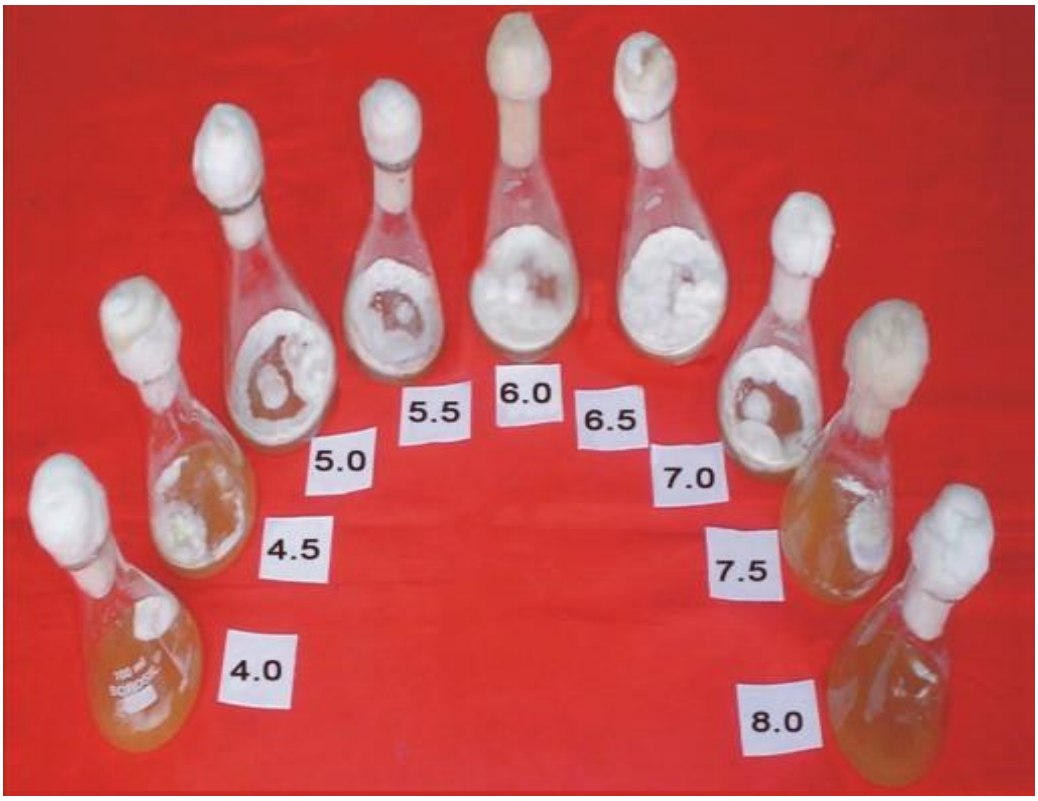


Plate 9: Effect of pH on dry mycelial weight of C. truncatum

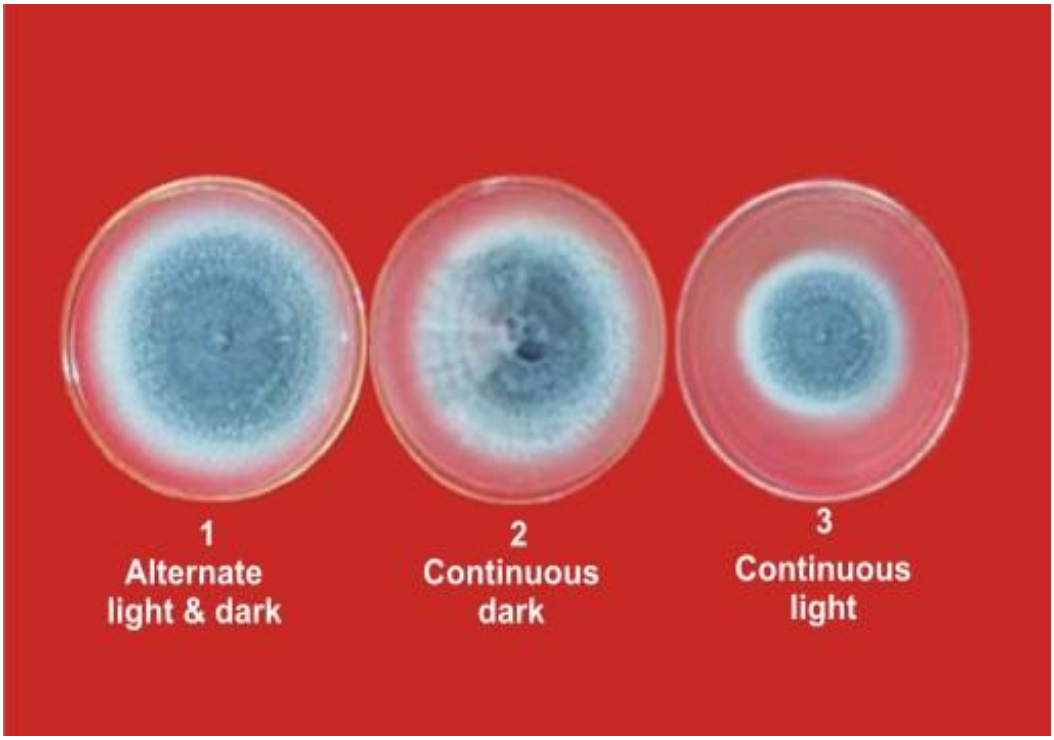


Plate 10: Effect of light on mycelial growth of C. truncatum

Table 17: Effect of environmental factors in relation to spore load of *Colletotrichum truncatum* and disease progression during *kharif* 2006 at ARS, Bidar

Std. week	Stage of crop (days)	Average weekly spore load	Weekly PDI	Temperature (^{0} c)		Relative humidity (%)		Total rainfall (mm)
				Maximum	Minimum	Morning	Evening	
24	6-12	0	0	34.51	23.60	77.43	37.14	4.4
25	13-19	4.70	9.26	32.49	22.37	89.57	55.86	20.2
26	20-26	14.32	13.58	31.26	21.91	92.86	58.00	18.4
27	27-33	20.43	21.65	30.23	21.66	90.71	62.43	8.7
28	34-40	18.50	29.32	31.46	21.94	86.86	51.71	1.5
29	41-47	15.21	38.02	32.34	21.86	88.14	49.00	8.8
30	48-54	17.10	50.11	28.74	21.14	94.86	71.00	83.4
31	55-61	22.14	62.21	26.56	20.57	96.00	79.14	173.2
32	62-68	20.75	68.50	29.03	20.69	95.86	63.71	17.9

Table 18: Effect of environmental factors in relation to spore load of *Colletotrichum truncatum* and disease progression during *kharif* 2007 at ARS, Bidar

Std. week	Stage of crop (days)	Average weekly spore load	Weekly PDI	Temperature (^{0} c)		Relative humidity (%)		Total rainfall (mm)
				Maximum	Minimum	Morning	Evening	
24	05-11	0.00	0.00	34.69	22.97	86.29	41.29	36.2
25	12-18	5.10	10.40	32.14	21.63	90.00	66.71	129.7
26	19-25	16.21	15.21	27.77	20.24	95.00	70.14	63.0
27	26-32	15.18	25.82	30.03	21.37	87.71	60.71	0.0
28	33-39	16.62	34.78	30.43	21.31	88.14	52.86	3.0
29	40-46	19.13	41.25	30.43	21.31	90.00	66.29	22.4
30	47-53	14.24	54.10	31.00	21.26	88.86	59.57	21.0
31	54-60	18.30	65.01	30.09	20.54	94.00	60.71	80.1
32	61-67	23.05	70.12	29.03	20.77	91.86	63.86	6.6

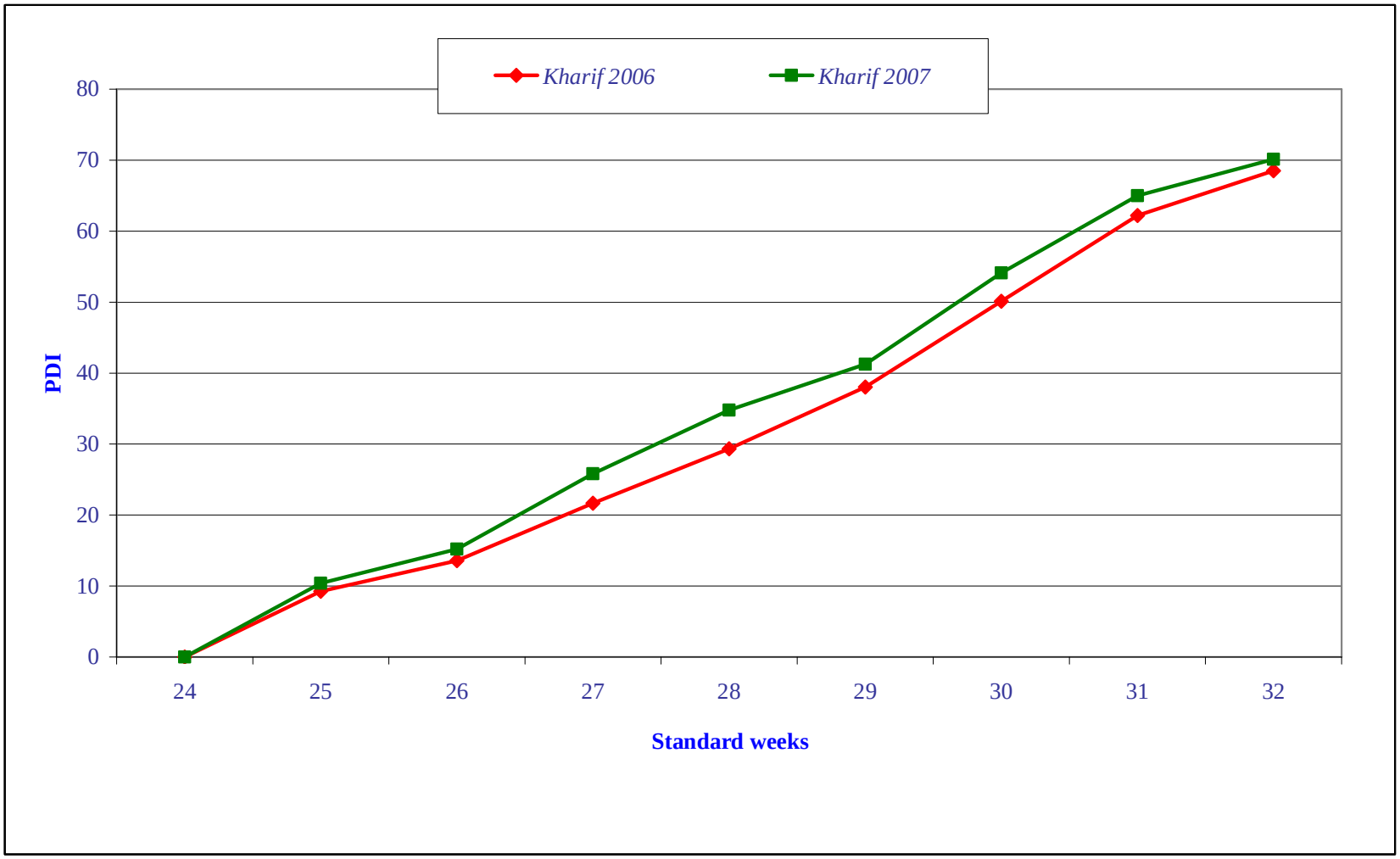


Fig. 12: Disease progress of anthracnose of greengram during kharif 2006 and kharif 2007

Plate 11a. Spore trap for exposure of stationary slide coated with Vaseline

Plate11b. Spore trap mounted in greengram experimental field for sampling of air borne conidia

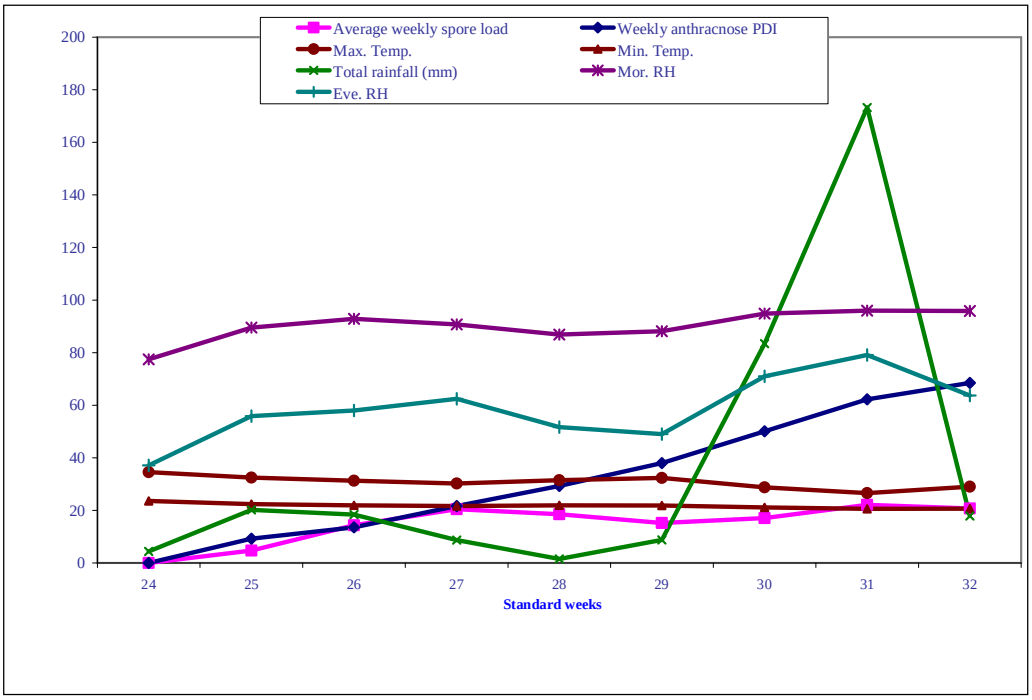


Fig. 13a: Development of spore load, PDI and meteorological parameters associated during kharif 2006

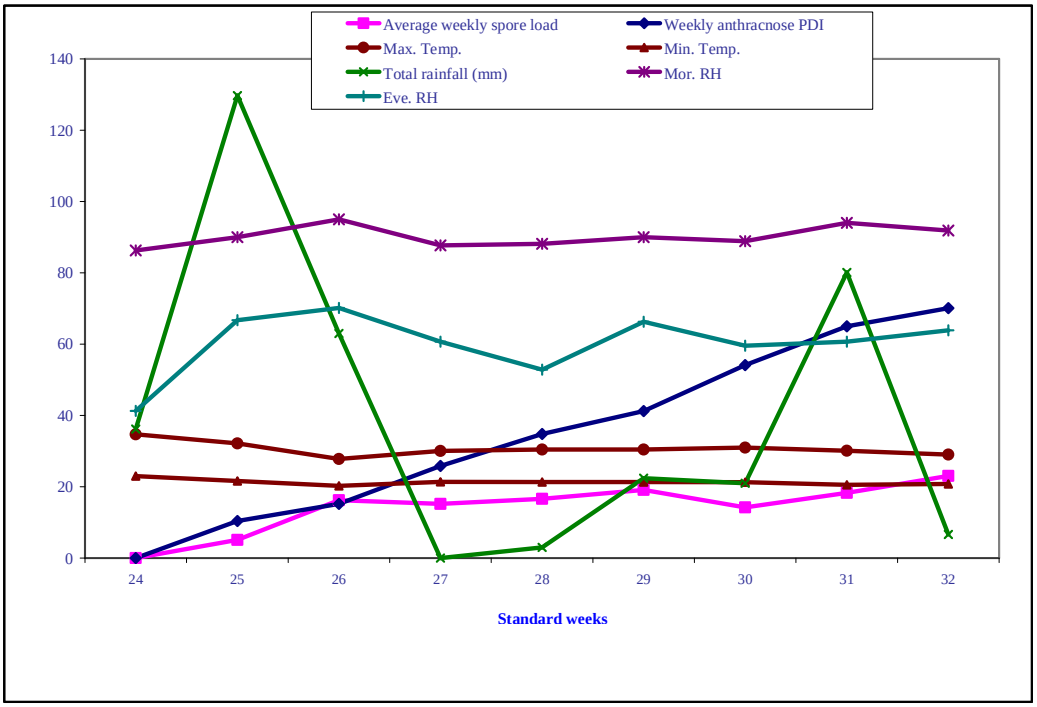


Fig. 13b: Development of spore load, PDI and meteorological parameters associated during kharif 2007

Table 19: Correlation between weekly spore load of *Colletotrichum truncatum* in relation to weather parameters

Sl. No.	Weather parameters	Correlation coefficient (r)		
		2006	2007	Pooled
1	Maximum temperature (ºc)	- 0.821**	- 0.851**	- 0.830**
2	Minimum temperature (ºc)	- 0.891**	- 0.812**	- 0.806**
3	Relative humidity (%) morning	0.762**	0.529	0.657**
4	Relative humidity (%) evening	0.728*	0.552*	0.645**
5	Rainfall (mm)	0.393	- 0.427	0.047

** significant at 1% probability
* significant at 5% probability

Table 20: Multiple linear regression of spore load of *Colletotrichum truncatum* causing anthracnose of greengram in relation to weather parameters

Year	Constant (a)	X_1	X_2	X_3	X_4	X_5	R^2
2006	344.10	- 2.42	- 8.32	- 0.83	0.07	- 0.07	0.90
2007	95.95	3.89	-12.39	0.52	0.36	- 0.15	0.93
Pooled	135.85	- 2.55	- 2.18	0.09	- 0.04	- 0.05	0.81

X_1 = Maximum temperature (ºc)
X_2 = Minimum temperature (ºc)
X_3 = Relative humidity (%) morning
X_4 = Relative humidity (%) evening
X_5 = Rainfall (mm)
R^2 = Coefficient of determination

Table 21: Multiple linear regression equation of spore load of *Colletotrichum truncatum* causing anthracnose of greengram in relation to weather parameters

Year	Multiple linear regression equation
2006	$Y=344.10 - 2.42X_1 - 8.32X_2 - 0.83X_3 + 0.07X_4 - 0.07X_5$
2007	$Y= 95.95 + 3.89X_1 - 12.39X_2 + 0.52X_3 + 0.36X_4 - 0.15X_5$
Pooled	$Y=135.85 - 2.55X_1 - 2.18X_2 + 0.09X_3 - 0.04X_4 - 0.05X_5$

X_1 = Maximum temperature (ºc)
X_2 = Minimum temperature (ºc)
X_3 = Relative humidity (%) morning
X_4 = Relative humidity (%) evening
X_5 = Rainfall (mm)

Whereas in 2007, the multiple linear regression equation was fitted as $Y = 95.95 + 3.89 X_1 – 12.39 X_2 + 0.52 X_3 + 0.36 X_4 – 0.15 X_5$

The pooled multiple linear regression equation for both the years was fitted as $Y = 135.85 – 2.55 X_1 – 2.18 X_2 + 0.09 X_3 – 0.04 X_4 – 0.05 X_5$ (Table 21).

Where, X_1 = maximum temperature (^{0}C), X_2 = minimum temperature (^{0}C), X_3 = Relative humidity (%) morning, X_4 = relative humidity (%), evening and X_5 = rainfall (mm).

This explains that, when there is increase of one unit of maximum and minimum temperature, evening relative humidity and rainfall, the spore load was decreased by 2.55, 2.18, 0.04 and 0.05, while it was increased by 0.09 units with respect to morning relative humidity. The weather factors influenced the spore load of *C. truncatum* to the extent of 81.00 per cent.

4.6.1.2 Effect of weather parameters on the development and spread of anthracnose of greengram

The knowledge of weather conditions predisposing for development and spread of the disease is important to organize Agro Advisory Services (AAS) for the farmers to take up timely management practices (Table 17 and 18).

During 2006, the anthracnose symptoms were first observed on 25th SW, when the crop was at 19 DAS. The severity increased slowly and reached as high as 68.50 per cent during 32nd SW. During the previous week, maximum temperature of 26.56^{0}C, minimum temperature of 20.57^{0}C, morning relative humidity of 96.00 per cent, evening relative humidity of 79.14 per cent and 173.2 mm rainfall were observed and depicted in Fig. 13a.

During 2007, the anthracnose symptoms were first observed on 25th SW when crop was at 18 DAS. The severity was upto 70.12 per cent at 32nd SW. The rainfall of 80.1 mm with minimum temperature of 20.54^{0}C and maximum temperature of 30.09^{0}C, morning relative humidity of 94.00 per cent and evening relative humidity of 60.71 per cent prevailed during previous week (Fig. 13b).

Table 22: Correlation between percent disease index (PDI) of anthracnose of greengram in relation to weather parameters

Sl. No.	Weather parameters	Correlation coefficient (r)		
		2006	2007	Pooled
1	Maximum temperature (ºc)	- 0.837**	- 0.479	- 0.669*
2	Minimum temperature (ºc)	- 0.909**	- 0.575*	- 0.734**
3	Relative humidity (%) morning	0.741**	0.376	0.582*
4	Relative humidity (%) evening	0.721*	0.276	0.528
5	Rainfall (mm)	0.575*	- 0.305	0.188

** significant at 1% probability
* significant at 5% probability

The data on both the years indicated that 25th and 26th standard weeks were highly favourable for the first appearance of the disease. The weather factors played an important role in the initiation and further spread of disease. It gives information to design supervisory control measures of disease in order to get expected yield.

4.6.1.3 Correlation and multiple linear regression analysis between severity of anthracnose in relation to weather parameters

The analysis was made to establish the relationship between weather factors *viz.*, maximum and minimum temperatures, morning and evening relative humidity and rainfall with per cent disease index of disease in highly susceptible variety 'Chinamung' through correlation and multiple linear regression analysis.

The relationship between PDI and weather factors during 2006 (Table 22) indicated significant negative correlation between maximum and minimum temperature with a correlation coefficient of -0.837 and -0.909, respectively. While, relative humidity morning (0.741), relative humidity evening (0.721) and rainfall (0.575) were positively correlated with PDI.

Similarly during 2007, PDI was negatively correlated with temperature and positively with relative humidity. However, the pooled analysis of both the years revealed significant negative correlation with maximum temperature (-0.669) and minimum temperature (-0.734), while it was positively correlated with relative humidity morning (0.582), relative humidity evening (0.528) and rainfall (0.188).

The multiple linear regression of PDI in relation to weather parameters during 2006 (Table 23) indicated that the regression coefficients for maximum temperature, minimum temperature, morning relative humidity, evening relative humidity and rainfall were found to be 16.37, -75.27, -2.72, 0.92 and 0.09, respectively during 2006 and in the same way, 34.81, -21.93, 15.15, 0.02 and -0.90, respectively during 2007. The pooled regression coefficients for both the years showed regression coefficients for weather parameters as maximum temperature (-2.42), minimum temperature (-22.06), morning relative humidity (0.69), evening relative humidity (1.01) and rainfall (0.01).

Table 23: Multiple linear regression of percent disease index of anthracnose of greengram caused by *Colletotrichum truncatum* in relation to weather parameters

Year	Constant (a)	X_1	X_2	X_3	X_4	X_5	R^2
2006	1349.88	16.37	- 75.27	-2.72	0.92	0.09	0.98
2007	-1885.52	34.81	- 21.93	15.15	0.02	- 0.90	0.89
Pooled	579.69	- 2.42	- 22.06	0.69	1.01	0.01	0.57

X_1 = Maximum temperature (ºc)
X_2 = Minimum temperature (ºc)
X_3 = Relative humidity (%) morning
X_4 = Relative humidity (%) evening
X_5 = Rainfall (mm)
R^2 = Coefficient of determination

During 2006, the multiple linear regression equation was fitted to the data and the equation arrived for the weather parameters was $Y = 1349.88 + 16.37 X_1 – 75.27 X_2 – 2.72 X_3 + 0.92 X_4 + 0.09 X_5$, whereas during 2007, the multiple linear regression equation was fitted as $Y = -1885.52 + 34.81 X_1 – 21.93 X_2 + 15.15 X_3 + 0.02 X_4 – 0.90 X_5$. The pooled multiple linear regression equation for both the years was fitted as $Y = 579.69 – 2.42 X_1 – 22.06 X_2 + 0.69 X_3 + 1.01 X_4 + 0.01 X_5$ (Table 24). Where, X_1 = maximum temperature (^{0}C), X_2 = minimum temperature (^{0}C),X_3 = relative humidity (%) morning, X_4 = relative humidity (%) evening and X_5 = rainfall (mm).

This revealed that when there was increase in one unit of maximum and minimum temperature, the per cent disease index decreased by 2.42 and 22.06 units, respectively, while when there was increase in one unit of morning and evening relative humidity and rainfall, the per cent disease index increased by 0.69, 1.01 and 0.01 units, respectively. The weather factors influenced the disease incidence in Chinamung to the extent of 57.00 per cent.

4.6.1.4 Disease prediction models

Weather factors play an important role in disease development, when the vulnerable host and virulent pathogen coincide in a situation. An attempt was made to predict the severity of anthracnose using 2nd degree polynomial equation model.

The disease severity was recorded at weekly interval on susceptible variety Chinamung during 2006 and 2007 (Table 17 and 18). Since, the disease progress curve behaved in a same manner during both *kharif* 2006 and *kharif* 2007 (Fig. 12), the 2nd degree polynomial function method was used to develop disease prediction models. The values were calculated and the models were developed to predict the severity of anthracnose. Observed and predicted disease severity of anthracnose of greengram during *kharif* 2006 and *kharif* 2007 is given in Table 25 and Fig. 14 and 15.

Table 24: Multiple linear regression equation of percent disease index of anthracnose of greengram in relation to weather parameters

Year	Multiple linear regression equation
2006	$Y = 1349.88 + 16.37X_1 - 75.27X_2 - 2.72X_3 + 0.92X_4 + 0.09X_5$
2007	$Y = -1885.52 + 34.81X_1 - 21.93X_2 + 15.15X_3 + 0.02X_4 – 0.90X_5$
Pooled	$Y = 579.69 - 2.42X_1 - 22.06X_2 + 0.69X_3 + 1.01X_4 + 0.01X_5$

X_1 = Maximum temperature (ºc)
X_2 = Minimum temperature (ºc)
X_3 = Relative humidity (%) morning
X_4 = Relative humidity (%) evening
X_5 = Rainfall (mm)

Kharif 2006

$Y = -4.90 + 5.37 X + 0.33 X^2$ with $R^2 = 0.99$

Kharif 2007

$Y = -7.60 + 7.87 X + 0.11 X^2$ with $R^2 = 0.99$

The models had highest coefficient of determination values with 99 per cent for *kharif* 2006 and *kharif* 2007.

4.6.2 Effect of date of sowing on the incidence of anthracnose of greengram

A field experiment was conducted during *kharif* season of 2006 and 2007 with six different sowing dates starting from 4th June to 9th July at weekly intervals at Agricultural Research Station, Bidar to know the effect of different dates of sowing and the meteorological conditions associated with the disease development (Plate 12). The information on the incidence of disease as affected by different dates of sowing and also to know the influence of meteorological conditions in disease development will be very much useful to adjust the sowing times for growing good crop under very low disease pressure. The results are presented in Table 26 and depicted in Fig. 16.

During 2006, there was a significant difference in the severity of anthracnose at different sowing dates. The per cent disease index varied from 24.29 to 59.87 per cent. The least PDI was recorded on crop sown on 4th June (24.29%) and was found on par with PDI of the crop sown on 11th June (26.10%), while the highest PDI was recorded on crop sown on 9th July (59.87%).

Similar results were obtained during 2007 also. The least severity of 25.77 per cent was recorded on crop sown on 4th June, while maximum PDI on crop sown on 9th July (61.24%). The mean PDI ranged from 25.03 to 60.56 per cent. The mean data of two years indicated the same trend as observed in individual years with respect to per cent disease index of anthracnose.

The grain yield in both the years indicated that higher yields (9.61 q/ha and 9.18 q/ha) were obtained in 4th June sowing of 2006 and 2007, respectively. The mean yield data

Table 25: Observed and predicted PDI of anthracnose of greengram caused by *Colletotrichum truncatum* during the progression of disease for *kharif* 2006 and 2007

Time interval (week)	Percent Disease Index					
	Kharif 2006			*Kharif* 2007		
	Observed	Predicted	Deviation	Observed	Predicted	Deviation
1	9.26	7.05	2.21	10.40	6.56	3.84
2	13.58	15.40	- 1.82	15.21	20.03	- 4.82
3	21.65	26.38	- 4.73	25.82	21.23	4.59
4	29.32	25.37	3.95	34.78	40.09	- 5.31
5	38.02	40.50	- 2.48	41.25	41.03	0.22
6	50.11	46.64	3.47	54.10	56.82	- 2.72
7	62.21	64.54	- 2.33	65.01	64.39	0.62
8	68.50	66.77	1.73	70.12	66.54	3.58

revealed that highest mean yield of 9.40 q per ha was obtained in crop sown on 4th June followed by 11th June (8.83 q/ha).

In the present study, severity of anthracnose of greengram was found to be influenced by environmental factors, which prevailed during crop growth period. Table 27 indicated that in both the years, the crop sown during 4th and 11th June recorded a lower per cent disease index coupled with higher yield. This could be due to higher temperature of 31^{0}C during the crop growth period coupled with lower humidity (88 – 89%) which were less congenial to the disease development. Whereas, the disease severity was maximum at the end of June month onwards. During that period, the weather conditions were very much congenial i.e., moderate temperature of 29^{0}C coupled with higher humidity of 90 to 92 per cent, with respect to rainfall received with range of 3.10 to 5.91 mm, though the amount received was less compared to early sowings but there was frequent rains received during crop growth period. The disease coincidence of the favourable period with stage of the crop led to considerable increase in disease severity.

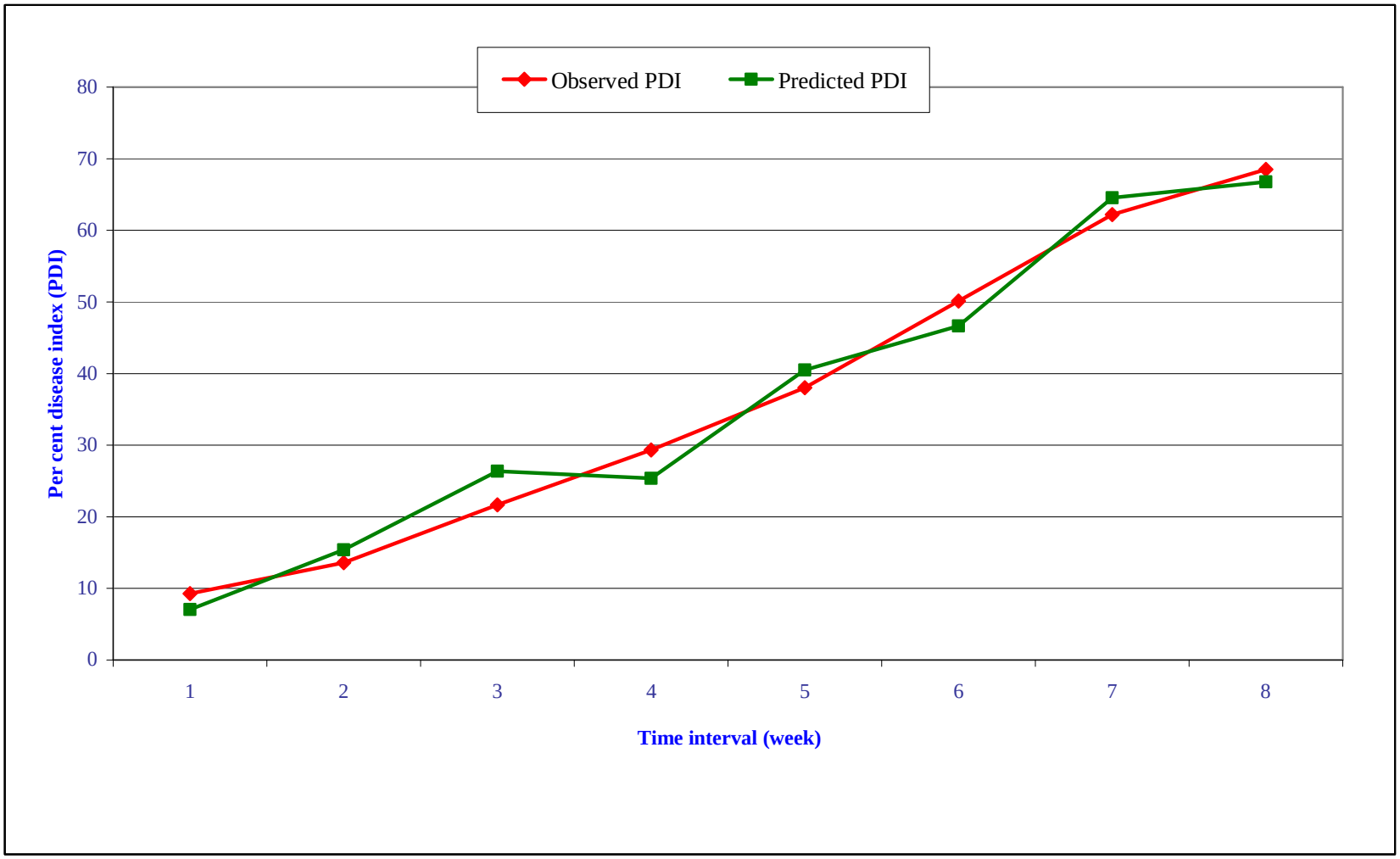


Fig. 14: Observed and predicted PDI values during 2006

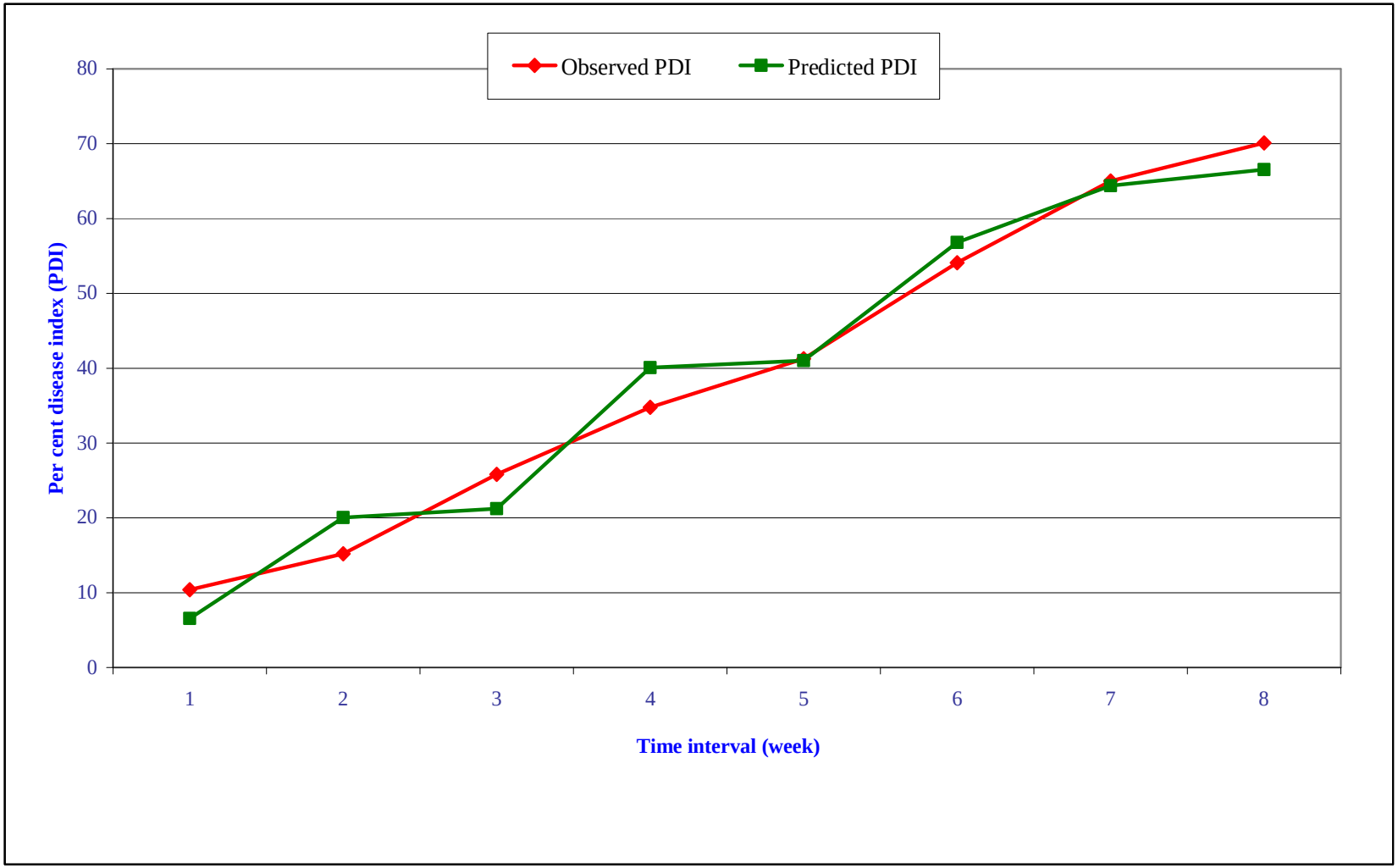


Fig. 15: Observed and predicted PDI values during 2007

Table 26: Effect of date of sowing on percent disease index of anthracnose and yield of greengram during *kharif* 2006 and 2007

Sl. No.	Sowing date	Percent disease index at 40 DAS			Grain yield (q/ha)		
		K-06	K-07	Mean	K-06	K-07	Mean
1	4th June	24.29 (29.52)*	25.77 (30.50)	25.03	9.61	9.18	9.40
2	11th June	26.10 (30.72)	27.67 (31.73)	26.89	9.06	8.60	8.83
3	18th June	28.78 (32.44)	29.65 (32.99)	29.22	7.87	7.34	7.61
4	25th June	32.91 (35.00)	35.07 (36.31)	33.99	6.91	6.59	6.75
5	2nd July	52.17 (46.24)	55.45 (48.13)	53.81	5.97	5.43	5.70
6	9th July	59.87 (50.69)	61.24 (51.50)	60.56	5.34	5.02	5.18
	S.Em±	0.68	0.70		0.19	0.20	
	CD at 5%	2.07	2.10		0.56	0.60	

*values in parenthesis are arcsine transformed values

Correlation coefficients between disease severity and weather parameters during both the years (pooled) revealed that maximum and minimum temperature have significantly negative correlation with disease. However, correlation coefficient with relative humidity and rainfall were positive but non-significant (Table 28).

4.6.3 Survival of *C. truncatum* in debris and seed

4.6.3.1 Survival of conidia of *C. truncatum* under different storage conditions

Viability of conidia of *C. truncatum* in different storage conditions was studied at storage periods from 15 to 360 days at an interval of 15 days and the results obtained are shown in Table 29.

The results revealed that, per cent viability of conidia decreased with increase in storage period in all conditions tested. The conidia remained viable for 120 days when kept under glasshouse conditions, whereas they remained viable for 210 days at room conditions. Similarly, the viability of conidia remained for 240 days under tree shade condition. Maximum

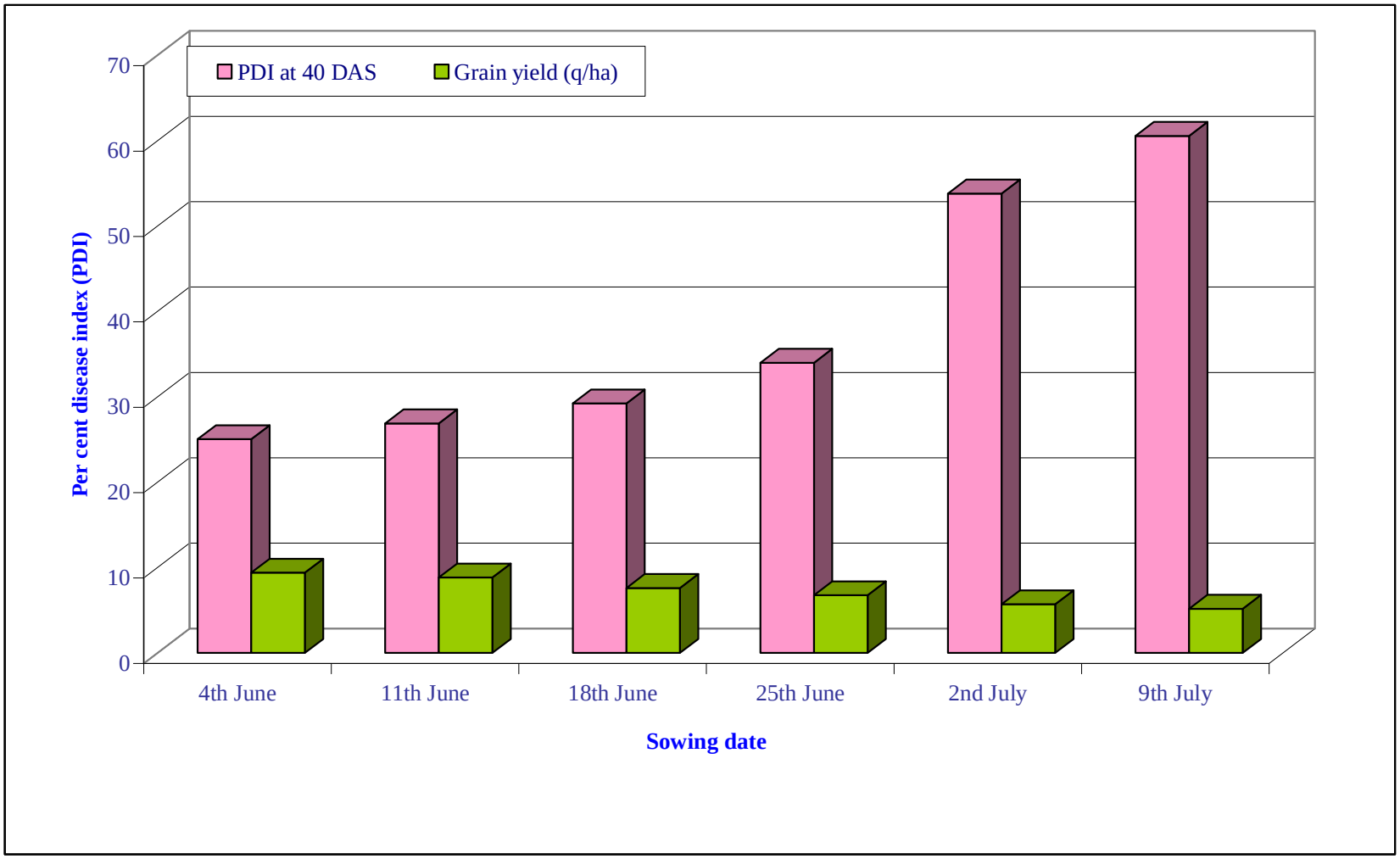


Fig. 16: Effect of date of sowing on per cent disease index of anthracnose and grain yield of greengram during kharif 2006 and 2007

Table 27: Effect of dates of sowing and environmental factors in relation to anthracnose of greengram caused by *Colletotrichum truncatum* during *kharif* 2006 and 2007

Sl. No	Date of sowing	PDI at 40 DAS	Temperature (^{0} c)		Relative humidity (%)		Total rainfall (mm)
			Maximum	Minimum	Morning	Evening	
Kharif 2006							
1	4th June	24.29	31.80	22.10	88.16	53.23	155.12
2	11th June	26.10	30.95	21.88	89.55	58.04	318.64
3	18th June	28.78	30.26	21.52	91.86	61.36	332.08
4	25th June	32.91	29.93	21.35	92.47	62.39	330.96
5	2nd July	52.17	29.87	21.27	92.25	61.95	313.04
6	9th July	59.87	29.96	21.16	92.05	60.73	309.12
Kharif 2007							
1	4th June	25.77	31.73	21.47	89.02	57.39	356.16
2	11th June	27.67	30.95	21.33	90.00	59.79	355.60
3	18th June	29.65	30.24	21.05	90.70	62.61	325.92
4	25th June	35.07	29.91	21.03	90.57	61.46	202.72
5	2nd July	55.45	29.25	21.09	90.29	60.05	173.60
6	9th July	61.24	29.22	21.07	91.11	60.86	202.72

Table 28: Correlation studies of weather parameters with anthracnose severity on greengram

Sl. No	Weather parameters	Correlation coefficient (r)		
		2006	2007	Pooled
1	Maximum temperature (^{0}C)	- 0.680*	- 0.869**	- 0.782**
2	Minimum temperature (^{0}C)	- 0.823**	- 0.551*	- 0.600*
3	Relative humidity (%) morning	0.619*	0.605*	0.517
4	Relative humidity (%) evening	0.516	0.196	0.389
5	Rainfall (mm)	0.331	- 0.838**	0.329

** significant at 1% probability

* significant at 5% probability

Experimental field View

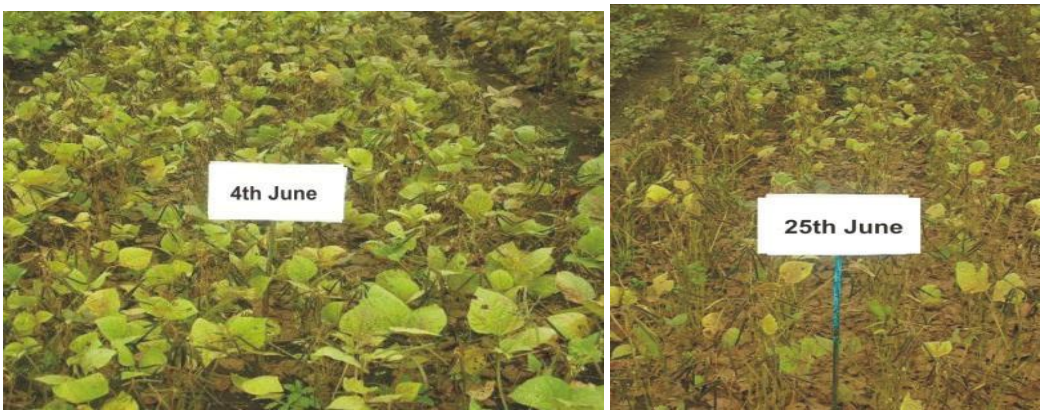


Plate 12: Effect of Date of sowing on the severity of anthracnose

period of viability of spores remained upto 360 days under freeze conditions. The lowest period of viability of conidia remained for 90 days was under field conditions.

4.6.3.2 Survival in infected seeds

Studies on survival of *C. truncatum* and germination percentage in greengram seeds was undertaken for a period of 360 days at an interval of one month and the results are presented in Table 30.

The data revealed that there was a sharp decline in survivability of the fungus over the period of storage, however the germination percentage increased with the time, further it was observed that initially *C. truncatum* fungus recorded 23.50 per cent survival in seed at 30 days. Later there was a gradual decrease in the survivability of fungus. The fungus remained viable at low percentage (7.25) upto 360 days. The germination percentage gradually increased with the increase in the storage period and reached upto 87.00 per cent after 360 days of storage.

4.6.4 Effect of plant age in relation to infection of *C. truncatum*

Age of the plant is important for the development of disease. A study was conducted to find out the susceptible stage of the greengram crop for maximum anthracnose infection (Table 31 and Fig. 17).

The data presented in Table 31 revealed that the greengram plants when inoculated at different plant age *i.e.,* 5, 10, 15, 20, 25, 30, 35, 40, 45 and 50 days showed *C. truncatum* infection. The per cent disease severity on five days old seedlings was 3.40. It gradually increased at later stage. The maximum disease severity of 68.92 per cent was observed on 40 days old plants, followed by 35 days (62.05%) and 45 days (52.20%), further the disease severity decreased to 22.87 per cent on 50 days old plants.

4.6.5 Host range studies

The possibility of existence of alternative hosts was studied. Eight different pulses as mentioned in the “Material and Methods” were examined for their reaction to *C. truncatum* under artificial inoculated conditions.

The results of the disease reaction on different pulses are presented in Table 32. It was confirmed that among the eight pulse crops tested, anthracnose symptom developed on only three hosts *viz.*, blackgram (*Vigna mungo*), horsegram (*Macrotyloma uniflorum*) and greengram (*Vigna radiata*).

Infected leaves showed the symptoms of dark brown lesions which gave blighted appearance to the seedlings. As the disease progressed such lesions coalesced turned necrotic. Soybean (*Glycine max*), cowpea (*Vigna unguiculata*), redgram (*Cajanus cajan*), bengalgram (*Cicer arietinum*), frenchbean (*Phaseolus vulgaris*) and clusterbean (*Cyamopsis tetragonoloba*) did not show any symptom of anthracnose.

4.6.6 Cross inoculation studies

The cross inoculation studies were conducted to know the cross infectivity of anthracnose pathogen from horsegram and blackgram on greengram and vice-versa as mentioned in the “Material and Methods”. In cross inoculation study, the conidial suspension of *C. truncatum* from greengram infected blackgram, greengram and horsegram. Similarly, conidial suspension prepared from isolates of blackgram and horsegram infected greengram (Table 33 and Plate 13).

4.7 Screening of genotypes against anthracnose and biochemical factors of resistance

4.7.1 Screening of greengram genotypes under artificial inoculated conditions against anthracnose

Thirty greengram genotypes were evaluated under greenhouse artificial inoculation with *C. truncatum* fungus, to find out the source of host resistance if any during 2006-07. The results are presented in Table 34.

Table 29: Studies on survival of conidia of *Colletotrichum truncatum* causing anthracnose of greengram under different conditions

Sl. No	Storage period (Days)	Percent viable conidia at different storage conditions				
		Freeze (4-5^{0}C)	Tree shade (18-22^{0}C)	Room/Lab (20-25^{0}C)	Glass house (25-28^{0}C)	Field (28-30^{0}C)
1.	15	87.16	82.15	80.18	76.73	66.17
2.	30	85.75	76.52	77.26	68.15	57.23
3.	45	83.23	70.23	73.10	61.24	42.30
4.	60	80.41	67.88	69.55	55.18	30.75
5.	75	77.50	63.40	65.33	46.29	15.20
6.	90	73.62	54.73	56.81	23.11	2.86
7.	105	70.78	48.25	50.42	10.35	0
8.	120	67.45	40.08	42.36	2.17	0
9.	135	63.81	35.66	36.91	0	0
10.	150	60.11	30.19	29.24	0	0
11.	165	57.75	25.43	21.66	0	0
12.	180	54.23	20.78	15.38	0	0
13.	195	51.39	17.19	10.75	0	0
14.	210	48.56	13.86	3.28	0	0
15.	225	45.90	8.10	0	0	0
16.	240	41.27	5.27	0	0	0
17.	255	38.15	0	0	0	0
18.	270	35.78	0	0	0	0
19.	285	34.50	0	0	0	0
20.	300	30.15	0	0	0	0
21.	315	29.00	0	0	0	0
22.	330	25.22	0	0	0	0
23.	345	20.19	0	0	0	0
24.	360	5.23	0	0	0	0

It was revealed that among the thirty genotypes evaluated, none of the genotypes were found to be immune. Two genotypes *viz.*, TM-96-2 and TARM-18 have shown resistant reaction, three genotypes *viz.*, BGS-9, TM-98-50 and TM-97-55 showed moderately resistant reaction. Two genotypes *viz.*, Pusa baisaki and Sel-4 were found susceptible and remaining 23 genotypes *viz.*, Chinamung, Vaibhav, KG-06-1, Yellowmung, DGGV-04, DGS-052, BPMR-1, DLGG-22, DGGS-16, BPMR-145, HMV-6, ML-5, HVM-1, PD-51, OBGG-52, ML-131, KM-6-146, DGS-051, KM-5-133, DGGS-27, KM-5-141, KM-5-134 and KM-6-210 showed highly susceptible reaction to *C. truncatum* (Plate 14).

Table 30: Studies on survival of *Colletotrichum truncatum* in greengram seeds and its effect on germination

Sl. No.	Storage period (in days)	Percent Survival of fungus	Percent germination of seed
1.	30	23.50	54.00
2.	60	22.15	55.00
3.	90	22.00	57.00
4.	120	24.00	57.00
5.	150	18.75	60.00
6.	180	17.10	63.00
7.	210	14.00	66.00
8.	240	13.50	69.00
9.	270	13.00	74.00
10.	300	10.75	76.00
11.	330	9.00	81.00
12.	360	7.25	87.00

Table 31: Effect of age of plant in relation to anthracnose severity in greengram

Sl. No.	Age of the plant (days)	Percent disease index (PDI)
1	5	3.40 (10.62)*
2	10	6.16 (14.37)
3	15	9.11 (17.56)
4	20	16.26 (23.78)
5	25	21.77 (27.81)
6	30	35.12 (36.34)
7	35	62.05 (51.98)
8	40	68.92 (56.12)
9	45	52.20 (46.26)
10	50	22.87 (28.57)
S.Em±		0.56
CD at 1%		2.29

*Values in parenthesis are arc-sine transformed values

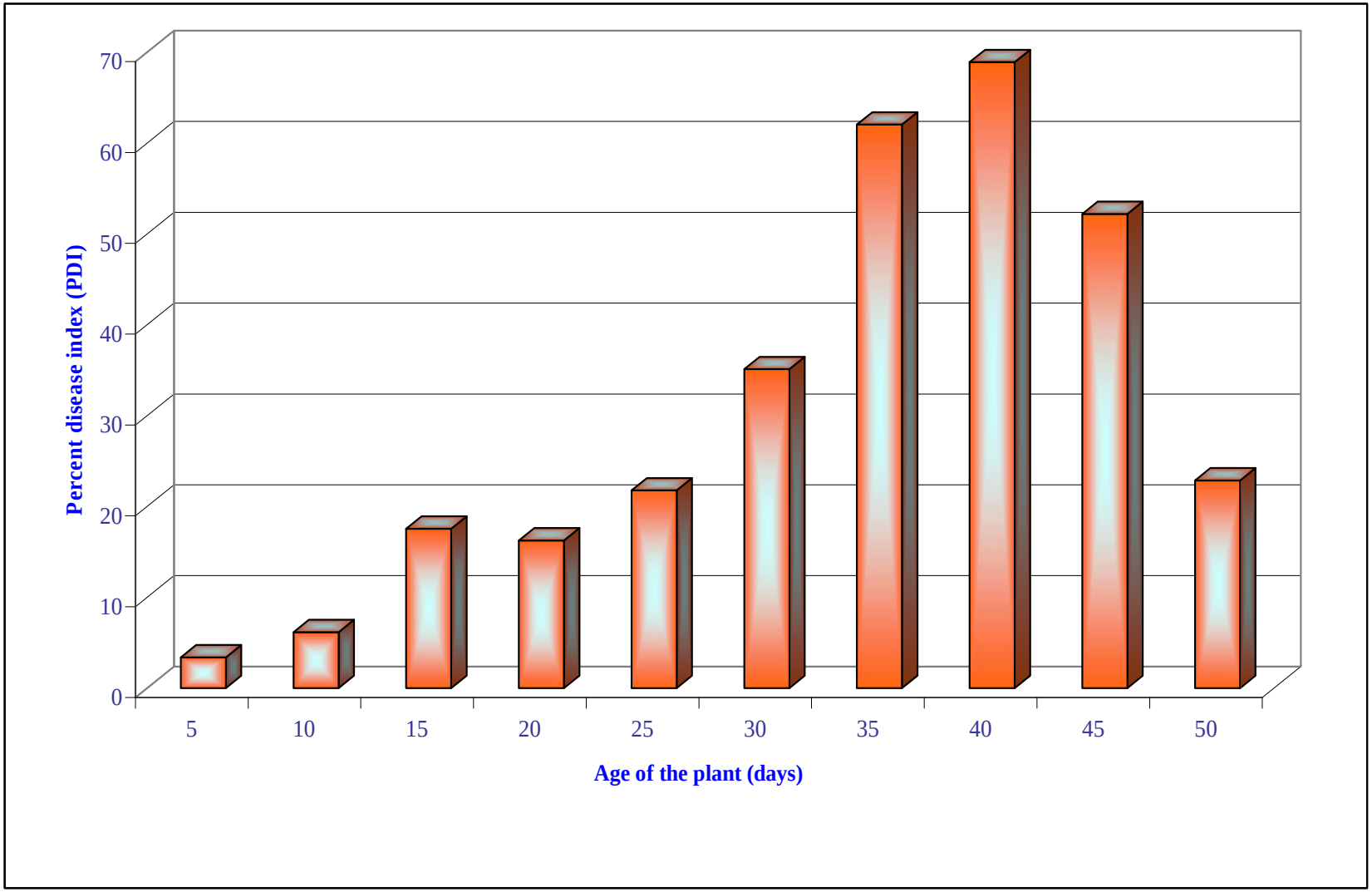


Fig. 17: Effect of the plant age in relation to anthracnose severity in greengram

Table 32: Reaction of different pulses to greengram isolate of *Colletotrichum truncatum*

Sl. No.	Host plant	Botanical name	Reaction
1.	Blackgram	*Vigna mungo*	+
2.	Soybean	*Glycine max*	-
3.	Horse gram	*Macrotyloma uniflorum*	+
4.	Cow pea	*Vigna unguiculata*	-
5.	Redgram	*Cajanus Cajun*	-
6.	Bengalgram	*Cicer arietinum*	-
7.	French bean	*Phaseolus vulgaris*	-
8.	Cluster bean	*Cyamopsis tetragonoloba*	-
9.	Greengram	*Vigna radiata*	+

+ = Infection observed

- = No infection

4.7.2 Field evaluation of promising greengram varieties for disease resistance

To identify the sources of resistance, six promising varieties were screened against anthracnose with 0 to 9 scale under field conditions at ARS, Bidar during *kharif* 2006 and 2007 as explained in Material and Methods. The data on reaction of each variety is presented in Table 35.

During 2006, the cultivars *viz.*, TM-96-2 and TARM-18 were found resistant and BGS-9 showed moderately resistant reaction while Sel-4 found susceptible and Yellowmung and Chinamung varieties were found highly susceptible. During 2007, TARM-18 alone was found resistant and cultivars *viz.*, TM-96-2 and BGS-9 were found moderately resistant while others showed highly susceptible reaction (Plate 15).

4.7.3 Biochemical factors of resistance to anthracnose of greengram

The investigations on biochemical factors of resistance against *C. truncatum* were carried out and results are presented in Table 36a, 36b and 36c and depicted in Fig. 18.

4.7.3.1 Chlorophyll content

Destruction of chloroplast is a common feature of diseased tissues. Disease may reduce photosynthetic rate, phosphorylation and CO_2 assimilation. These changes may be partially or completely accounted by reduction in chlorophyll content. Hence, rate of chlorophyll loss in the greengram crop as influenced by the development of anthracnose was studied. The chlorophyll 'a', chlorophyll 'b' and total chlorophyll content of different genotypes are presented in Table 36a.

Table 33: Cross inoculation study of *Colletotrichum truncatum* – a causal agent of anthracnose of greengram

Sl. No	Hosts	*Vigna radiata*	*Vigna mungo*	*Macrotyloma uniflorum*
1.	*Vigna radiata*	+	+	+
2.	*Vigna mungo*	+	+	+
3.	*Macrotyloma uniflorum*	+	+	+

+ = Infection observed

Infection on Vigna radiate

Infection on Vigna mungo

Infection on Macrotyloma uniflorum

Plate 13: Cross inoculation study of C. truncatum

Table 34: Reaction of greengram genotypes against anthracnose under artificial inoculated condition

Sl. No	Grade	Disease reaction	Name of the genotypes
1.	0	Immune	Nil
2.	1	Resistant	TM-96-2, TARM-18
3.	3	Moderately resistant	BGS-9, TM-98-50, TM-97-55
4.	5	Moderately susceptible	Nil
5.	7	Susceptible	Pusa baisaki, Sel-4
6.	9	Highly susceptible	China mung, Vaibhav, KG-06-1, Yellow mung, DGGV-04, DGS-052, BPMR-1, DLGG-22, DGGS-16, BPMR-145, HMV-6, ML-5, HVM-1, PD-51, OBGG-52, ML-131, KM-6-146, DGS-051,KM-5-133,DGGS-27,KM-5-141,KM-5-134,KM-6-210

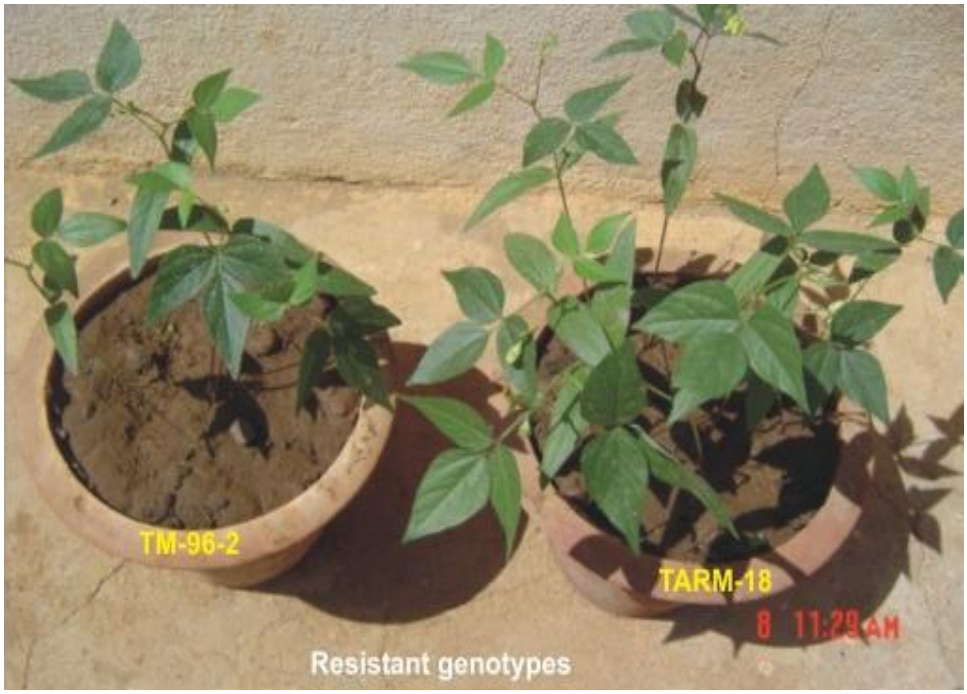


Plate 14: Screening of greengram genotypes under artificial inoculated condition against C. truncatum

Table 35: Field evaluation of promising greengram entries for disease resistance

Sl. No	Name of the genotype	Maximum disease grade	
		2006	2007
1.	Sel-4	7	9
2.	BGS-9	3	3
3.	Yellow mung	9	9
4.	China mung	9	9
5.	TM-96-2	1	3
6.	TARM-18	1	1

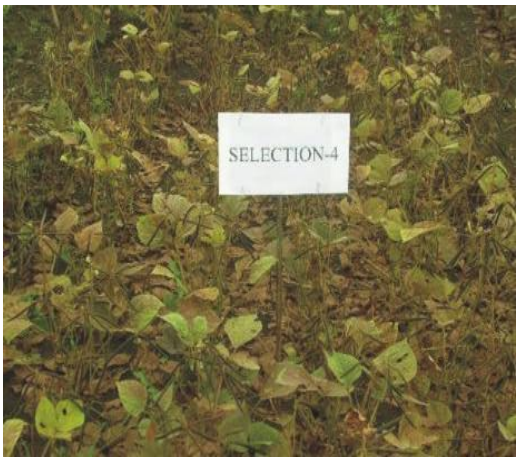
The resistant and moderately resistant varieties recorded higher amounts of chlorophyll ‘a’, ‘b’ and total chlorophyll than susceptible varieties in both healthy and diseased leaves. In all the varieties, there was decrease in chlorophyll ‘a’, chlorophyll ‘b’ and total chlorophyll content in diseased leaves. The least decrease per cent in chlorophyll ‘a’ was 4.25 per cent in TM-96-2 and TARM-18 (both resistant), followed by 4.41 per cent in BGS-9 and TM-97-55 (both moderately resistant), whereas highest decrease of 58.11 per cent in Sel-4, Chinamung and Yellowmung (all susceptible). But, chlorophyll ‘b’ was found to be more sensitive which was also reduced at higher rates in susceptible varieties to 74.93 per cent.

Total chlorophyll was also in the same trend, as it was cumulative effect of chlorophyll ‘a’ and ‘b’. Its reduction was more in susceptible varieties (64.24%) than resistant (14.88%) and moderately resistant (16.28%) varieties.

4.7.3.2 Sugar content

The present study revealed the variations in sugar content between healthy and infected greengram leaves. This study will help to understand some aspects of biochemical defense mechanisms, operating in the host.

The Table 36b indicated that total sugars, reducing sugar and non-reducing sugar contents were higher in healthy leaves of susceptible genotypes than of resistant ones. In diseased leaves their amount decreased in both resistant and susceptible genotypes. But, this decrease in sugar content was at higher rates in susceptible varieties compared to resistant and moderately resistant varieties.



Section – 4

Chinamung

Yellow mung

BGS – 9

TARM – 18

TM – 96 -2

Plate 15: Field evaluation of promising green gram varieties for disease resistance

Table 36a: Effect of anthracnose on chlorophyll content (mg/g of fresh weight) in varieties of green gram with different levels of resistance

Chlorophyll	Type of leaves	Resistant		Mean	Per cent decrease over healthy	Moderately resistant		Mean	Per cent decrease over healthy	Susceptible/ Highly susceptible			Mean	Per cent decrease over healthy
		TM-96-2	TARM-18			BGS-9	TM-97-55			Sel-4	China mung	Yellow mung		
a. Chlorophyll 'a'	Healthy	0.813	1.306	1.060	4.25	1.102	0.850	0.976	4.41	0.653	0.701	0.645	0.666	58.11
	Diseased	0.780	1.250	1.015		1.060	0.806	0.933		0.262	0.310	0.265	0.279	
b. Chlorophyll 'b'	Healthy	0.601	0.733	0.667	31.78	0.495	0.512	0.504	39.29	0.380	0.422	0.337	0.379	74.93
	Diseased	0.503	0.407	0.455		0.311	0.301	0.306		0.077	0.119	0.088	0.095	
c. Total Chlorophyll	Healthy	1.414	2.039	1.727	14.88	1.597	1.362	1.480	16.28	1.033	1.123	0.982	1.046	64.24
	Diseased	1.283	1.657	1.470		1.371	1.107	1.239		0.339	0.429	0.353	0.374	

Table 36b: Effect of anthracnose on sugar content (mg/g of fresh weight) in varieties of greengram with different levels of resistance

Sugars	Type of leaves	Resistant		Mean	Per cent decrease over healthy	Moderately resistant		Mean	Per cent decrease over healthy	Susceptible/ Highly susceptible			Mean	Per cent decrease over healthy
		TM-96-2	TARM-18			BGS-9	TM-97-55			Sel-4	China mung	Yellow mung		
a. Reducing sugar	Healthy	9.32	9.15	9.24	8.01	8.03	9.29	8.66	8.78	9.51	9.85	9.74	9.70	9.07
	Diseased	8.10	8.90	8.50		7.12	8.68	7.90		8.74	8.75	8.97	8.82	
b. Non reducing sugar	Healthy	5.79	6.09	5.94	5.40	6.11	6.15	6.13	5.38	7.01	6.31	6.87	6.73	5.49
	Diseased	5.31	5.93	5.62		5.52	6.08	5.80		6.68	6.01	6.40	6.36	
c. Total sugar	Healthy	15.11	15.24	15.18	6.98	14.14	15.44	14.79	7.36	16.52	16.16	16.61	16.43	7.61
	Diseased	13.41	14.83	14.12		12.64	14.76	13.70		15.42	14.76	15.37	15.18	

The least rate of decrease in reducing sugar content in diseased leaves was 8.01 per cent in TM-96-2 and TARM-18 (both resistant) followed by 8.78 per cent in BGS-9 and TM-97-55 (both moderately resistant), whereas highest per cent decrease of 9.07 in susceptible varieties. Similarly, lowest per cent decrease in non-reducing sugar content (5.40%) was observed in resistant varieties followed by 5.38 per cent in moderately resistant varieties, while highest (5.49%) in susceptible varieties.

In all, the decrease in total sugar content was 6.98 and 7.36 per cent in resistant and moderately resistant varieties, respectively whereas highest per cent decrease of 7.61 was found in susceptible varieties.

4.7.3.3 Phenols

Phenols also play an important role in imparting disease resistance. In the present investigations, both total phenol and ortho-dihydroxy phenols were estimated. The results are presented in Table 36c.

The study indicated that healthy leaves of resistant genotypes contained higher amount of total phenol and O.D. phenol than susceptible. In diseased leaves their amount increased in both the genotypes.

In Table 36c, it is clear that the per cent increase in total phenols was higher (25.47%) in resistant varieties viz., TM-96-2 and TARM-18 followed by 20.90 per cent in moderately resistant varieties viz., BGS-9 and TM-97-55, but it was least of 19.61 per cent in susceptible varieties. Similarly, highest per cent increase in ortho-dihydroxy phenols of 26.36 was found in resistant varieties followed by 25.35 per cent in moderately resistant varieties while it was only 21.61 per cent in susceptible varieties.

4.8 Disease management

4.8.1 *In vitro* evaluation of fungicides

Eight systemic and five non-systemic fungicides were evaluated at three concentrations in the laboratory for their efficacy against *C. truncatum* through poison food technique. The data are presented in Table 37 and 38 and depicted in Fig. 19 and 20.

The results presented in Table 37 revealed that, there was a significant difference between the systemic fungicides, concentrations and interactions. Propiconazole, carbendazim, thiophanate methyl and benomyl inhibited cent per cent of mycelial growth of *C. truncatum* at all the three concentrations (0.05, 0.1 and 0.15%). Hexaconazole and tricyclazole also showed cent per cent inhibition at 0.1 and 0.15 per cent concentrations, while penconazole and difenconazole showed cent per cent inhibition at 0.15 per cent concentration. The least inhibition of mycelial growth among the systemic fungicides was observed in penconazole (59.45%) at 0.05 per cent concentration (Plate 16).

Among the non-systemic fungicides tested, it is evident form the Table 38 that the maximum mycelial growth inhibition was obtained by carbendazim + mancozeb (100%) at all the three concentrations (0.1, 0.2 and 0.25%), followed by chlorothalonil (100%) at 0.25 per cent. The least inhibition of mycelial growth was observed in copper oxychloride (35.82%) at 0.1 per cent concentration (Plate 17).

4.8.2 *In vitro* evaluation of botanicals

Ten plant extracts were evaluated at three concentrations in the laboratory for their efficacy against *C. truncatum* through poison food technique as detailed in Material and Methods. The data are presented in Table 39 and depicted in Fig. 21.

Table 39 revealed that amongst the ten plant extracts evaluated, azadirachtin at 10 per cent concentration was found to be best in inhibiting the mycelial growth of *C. truncatum* (63.34%) and found significantly superior over all the other extracts, followed by eucalyptus oil (60.62%), garlic (59.44%) and neem seed kernel extract (56.63%) at 10 per cent. Least inhibition of mycelial growth of *C. truncatum* was recorded in bellary jali (35.55%) at 5 per cent concentration (Plate 18).

Table 36c: Effect of anthracnose on phenol content (mg/g of fresh weight) in varieties of greengram with different levels of resistance

Phenols	Type of leaves	Resistant		Mean	Per cent increase over healthy	Moderately resistant		Mean	Per cent increase over healthy	Susceptible/ Highly susceptible			Mean	Per cent increase over healthy
		TM-96-2	TARM-18			BGS-9	TM-97-55			Sel-4	China mung	Yellow mung		
a. Total phenols	Healthy	5.44	5.24	5.34	25.47	5.27	5.35	5.31	20.90	4.12	4.16	4.10	4.13	19.61
	Diseased	6.41	6.99	6.70		6.16	6.68	6.42		5.15	5.21	4.46	4.94	
b. Orthodihydroxy phenols	Healthy	3.55	3.80	3.68	26.36	3.64	3.46	3.55	25.35	2.72	3.21	2.95	2.96	21.61
	Diseased	4.41	4.89	4.65		4.59	4.31	4.45		3.42	3.80	3.58	3.60	

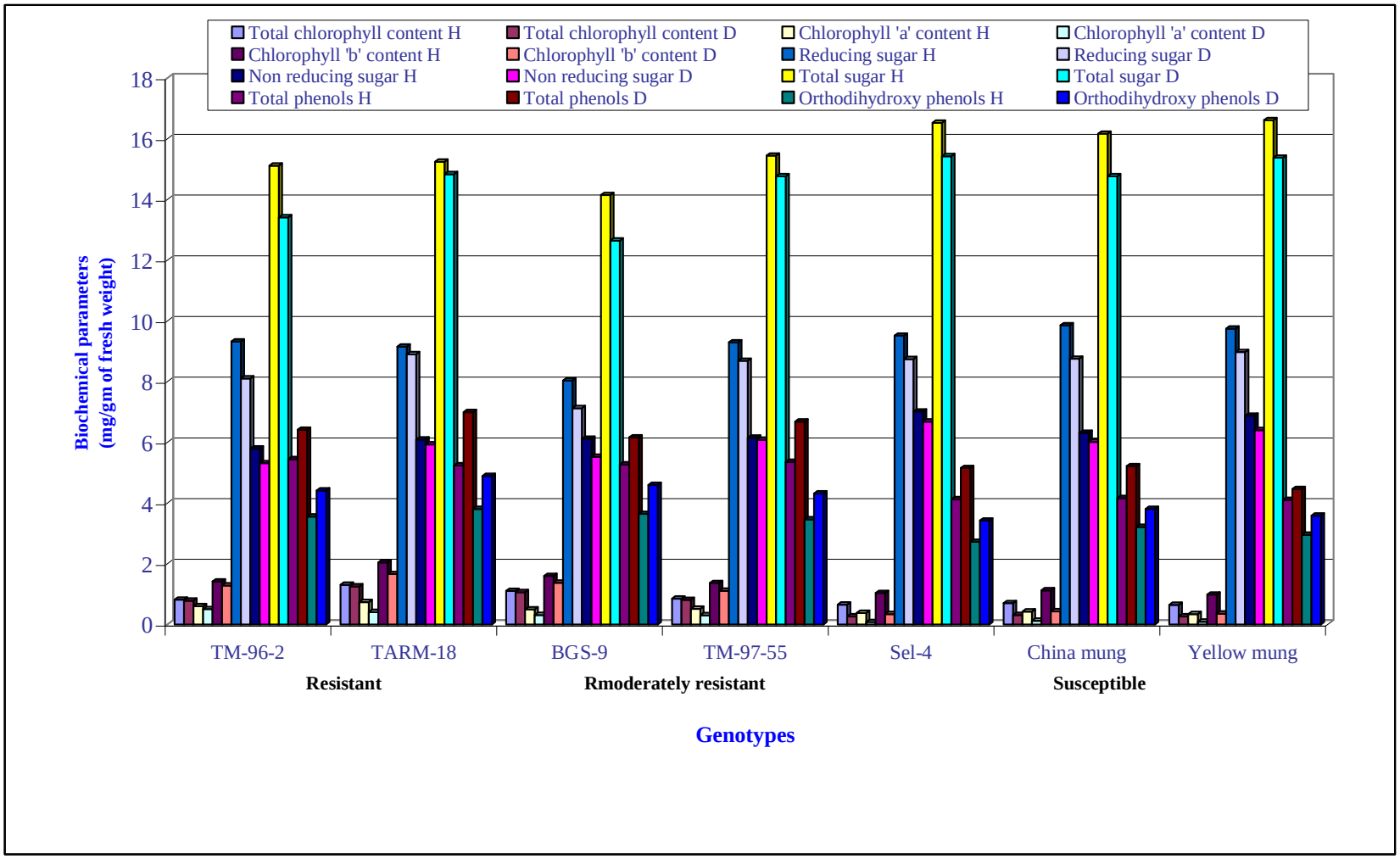


Fig. 18: Biochemical parameters in healthy and diseased leaves of resistant, moderately resistant and susceptible varieties as influenced by anthracnose of greengram

Table 37: *In vitro* evaluation of systemic fungicides against *Colletotrichum truncatum*

Sl. No	Chemicals	Percent inhibition of radial growth over control			
		0.05%	0.1%	0.15%	Mean
1.	Propiconazole 25%	100.00 (90.05)*	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)
2.	Hexaconazole 5%	96.20 (78.85)	100.00 (90.05)	100.00 (90.05)	98.73 (86.31)
3.	Carbendazim 50 WP	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)
4.	Thiophanate methyl 70 WP	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)
5.	Penconazole 10 EC	59.45 (50.47)	69.60 (56.56)	100.00 (90.05)	76.35 (65.69)
6.	Difenconazole 25 EC	60.17 (50.89)	70.43 (57.09)	100.00 (90.05)	76.86 (66.01)
7.	Benomyl 50 WP	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)
8.	Tricyclozole 75 WP	72.51 (58.41)	100.00 (90.05)	100.00 (90.05)	90.83 (79.50)
	Mean	86.04 (74.85)	92.50 (81.74)	100.00 (90.05)	
		S.E m ±		C.D at 1%	
	Fungicide(F)	0.12		0.45	
	Concentration (C)	0.07		0.26	
	FXC	0.20		0.75	

* Values in parenthesis are arcsine transformed values

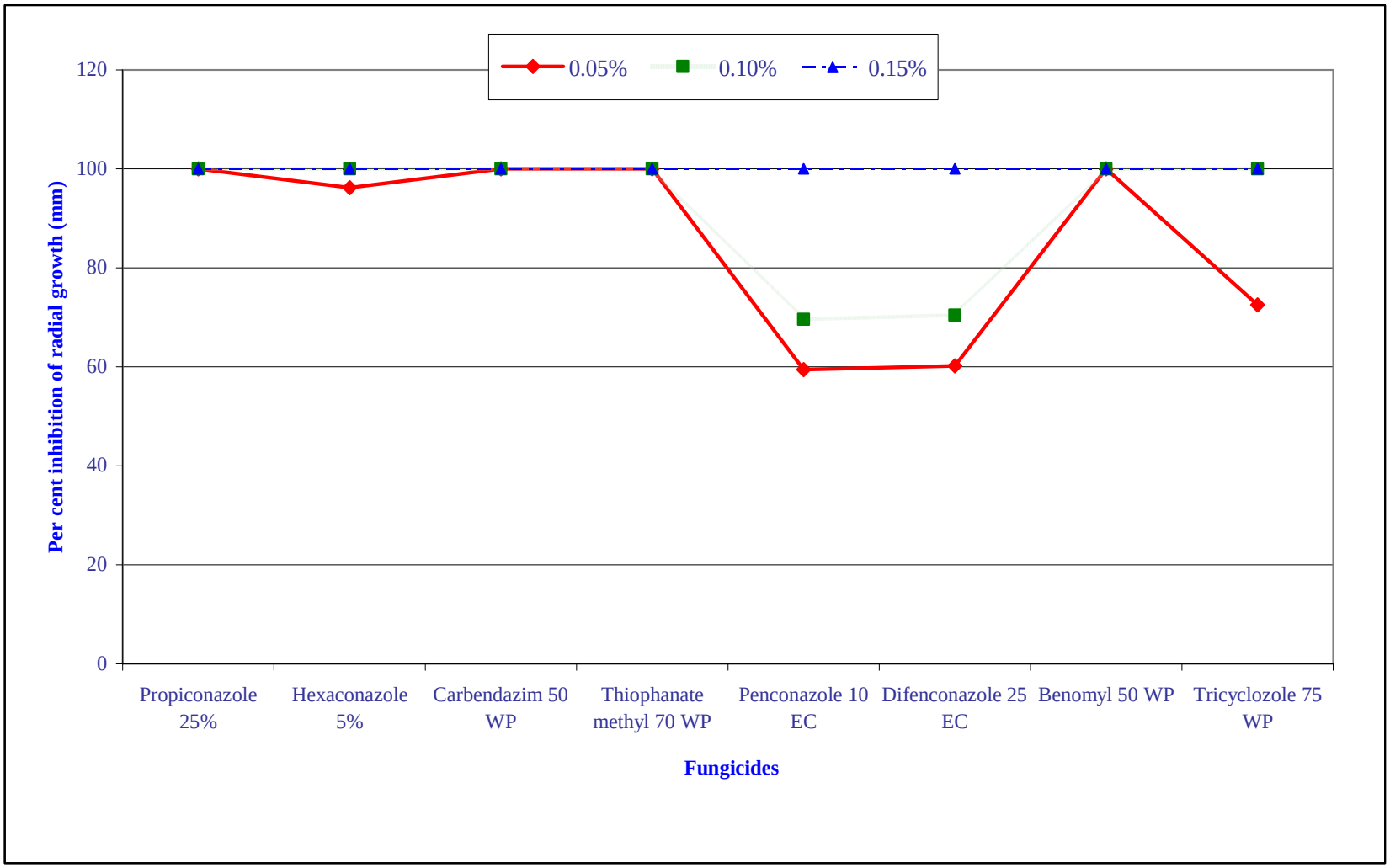


Fig. 19: In vitro evaluation of systemic fungicides against C. truncatum

Table 38: *In vitro* evaluation of non systemic fungicides against *Colletotrichum truncatum*

Sl. No	Chemicals	Percent inhibition of radial growth over control			
		0.1%	0.2%	0.25%	Mean
1.	Mancozeb 75 WP	60.36 (51.00)*	66.52 (54.67)	85.55 (67.70)	70.81 (57.80)
2.	Chlorothalonil 75 WP	84.39 (66.76)	87.81 (69.60)	100.00 (90.05)	90.73 (75.47)
3.	Propineb 75 WP	46.52 (43.02)	55.48 (48.17)	61.46 (51.65)	54.49 (47.61)
4.	Carbendazim 12% + Mancozeb 63%	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)	100.00 (90.05)
5	Copper oxy chloride 50 WP	35.82 (36.78)	45.47 (42.42)	53.61 (47.09)	44.97 (42.10)
	Mean	65.42 (57.52)	71.06 (60.98)	80.12 (69.30)	
		S.E m ±		C.D at 1%	
	Fungicide(F)	0.11		0.42	
	Concentration (C)	0.09		0.34	
	FXC	0.21		0.80	

* Values in parenthesis are arcsine transformed values

4.8.3 *In vitro* evaluation of bioagents

Six bioagents were evaluated for their efficacy against *C. truncatum* through dual culture technique as explained in Material and Method. The results of the study are presented in Table 40. *Trichoderma harzianum* gave highest growth inhibition (64.38%) followed by *Gliocladium virens* (58.47%), *T. koningii* (54.37%) and *T. viride* (50.46%). The least growth inhibition of the fungus was observed in *Bacillus subtilis* (35.44%) and *Pseudomonas fluorescens* (26.56%) (Plate 19 and Fig. 22).

4.8.4 *In vitro* evaluation of ITKs

Table 41 revealed that spore germination of *C. truncatum* differed significantly with dilutions and their interactions. The per cent inhibition of spore germination of *C. truncatum* over control was significantly higher in cow urine (75.57%) followed by fermented butter milk (64.59%) and panchagavya (62.70%). The least per cent inhibition of spore germination was recorded in cow milk (56.10%) followed by vermiwash (59.73%) (Fig. 23).

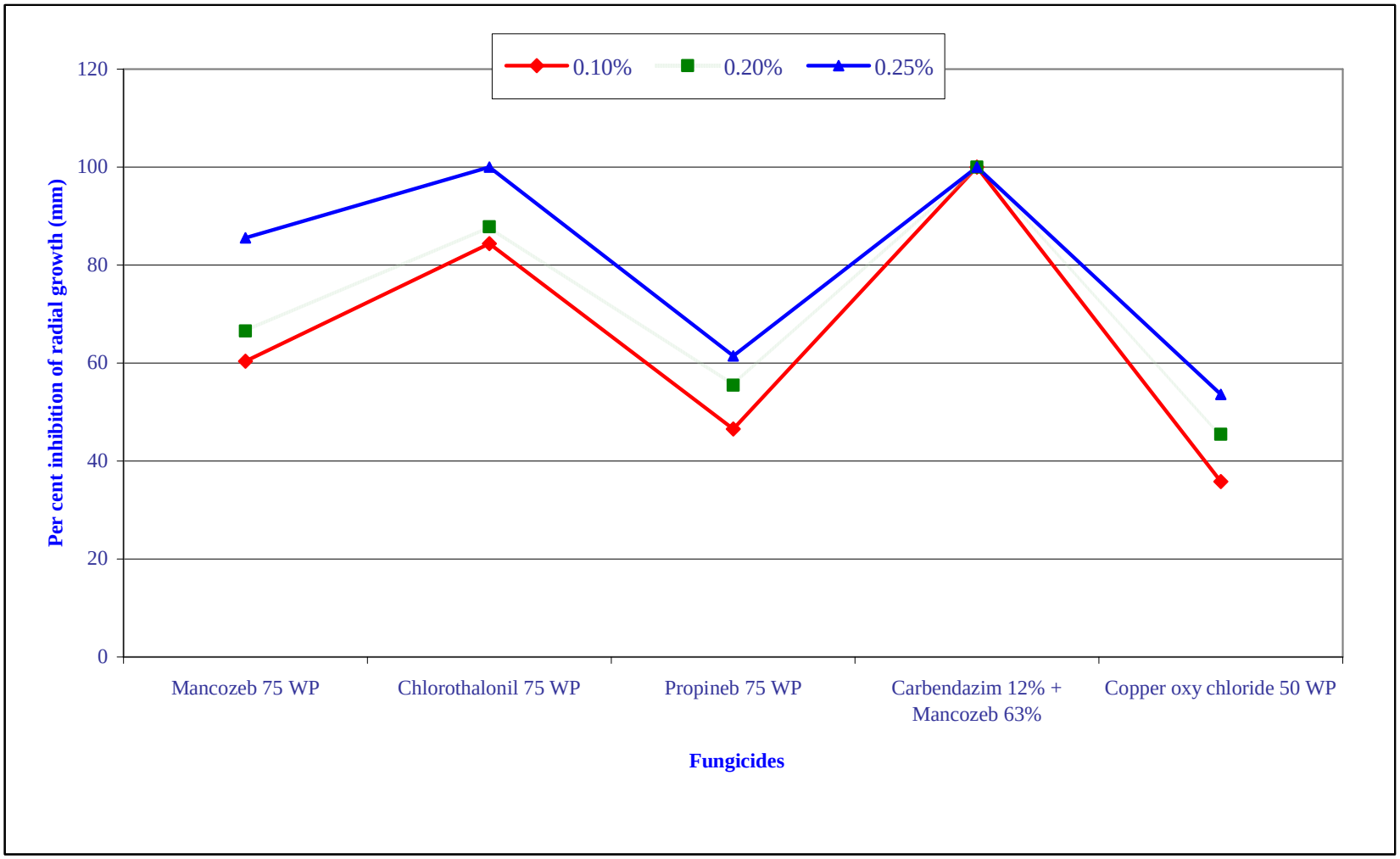
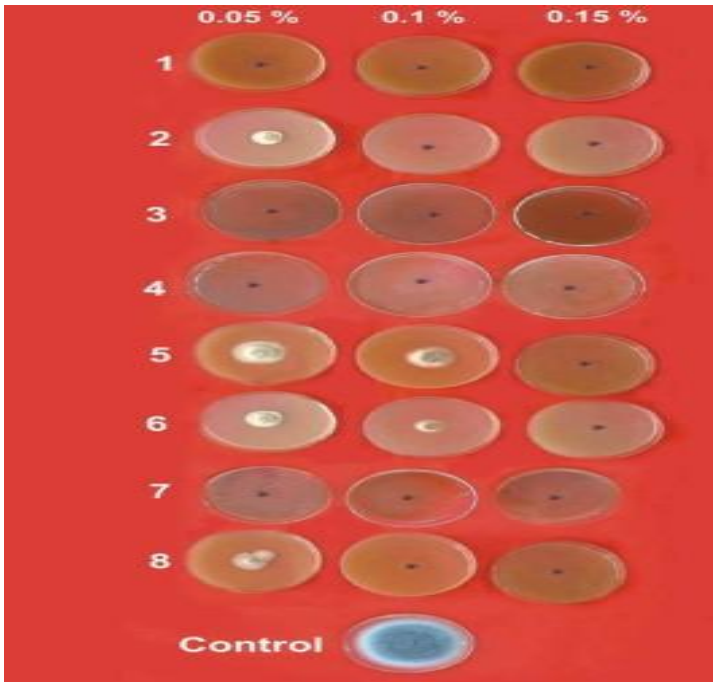
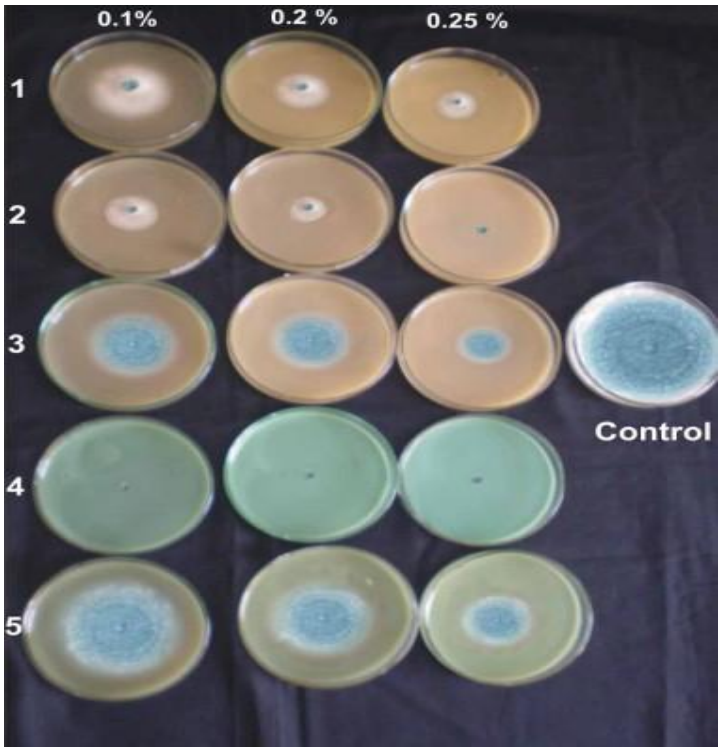


Fig. 20: In vitro evaluation of non-systemic fungicides against C. truncatum



1.Propiconazole 2. Hexaconazole 3. Carbendazim 4. Thiophanate methyl
5. Penconazole 6. Difenconazole 7. Benomyl 8. Tricyclazole

Plate 16: In vitro evaluation of systemic fungicides against C. truncatum



1.Mancozeb 2.Chlorothalonil 3.Propineb 4.Carbendazim+Mancozeb 5.Copper oxy chloride

Plate 17: In vitro evaluation non systemic fungicides against C.truncatum

Table 39: *In vitro* evaluation of botanicals against *Colletotrichum truncatum*

Sl. No	Botanicals	Percent inhibition of radial growth over control			
		5%	7.5%	10%	Mean
1.	Ballary Jali (leaf)	35.55 (36.62)*	42.58 (40.75)	46.76 (43.16)	41.63 (40.18)
2.	Cynodon (plant)	40.64 (39.62)	45.62 (45.51)	50.42 (45.26)	45.56 (42.46)
3.	Parthenium (plant)	40.48 (39.53)	47.31 (43.48)	52.47 (46.44)	46.75 (43.15)
4.	Garlic (bulb)	43.37 (41.21)	49.25 (44.59)	59.44 (50.46)	50.69 (45.42)
5.	Onion (bulb)	39.28 (38.83)	46.61 (43.08)	52.33 (46.36)	46.07 (42.75)
6.	Neem (kernel)	42.17 (40.51)	48.33 (44.06)	56.63 (48.83)	49.04 (44.47)
7.	Eucalyptus (oil)	47.37 (43.51)	50.23 (45.15)	60.62 (51.16)	52.74 (46.61)
8.	Azadirachtin (herbal product)	47.45 (43.56)	51.39 (45.82)	63.34 (52.76)	54.06 (47.38)
9.	Turmeric (rhizome)	39.47 (38.94))	44.36 (41.78)	49.46 (44.71)	44.43 (41.81)
10	Ginger (rhizome)	37.48 (37.77)	44.47 (41.84)	48.56 (44.20)	43.50 (41.27)
	Mean	41.33 (40.01)	47.02 (43.31)	54.00 (47.33)	
		S.E m ±		C.D at 1%	
	Botanicals (B)	0.14		0.52	
	Concentration (C)	0.08		0.30	
	BXC	0.24		0.90	

* Values in parenthesis are arcsine transformed values

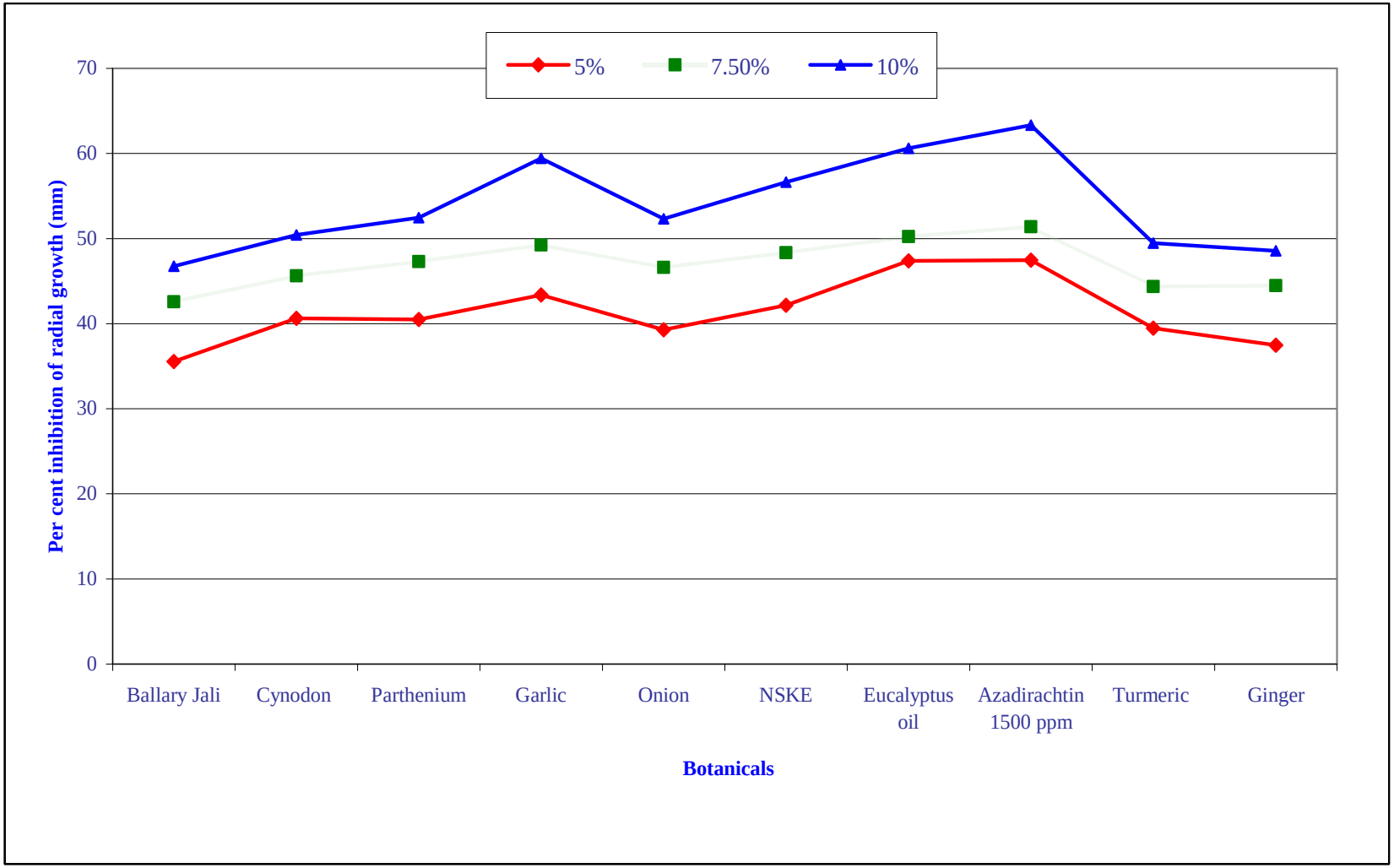


Fig. 21: In vitro evaluation of botanicals against C. truncatum

Table 40: *In vitro* evaluation of bioagents against *Colletotrichum truncatum*

Sl. No.	Bio-agents	Percent inhibition
1	*Gliocladium virens*	58.47 (49.88)*
2	*Trichoderma koningii*	54.37 (47.51)
3	*Trichoderma viride*	50.46 (45.26)
4	*Trichoderma harzianum*	64.38 (53.35)
5	*Pseudomonas fluorescens*	26.56 (31.02)
6	*Bacillus subtilis*	35.44 (36.54)
S.Em±		0.13
CD at 1%		0.53

* Values in parenthesis are arcsine transformed values

Among the dilutions, 1:2 dilution recorded maximum inhibition (74.38%) of spore germination followed by 1:5 dilution (71.61%) and 1:10 dilution (67.78%).

Cow urine at 1:2, 1:5 and 1:10 dilutions recorded significantly higher per cent inhibition of spore germination, which were on par with each other. Among all ITK, cow urine was found to be superior.

4.8.5 Integrated disease management

The experiment on integrated disease management of anthracnose of greengram using fungicides, herbal products (eucalyptus oil and azadirachtin), ITKs (cow urine) and bioagent formulations (*Trichoderma harzianum*) was conducted during *kharif*, 2006 and *kharif* 2007. The results are presented in Table 42, 43 and 44 and Fig. 24 and Plate 20.

4.8.6 .Per cent disease index (PDI)

The results obtained during 2006 revealed that, all the treatments were significantly superior over untreated control (Table 42). From the data, it is clear that the least per cent disease index was found in the treatments *viz.*, T_{16} (foliar spray of propiconazole) and T_{15} (foliar spray of hexaconazole) and they were found on par with each other with PDI of 18.88 and 19.85 per cent, respectively and they were significantly superior over other treatments. This was followed by T_{11} (benomyl seed treatment + foliar spray of benomyl), T_{10} (carbendazim seed treatment + foliar spray of carbendazim) and T_{12} (thiophanate methyl seed treatment + foliar spray of thiophanate methyl) which recorded 20.10, 23.50 and 25.18 PDI, respectively. Whereas, the other treatments like T_{13} (carbendazim + mancozeb seed treatment + foliar spray of carbendazim + mancozeb) and T_{14} (tricyclazole + foliar spray of tricyclazole) were statistically on par with each other with PDI of 28.08 and 29.05 per cent,

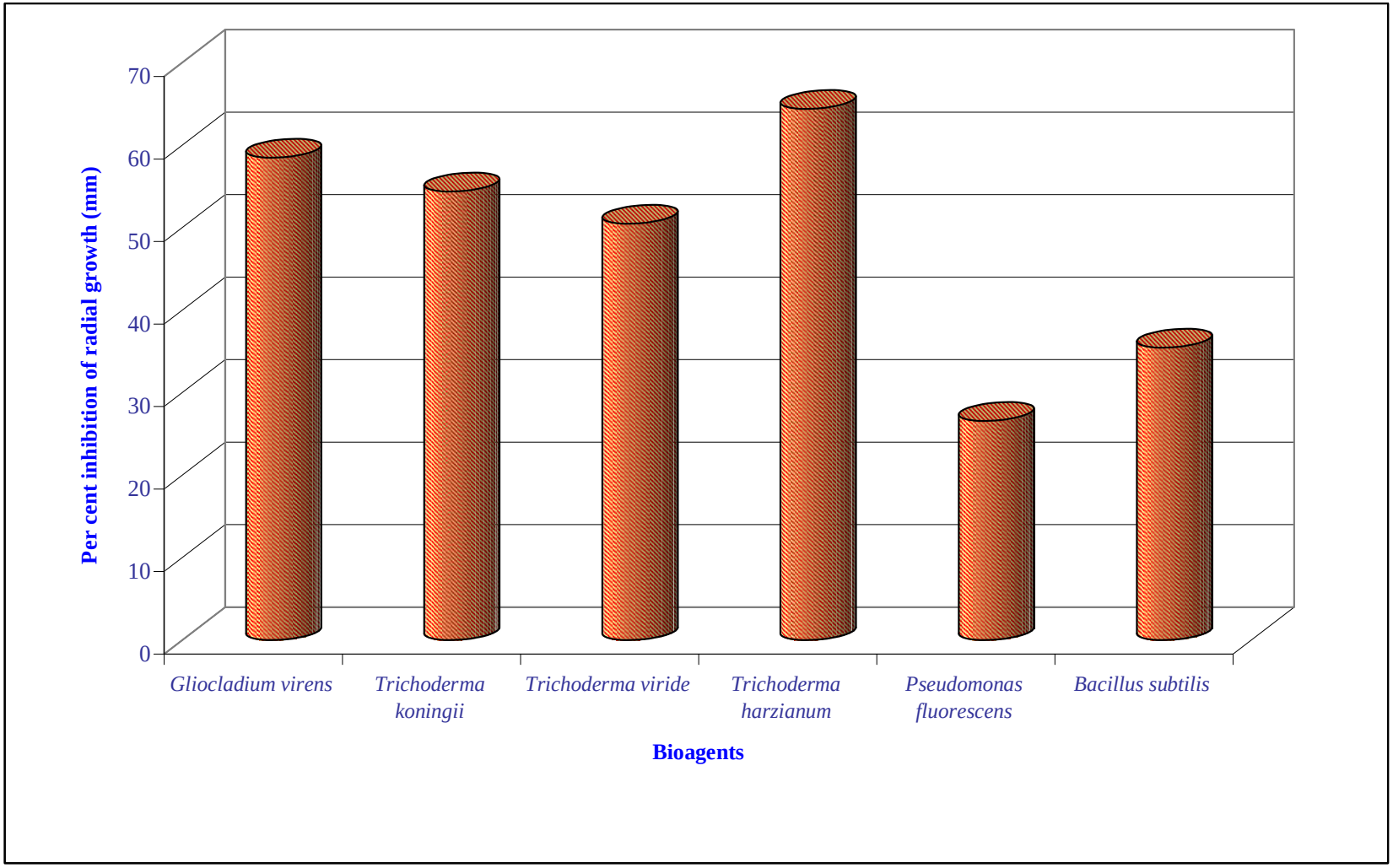
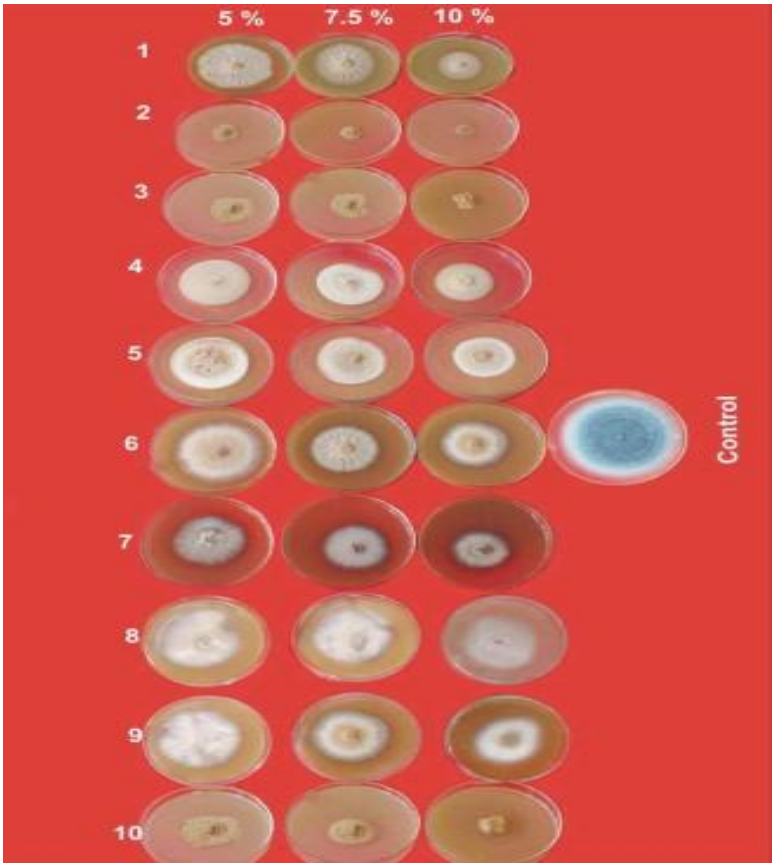
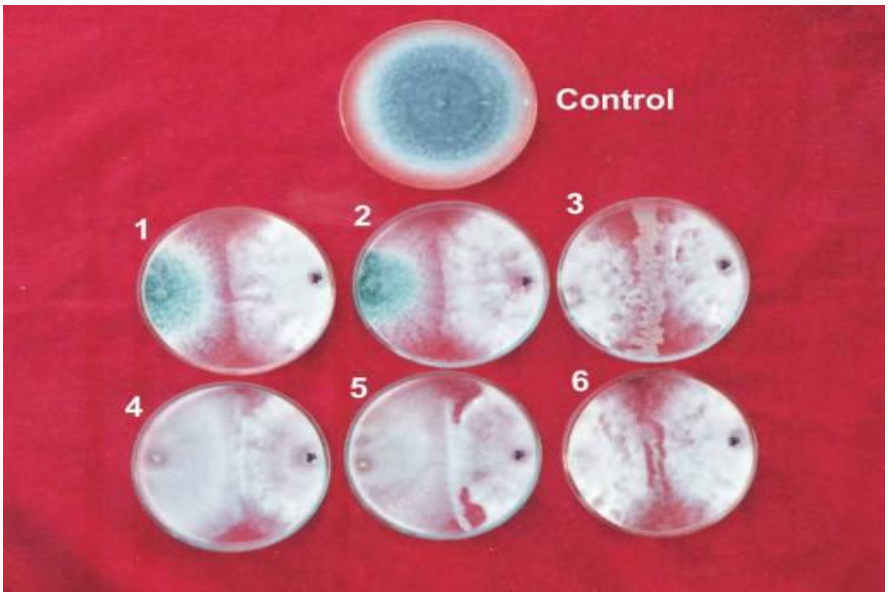


Fig. 22: In vitro evaluation of bioagent against C. truncatum



1.Cynodon 2.Azadirachitin 3.Eucalyptusoil 4. NSKE 5. Onion 6. Turmeric
7.Parthenium 8. Bellary Jali 9.Ginger 10. Garlic

Plate 18: Invitro evaluation of botanicals against C.truncatum



1. Trichoderma viride 2. Trichoderma koningii 3. Pseudomonas flurescens
4. Trichoderma virens 5. Trichoderma harzianum 6. Bacillus subtilis

Plate 19: Invitro evalution of bioagents against C. truncatum

Table 41: Effect of ITK's on spore germination of *Colletotrichum truncatum*

Sl. No	Name of ITK	Percent inhibition of spore germination over control at different concentrations					Mean
		1:2	1:5	1:10	1:15	1:20	
1.	Cow urine	82.08 (64.93)*	81.77 (64.72)	80.99 (64.13)	72.35 (58.25)	60.63 (51.12)	75.57 (60.63)
2.	Fermented Butter milk	75.50 (60.31)	72.60 (58.42)	69.51 (56.46)	55.29 (48.02)	50.05 (45.01)	64.59 (53.64)
3.	Cow milk	69.25 (56.30)	65.42 (53.96)	58.46 (49.85)	45.22 (42.24)	42.15 (40.47)	56.10 (48.56)
4.	Vermiwash	71.38 (57.63)	67.61 (55.29)	63.00 (52.51)	50.91 (45.51)	45.74 (42.54)	59.73 (50.70)
5	Panchagavya	73.69 (59.12)	70.65 (57.18)	66.92 (54.87)	53.43 (46.95)	48.81 (44.30)	62.70 (52.48)
	Mean	74.38 (59.66)	71.61 (57.91)	67.78 (55.57)	55.44 (48.19)	49.48 (44.69)	
		S.E m ±			C.D at 1%		
ITK's (I)		0.10			0.51		
Dilutions (D)		0.10			0.51		
IXD		0.22			1.14		

* Values in parenthesis are arcsine transformed values

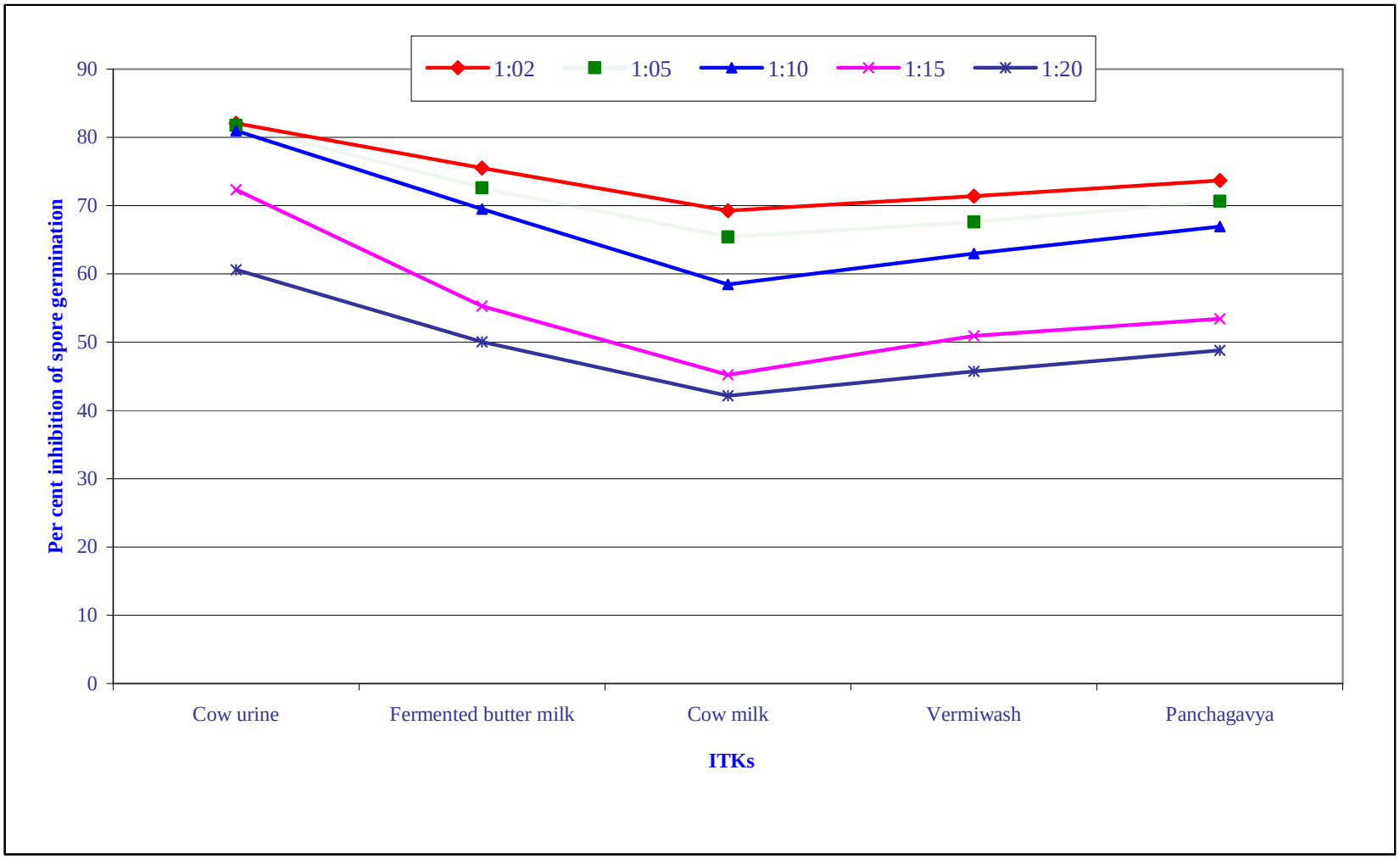


Fig. 23: Effect of ITKs on spore germination of C. truncatum

Table 42: Integrated disease management of anthracnose of greengram during *Kharif*, 2006

Sl. No.	Treatment	Percent disease index	Percent reduction in disease over control	Grain yield (q/ha)	Per cent grain yield increase over control	Stalk yield (q/ha)	B:C ratio
1	*Trichoderma harzianum* at 4 g/kg of seed (SD)	51.05 (45.58)*	6.01	7.94	0.63	7.98	4.88
2.	Carbendazium at 2 g/kg of seed (SD)	47.10 (43.34)	13.23	7.99	1.27	8.43	9.77
3.	Benomyl at 2 g/kg of seed (SD)	46.92 (43.23)	13.56	8.02	1.65	8.55	7.95
4.	Thiophanate methyl at 2 g/kg of seed (SD)	47.30 (43.45)	12.86	7.98	1.14	8.35	4.15
5.	Carbendazim + mancozeb at 2 g/kg of seed (SD)	48.75 (44.28)	10.19	7.97	1.01	8.17	7.29
6.	Tricyclozole at 2 g/kg of seed (SD)	49.26 (44.57)	9.25	7.96	0.89	8.08	2.44
7.	T_1 + One spray of Eucalyptus oil at 10%	41.67 (40.20)	23.23	8.23	4.31	11.10	0.03
8.	T_1 + One spray of cow urine at 10 %	43.85 (41.47)	19.22	8.19	3.80	10.96	0.60
9.	T_1 + One spray of Azadirachtin (1500 ppm) at 0.2%	39.28 (38.81)	27.63	9.03	14.45	11.89	5.84
10.	T_2 + One spray of carbendazim at 0.1%	23.50 (28.99)	56.71	11.23	42.08	14.09	22.35
11.	T_3 + One spray of Benomyl at 0.1%	20.10 (26.64)	62.97	11.43	44.87	14.28	12.47
12	T_4 + One spray of Thiophanate methyl at 0.1%	25.18 (30.11)	53.61	10.98	39.16	13.83	7.52
13.	T_5 + One spray of carbendazim + Mancozeb at 0.2%	28.08 (31.99)	48.27	10.12	28.26	12.97	8.95
14	T_6 + One spray of Tricyclozole at 0.1%	29.05 (32.61)	46.48	9.89	25.35	12.74	3.69
15	One spray of Hexaconazole at 0.1%	19.85 (26.46)	63.43	11.82	49.81	14.52	23.04
16	One spray of Propiconazole at 0.1%	18.88 (25.75)	65.22	12.16	53.99	14.82	14.10
17	Control	54.28 (47.45)	-	7.89	-	7.75	-
	S.E m ±	0.54		0.17		0.11	
	C.D at 5%	1.63		0.51		0.30	

* Values in parenthesis are arcsine transformed values.

Cost of the grain at Rs. 2200/qt and stalk at Rs. 50/qt. Labour charges for spray =Rs. 90/-.
Quantity of spray solution used per hectare : 500lit. Cost of fungicides / biorational in Rs/kg or litere *Trichoderma harzianum* (200), Carbendazim (450), Benomyl (1040), Thiophanate methyl (1600), Carbendazim + Mancozeb (460), Tricyclozole (2200), Eucalyptus oil (625), Azadirachtin (350), Hexaconazole (600), Propiconazole (1200) and cow urine (25).

respectively and they were significantly superior over untreated control. Among the biorationals used, the least incidence of anthracnose was noticed in T_9 (*Trichoderma harzianum* seed treatment + foliar spray of azadirachtin) followed by T_7 (*T. harzianum* seed treatment + foliar spray of eucalyptus oil) and T_8 (*T. harzianum* seed treatment + foliar spray of cow urine) with PDI of 39.28, 41.67 and 43.85 per cent, respectively and they were significantly superior over untreated control.

The highest per cent disease reduction of 65.22 was recorded in propiconazole followed by hexaconazole (63.43%) and benomyl seed treatment + foliar spray of benomyl (62.97%). Treatments like carbendazim seed treatment + foliar spray of carbendazim , thiophanate methyl seed treatment + foliar spray of thiophanate methyl and (carbendazim + mancozeb) seed treatment + foliar spray of (carbendazim + mancozeb) recorded considerably more per cent disease reduction 56.71, 53.61 and 48.27 per cent, respectively. The least per cent reduction of 6.01, 9.25 and 10.19 was recorded in *T. harzianum* seed treatment, tricyclazole seed treatment and carbendazim + mancozeb seed treatment, respectively.

The results obtained during *kharif* 2007 followed similar trend of results but in higher intensity of incidence of disease as observed during *kharif* 2006. All the treatments were significantly superior over untreated control (Table 43). The T_{16} (foliar spray of propiconazole) though it recorded the least disease index of 20.16 per cent. It was found on par with T_{15} (foliar spray of hexaconazole) and T_{11} (benomyl seed treatment + foliar spray of benomyl) which recorded 21.04 and 21.25 per cent disease index, respectively. The treatments *viz.*, T_{10} (carbendazim seed treatment + foliar spray of carbendazim) and T_{12} (thiophanate methyl seed treatment + foliar spray of thiophanate methyl) were found on par with each other with PDI of 24.77 and 26.63 per cent, respectively and they were found significantly superior over untreated control (56.07%). Among the biorationals used the least incidence was noticed in T_9 (*T. harzianum* seed treatment + foliar spray of azadirachtin) followed by T_7 (*T. harzianum* seed treatment + foliar spray of eucalyptus oil) and T_8 (*T. harzianum* seed treatment + foliar spray of cow urine) with PDI of 40.50, 42.95 and 45.02 per cent respectively and they were significantly superior over untreated control.

Further, the per cent reduction of disease was highest in propiconazole (64.04%) followed by hexaconazole (62.48%) and benomyl seed treatment + foliar spray of benomyl (62.10%). Carbendazim seed treatment + foliar spray of carbendazim, thiophanate methyl seed treatment + foliar spray of thiophanate methyl and (carbendazim + mancozeb) seed treatment + foliar spray of carbendazim + mancozeb also recorded per cent disease reduction of 55.82, 52.51 and 46.51, respectively. The least per cent disease reduction was recorded in *T. harzianum* seed treatment (5.40%), tricyclazole seed treatment (10.02%) and carbendazim + mancozeb seed treatment (10.81%).

The pooled data (Table 44) indicated that, the treatment propiconazole recorded the least PDI (19.52) followed by hexaconazole (20.45%) and benomyl seed treatment + foliar spray of benomyl (20.68%) while carbendazim seed treatment + foliar spray of carbendazim recorded the mean PDI of 24.14. The disease reduction of 64.62, 62.94, 62.52 and 56.25 per cent was observed in propiconazole, hexaconazole, benomyl seed treatment + foliar spray of benomyl and carbendazim seed treatment + foliar spray of carbendazim, respectively.

4.8.7 Grain yield

The highest yield of 12.16 q per ha during *kharif* 2006 was recorded in propiconzole (T_{16}) and the least yield was recorded in untreated control (7.89 q/ha), though propiconazole (T_{16}) was significantly superior over all other treatments, however it is on par with hexaconazole (11.82 q/ha), followed by benomyl seed treatment + foliar spray of benomyl (11.43 q/ha) and carbendazim seed treatment + foliar spray of carbendazim (11.23 q/ha). In the biorational treatments *viz.*, T, harzianum seed treatment + foliar spray azadirachtin, *T. harzianum* seed treatment + foliar spray of eucalyptus oil and *T. harzianum* seed treatment + foliar spray of cow urine recorded yield of 9.03, 8.23 and 8.19 q per ha, respectively (Table 42).

During *kharif* 2007, the maximum yield was recorded in propiconazole (11.45 q/ha) which was on par with hexaconazole (11.10 q/ha) followed by benomyl seed treatment + foliar spray of benomyl (10.84 q/ha), carbendazim seed treatment + foliar spray of carbendazim

Table 43: Integrated disease management of anthracnose of greengram during *Kharif*, 2007

Sl. No.	Treatment	Percent disease index	Percent reduction in disease over control	Grain yield (q/ha)	Per cent grain yield increase over control	Stalk yield (q/ha)	B:C ratio
1	*Trichoderma harzianum* at 4 g/kg of seed (SD)	53.04 (46.74)*	5.40	7.17	0.28	6.95	2.04
2.	Carbendazium at 2 g/kg of seed (SD)	48.48 (44.13)	13.54	7.23	1.26	7.62	9.15
3.	Benomyl at 2 g/kg of seed (SD)	48.05 (43.88)	14.30	7.26	1.68	7.71	7.54
4.	Thiophanate methyl at 2 g/kg of seed (SD)	48.83 (44.33)	12.91	7.21	0.98	7.42	3.35
5.	Carbendazim + mancozeb at 2 g/kg of seed (SD)	50.01 (45.00)	10.81	7.20	0.84	7.23	5.67
6.	Tricyclozole at 2 g/kg of seed (SD)	50.45 (45.26)	10.02	7.19	0.70	7.14	1.80
7.	T_1+One spray of Eucalyptus oil at 10%	42.95 (40.95)	23.39	7.67	7.42	9.85	0.04
8.	T_1 + One spray of cow urine at 10 %	45.02 (42.14)	19.71	7.54	5.60	9.72	0.75
9.	T_1 + One spray of Azadirachtin (1500 ppm) at 0.2%	40.50 (39.52)	27.77	8.45	18.35	10.63	6.61
10.	T_2 + One spray of carbendazim at 0.1%	24.77 (29.84)	55.82	10.55	47.76	12.81	22.88
11.	T_3 + One spray of Benomyl at 0.1%	21.25 (27.45)	62.10	10.84	51.82	13.02	12.98
12	T_4 + One spray of Thiophanate methyl at 0.1%	26.63 (31.06)	52.51	10.32	44.54	12.50	7.70
13.	T_5 + One spray of carbendazim + Mancozeb at 0.2%	29.99 (33.20)	46.51	9.46	32.49	11.64	9.26
14	T_6 + One spray of Tricyclozole at 0.1%	30.35 (33.43)	45.87	9.24	29.41	11.42	3.85
15	One spray of Hexaconazole at 0.1%	21.04 (27.30)	62.48	11.10	55.46	13.14	23.15
16	One spray of Propiconazole at 0.1%	20.16 (26.68)	64.04	11.45	60.36	13.37	14.22
17	Control	56.07 (48.48)	-	7.14	-	6.82	-
	S.E m ±	0.70		0.13		0.07	
	C.D at 5%	2.09		0.41		0.23	

* Values in parenthesis are arcsine transformed values

Cost of the grain at Rs. 2200/qt and stalk at Rs. 50/qt. Labour charges for spray =Rs. 90/-.

Quantity of spray solution used per hectare : 500lit. Cost of fungicides / biorational in Rs/kg or litere *Trichoderma harzianum* (200), Carbendazim (450), Benomyl (1040), Thiophanate methyl (1600), Carbendazim + Mancozeb (460), Tricyclozole (2200), Eucalyptus oil (625), Azadirachtin (350), Hexaconazole (600), Propiconazole (1200) and cow urine (25).

Table 44: IDM of anthracnose of greengram caused by *Colletotrichum truncatum* during *kharif* 2006 and 2007 - a pooled analysis

Sl. No.	Treatment	Percent disease index	Percent reduction in PDI over control	Grain yield (q/ha)	Percent increase in grain yield over control	Stalk yield (q/ha)	Percent increase in stalk yield over control	Benefit:Cost ratio
1.	*Trichoderma harzianum* at 4 g/kg of seed (SD)	52.03 (46.16)*	5.71	7.55	0.40	7.47	2.47	3.46
2.	Carbendazium at 2 g/kg of seed (SD)	47.79 (43.73)	13.39	7.61	1.20	8.03	10.15	9.46
3.	Benomyl at 2 g/kg of seed (SD)	47.49 (43.56)	13.94	7.64	1.60	8.13	11.52	7.75
4.	Thiophanate methyl at 2 g/kg of seed (SD)	48.07 (43.89)	12.89	7.60	1.06	7.89	8.23	3.75
5.	Carbendazim + manrope at 2 g/kg of seed (SD)	49.38 (44.64)	10.51	7.59	0.93	7.70	5.62	6.48
6.	Tricyclozole at 2 g/kg of seed (SD)	49.86 (44.92)	9.64	7.58	0.80	7.61	4.39	2.12
7.	T_1 + One spray of Eucalyptus oil at 10%	42.31 (40.58)	23.32	7.95	5.72	10.48	43.76	0.04
8.	T_1+ One spray of cow urine at 10 %	44.44 (41.80)	19.46	7.87	4.65	10.34	41.84	0.68
9.	T_1 + One spray of Azadirachtin (1500 ppm) at 0.2%	39.89 (39.17)	27.71	8.74	16.22	11.26	54.46	6.23
10.	T_2 + One spray of carbendazim at 0.1%	24.14 (29.42)	56.25	10.88	44.68	13.45	84.50	22.62
11.	T_3 + One spray of Benomyl at 0.1%	20.68 (27.04)	62.52	11.14	48.14	13.65	87.24	12.73
12.	T_4 + One spray of Thiophanate methyl at 0.1%	25.91 (30.59)	53.04	10.65	41.62	13.17	80.66	7.61
13.	T_5 + One spray of carbendazim + Mancozeb at 0.2%	29.04 (32.60)	47.37	9.79	30.19	12.31	68.86	9.11
14.	T_6 + One spray of Tricyclozole at 0.1%	29.70 (33.02)	46.18	9.57	27.26	12.08	65.71	3.77
15.	One spray of Hexaconazole at 0.1%	20.45 (26.88)	62.94	11.46	52.39	13.83	89.71	23.10
16.	One spray of Propiconazole at 0.1%	19.52 (26.22)	64.62	11.80	56.91	14.10	93.42	14.16
17.	Control	55.18 (47.97)	-	7.52	-	7.29	-	-
	SEm$\pm$	0.40		0.11		0.09		
	CD at 5%	1.18		0.35		0.28		

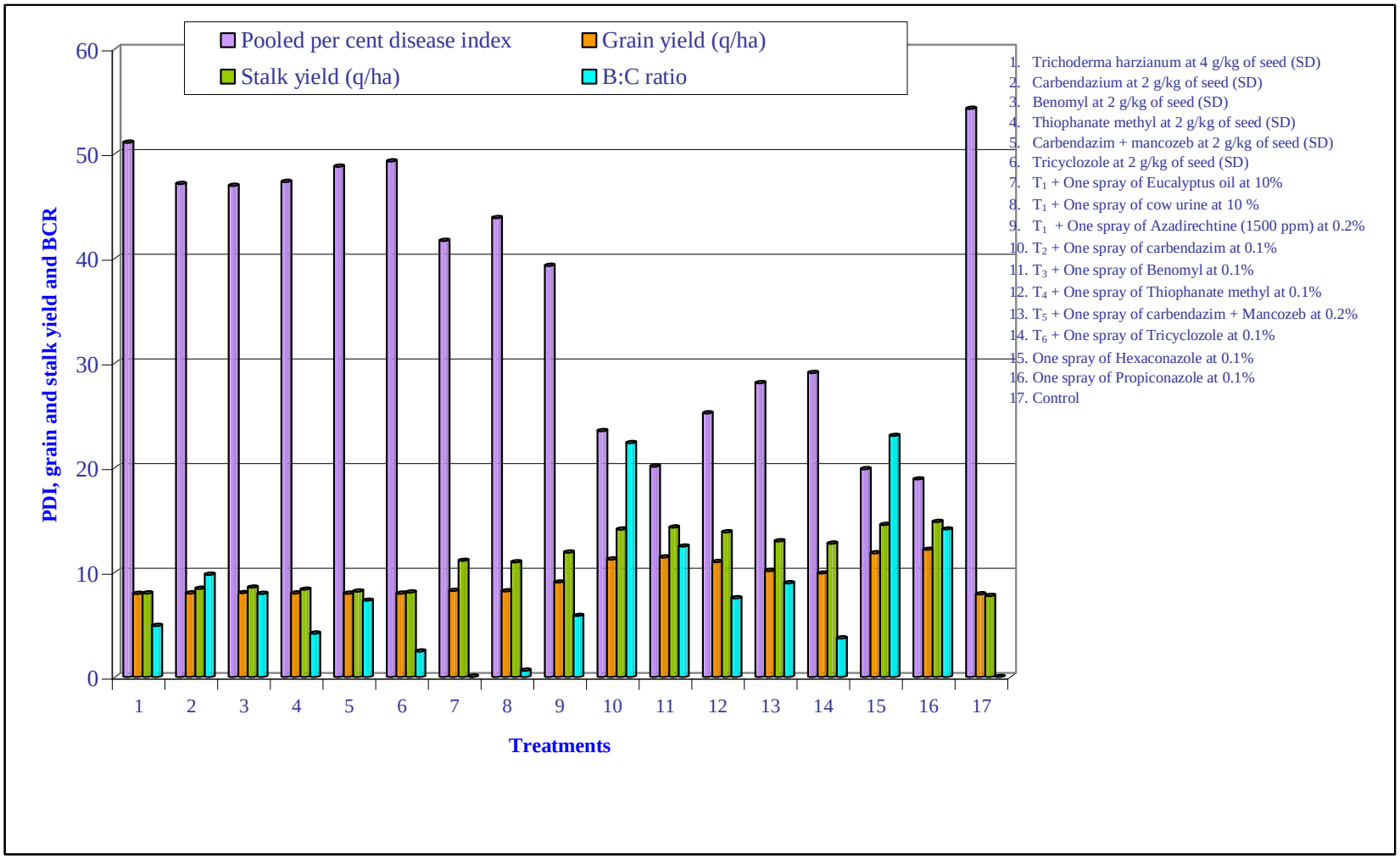
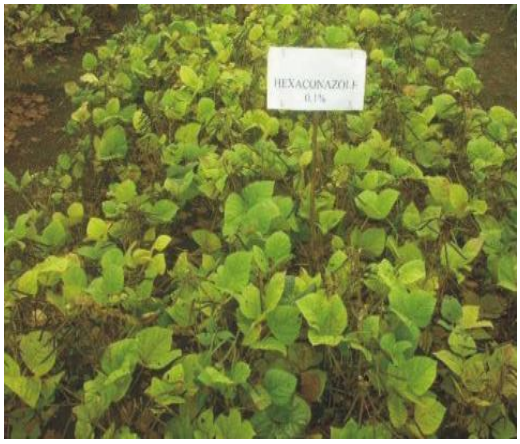


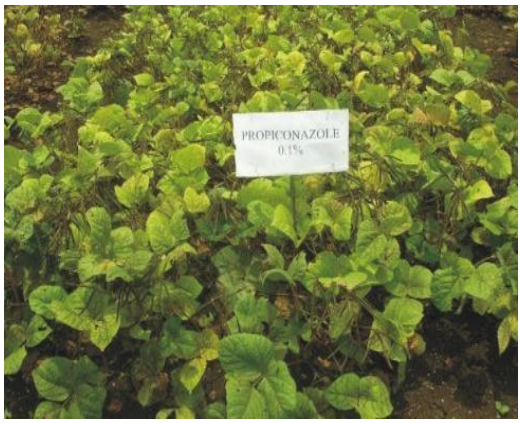
Fig. 24: Effect of different treatment combinations on pooled PDI of anthracnose, grain yield, stalk yield and BCR in greengram

Experimental field view



Hexaconazole

Carbendazim (seed treatment + spray)



Propiconazole

Benomyl (seed treatment + spray)

Control

Plate 20: Effect of different treatments on severity of anthracnose of greengram for effective management

(10.55 q/ha) and thiophanate methyl seed treatment + foliar spray of thiophanate methyl (10.32 q/ha). The yields of 8.45 q per ha and 7.67 q per ha were recorded in *T. harzianum* seed treatment + foliar spray of azadirachtin and *T. harzianum* seed treatment + foliar spray of eucalyptus oil treatment, respectively (Table 43). The pooled data (Table 44) revealed that the yield was highest in propiconazole (11.80 q/ha) which was on par with hexaconazole (11.46 q/ha) followed by benomyl seed treatment + foliar spray of benomyl (11.14 q/ha) and carbendazim seed treatment + foliar spray of carbendazim (10.88 q/ha), however both these treatments were on par with each other. The per cent increase in yield also showed the same trend with yield data.

4.8.8 Stalk yield

Maximum stalk yields were recorded in propiconazole (14.82 q/ha) and hexaconazole (14.52 q/ha) which were significantly superior and found on par with each other followed by benomyl seed treatment + foliar spray of benomyl (14.28 q/ha) during 2006. Whereas, during 2007, propiconazole (13.37 q/ha), hexaconazole (13.14 q/ha), benomyl seed treatment + foliar spray of benomyl (13.02 q/ha) and carbendazim seed treatment + foliar spray of carbendazim (12.81 q/ha) produced significantly more stalk yield than rest of the treatments and were found on par with each other.

The pooled data (Table 44) of two years indicated that the treatment propiconazole recorded the highest stalk yield (14.10 q/ha) followed by hexaconazole (13.83 q/ha) and benomyl seed treatment + foliar spray of benomyl (13.65 q/ha) with per cent increase over control of 93.42, 89.71 and 87.24 per cent, respectively.

4.8.9 Benefit:cost ratio (BCR)

During 2006, Table 42 revealed that highest benefit:cost ratio (BCR) was obtained in foliar spray of hexaconazole (23.04) followed by carbendazim seed treatment + foliar spray of carbendazim (22.35) and foliar spray of propiconazole (14.10) and the lowest in *T. harzianum* seed treatment + foliar spray of eucalyptus (0.03) and *T. harzianum* seed treatment + foliar spray of cow urine (0.60). During 2007 also, maximum benefit was recorded in foliar spray of hexaconazole (23.15) and carbendazim seed treatment + foliar spray of carbendazim (22.88) and the lowest in *T. harzianum* seed treatment + foliar spray of eucalyptus (0.04) and *T. harzianum* + foliar spray of cow urine (0.75) [Table 43]. From the pooled data of two years, it is evident that benefit:cost ratio of 23.10 was recorded in foliar spray of hexaconazole followed by 22.62 in carbendazim seed treatment + foliar spray of carbendazim (Table 44).

5. DISCUSSION

In India, majority of the population is mainly dependent on vegetarian food to meet their nutritional requirement. They wholly depend on pulses for protein requirement. Of the different pulses, greengram [*Vigna radiate* (L.) Wilczek] is one of the important pulse crops, which stands for its easy digestibility and nutritional aspects. However, the crop is subjected to various diseases caused by many pathogens. Anthracnose caused by *Colletotrichum truncatum* is one of the major diseases in greengram resulting in yield reduction of upto 24 to 67 per cent (Deeksha and Tripathi, 2002a). Now, the disease has become one of the major constraints for greengram cultivation, particularly in northern Karnataka. Laxman (2006) recorded 18.2 to 86.57 per cent incidence of the disease in northern Karnataka. Since, there were wide gaps in the research conducted with respect to this disease, the present investigations were carried out for survey of the disease to know the disease severity in different parts of Northern Karnataka, basic research on the nature of the fungus, its cultural and physiological characters, epidemiology of the disease to devise forecasting systems, estimation of loss in yield, reaction of genotypes, biochemical factors for resistance and finally management of disease through integrated approaches of hitherto neglected but important disease of greengram. Keeping these points in view, objectives were set and discussions on the experimental results are presented hereunder. The morphological characters like mycelium, acervuli, setae and conidia of the fungus resembled very closely with *Colletotrichum truncatum* which was described by Saxena and Sinha (1977), Bharadwaj and Singh (1986), Sinclair and Backman (1989) and Laxman (2006). Further, it was confirmed by Agharkar Research Institute, Pune. Thus causal fungus was identified as *Colletotrichum truncatum* (Schw.) Andrus and Moore.

Infection by *C. truncatum* on greengram was initially seen on the lower surface with large (10 to 12 mm diameter) lesions of bright blood red stains and the corresponding upper surface became reddish brown ring like spots of 8 to 10 mm and subsequently became chlorotic, later turned to form shot holes. At later stage, the same lesions appeared on the petioles, stem and pods. The lesions were elongated and brown coloured on petioles and stems. Similar symptoms were observed along with concentric ring of red colour with brown and shot holes in case of urdbean (Tripathi and Beniwal, 1977). Later, Roy (1982), Beniwal *et al.* (1983) and Agarwal (1991) reported that *C. truncatum* causing serious leaf spotting in greengram, may produce small, reddish brown, 1 to 4 mm diameter spots, later turned to form 'shot holes' and in severe cases, premature defoliation occurred and also brown to reddish blotches on petiole and pods. Similarly, Laxman (2006) observed *C. truncatum* leaf infection in the form of reddish brown chlorotic spot, which later became black. The studies conducted by Asian Vegetable Research and Development Centre, Taiwan (Anon., 1992a) indicated that *C. truncatum* caused brown lesion on leaves, petioles, stem and pods. In the present study also, the symptoms were noticed on leaves, stem and pods.

5.1 Survey and surveillance for disease incidence

In the present study, an intensive roving survey for anthracnose of greengram was carried out during *kharif* 2006 and 2007 in major greengram growing areas of northern Karnataka to get precise information on the distribution and intensity of the disease. The data on survey revealed that the anthracnose severity varied from locality to locality, because of type of variety grown, environmental conditions, cropping pattern and build up of inoculum. The average disease severity varied in various locations in different districts owing to varied agro-climatic conditions and also different cultivars used. In northern Karnataka, the disease severity was found more in Bidar district (49.43%) followed by Gulbarga (48.12%) and Gadag (44.59%) and the least in Bijapur with 23.86 per cent. Such variations in anthracnose severity and wide spread nature have been reported by earlier workers (Saxena and Gupta, 1981; Madhusudhan, 2002 and Laxman, 2006). However, on an average, disease severity was higher during 2007 (38.34%) compared to 2006 (35.53%). The higher incidence of anthracnose during *kharif* 2007 may be attributed to the temperature and relative humidity prevailing during the crop period which were favourable for disease development and spread.

With respect to individual talukas, Humnabad taluka (59.52%) of Bidar district recorded highest disease severity followed by Chincholli taluka (55.54%) of Gulbarga district, wherein conditions for development and spread of the disease were prevailing during *kharif* season. These observations are in agreement with the earlier reports of Varaprasad (2000) in chickpea blight and Laxman (2006) in anthracnose of greengram.

Maximum disease severity of 60.54 per cent recorded in Dubalgundi followed by Hallikhed (B) (60.21%) and Dhummansur villages (59.72%) of Humnabad taluka of Bidar district may be attributed to extensive and continuous cropping of greengram. While, lower disease severity (19.71%) recorded in Halsangi followed by Malaghan village (19.75%) of Bijapur district, wherein unfavourable environment conditions and less availability of infected seed and source of inoculum.

Further, the intensity varied to greater extent in different locations indicating the role of environment and/or existence of physiological races in the pathogen. In general, it is observed that the disease progress in natural conditions was in the second fortnight of July month which coincided with frequent rains with moderate temperature and high relative humidity. These results are in agreement with Ashok Kumar *et al.* (1999) in case of anthracnose of kidney bean and Thakur and Khare (1991) in anthracnose of greengram.

Considering the disease severity, the locations *viz*., Bidar and Gulbarga districts are identified as 'hot spots' for anthracnose of greengram. Intensive cultivation of greengram crop year after year, use of infected seed materials, non-adoption of disease management practices, favourable weather conditions and also the cultivation of highly susceptible varieties of greengram could be the reasons for higher incidence of disease in different locations of Karnataka state.

5.2 Loss assessment

Number of fungicide sprays required to manage the disease

In order to assess the loss due to anthracnose under natural conditions, a susceptible Chinamung variety was used. One to three fungicidal sprays of carbendazim (0.1%) were given in order to estimate the loss due to the disease.

5.2.1 Per cent disease index

Two years data indicated that maximum disease severity was recorded in treatment where no spray of carbendazim was taken in both the years. Whereas, the disease severity was very less in rest of the treatments where one to three sprays of carbendazim were taken. Among the sprayed treatments, one spray treatment recorded higher disease severity of 18.76 per cent than other treatments at all time of observations in both the years. So also, in crop receiving two sprays recorded significantly higher disease index of 16.20 per cent than plots receiving three sprays. The least disease severity was noticed in three sprays (14.83%) followed by two sprays in both the years of experiment and did not differ significantly with each other.

The disease reduction to the tune of 57.87 and 58.08 per cent was observed during kharif 2006 and 2007, respectively on crops sprayed with three times, while it was comparatively less in crop received one spray. Effectiveness of fungicidal sprays to lower the disease advancement is on record (Bharadwaj and Thakur, 1991; Shirshikar, 1995; Varaprasad, 2000; Madhusudhan, 2002 and Laxman, 2006).

AUDPC values gives an idea of diseae progression over a period of time, which inturn reveals the yield loss. In the present investigation, the AUDPC values of anthrcnose reduced considerably with increased number of sprays of carbendazim. These findings are in agreement with those of Benagi (1995), who reported the reduction in AUDPC values of late leaf spot of groundnut with increased number of chlorothalonil sprays. Similar views were expressed by Amaresh (2000) in *Alternaria* leaf blight of sunflower.

The results of the experiments from both the years clearly indicated that two fungicidal sprays are sufficient to reduce the disease severity. Similar views were put forth by Bharadwaj and Thakur (1991), Madhusudhan (2002), Deeksha and Tripathi (2002a) and Laxman (2006).

5.2.2 Grain yield

Application of fungicide significantly improved the grain yield than untreated control (no spray). Maximum yields were obtained from the plots sprayed with two and three times, which were attributed to the lower disease index in these treatments.

The grain yield loss was to the tune of 40.18 per cent in the absence of control measures of the disease. So, by minimizing the anthracnose severity, the yield loss could be reduced considerably. The disease incidence was higher in control plots than fungicide

applied plots, thus indicating relationship between the disease and also yield. The data revealed that, yield was reduced drastically in control plots as compared to fungicide applied plots. The yield was significantly higher in two sprays of carbendazim (12.59 q/ha) during *kharif* 2006 and *kharif* 2007 (12.01 q/ha) than one spray of carbendazim and also disease intensity was low in two sprays of fungicide. These findings are in accordance with the reports of Bharadwaj and Thakur (1991), Madhusudhan (2002), Deeksha and Tripathi (2002a) and Laxman (2006).

Therefore, the results of two years of field studies proved that, two sprays of carbendazim are sufficient to manage the disease and realize the economic yields.

5.2.3 Stalk yield

Similarly, spraying of fungicides significantly increased the stalk yield. The maximum stalk yield were obtained in plots receiving two to three sprays which is mainly due to lower disease index in these treatments. Increase in stalk yields are recorded from plots receiving (14.53 q/ha) two and three (14.67 q/ha) sprays than plots receiving one spray (9.88 q/ha) and untreated control (7.79 q/ha). These findings are in line with the reports of similar views obtained by Benagi (1995) in late leaf spot of groundnut and Deeksha and Tripathi (2002a) in anthracnose of blackgram. Therefore, studies indicated that giving two sprays could help to enhance the stalk yield level in greengram by way of controlling the disease.

5.2.4 Benefit:Cost ratio

Higher benefits were recorded from treatments with one and two sprays of carbendazim (0.1%) against anthracnose in greengram during both the years. This is in agreement with the earlier report of Madhusudhan (2002), Deeksha and Tripathi (2002a) and Laxman (2006). Though, the benefits were same in both the treatments farmers have to loose 2.83 and 4.65 q per ha of grain and stalk yield, respectively if they spray the crop once when compared to two fungicidal applications which are sufficient to overcome the outbreak, but it can not be practical to sacrifice 2.83 and 4.65 q per ha of grain and stalk yield, respectively to obtain higher benefits.

It may be inferred that, two sprays of carbendazim are essential to obtain maximum yield. When disease pressure is moderate to low, one spray of carbendazim (0.1%) is enough to realize maximum benefit. Whereas, when disease pressure is very high, two sprays of carbendazim are necessary to realize maximum benefits. Benefit:cost ratio is important for economic feasibility of farming community.

5.2.5 Crop loss model

Crop loss model plays a vital role in the prediction and forecasting of loss due to anthracnose, which is a pre-requisite for determining decision in thresholds and deployment of cost effective management practices. Hence, importance of disease is adjudged based on the loss in yield caused by the disease. Several workers have indicated the loss in yield (Schneider *et al*., 1976) of greengram crop against leaf spot and groundnut late leaf spot (Benagi, 1995).

Accurate information of loss is needed by farmer and plant protection specialists to develop decision thresholds for determining when cost effective management strategy should be deployed (Nutter, 1993). In the present study, yield loss models were developed by using PDI as input variable to predict loss due to anthracnose. During both the years and in pooled analysis, highly significant correlation coefficient (r) and coefficient of determination (R^2) were obtained for yield predictions with input variable PDI. Hence, the simple linear regression crop loss model was developed which depicts maximum correlation with given PDI and predicted yield. Model developed only helps to calculate the yield with given PDI. As fitting of the model is dependent on maximum R^2, it can fit anywhere from PDI taken at 25 to 60 DAS.

The relationship between PDI and yield were significant and negatively correlated, indicating that increase in per cent disease index leads to decrease in yield of greengram. This is in agreement with Sud and Singh (1985), who reported that the grain yield was negatively related to the severity of leaf spot and positively to green leaf area in case of blackgram. Regression equation showed that, there was a linear relationship between per cent disease index and yield. The fungus obtains nutrients from host and reduces the photosynthetic area of the leaf due to dark brown chlorotic spot on leaf. Thus, photosynthesis and translocation are affected which results in reduction in yield. Similarly, Singh and Shukla

(1988) reported that premature defoliation, affects the yield greatly and pod infection may result in complete loss in yield.

The significant coefficient of determination (R^2) clearly indicated the validity of the model developed. In the present study, the predicted yield loss values were nearer to the observed values and indicated that, the linear regression models fitted by using variable PDI is appropriate for prediction of yield loss due to anthracnose of greengram. Van der Plank (1963) suggested that yield loss may be related to disease and demonstrated the relationship using linear regression equation.

5.3 Cultural and physiological studies

5.3.1 Cultural studies

5.3.1.1 Growth of *C. truncatum* on potato dextrose broth at different incubation period

The growth phase of *C. truncatum* was studied by inoculating the fungus on potato dextrose broth and harvesting the growth starting from third day onwards upto 21st day. The findings of the experiment revealed that there was increase in the dry mycelial weight of fungus from third day onwards upto 15th day. Thereafter, there was a decline in the dry mycelial weight upto 21st day. The maximum dry mycelial weight of 214.13 mg was recorded on the 15th day and it was considered as the optimum period for growth of the fungus. The increase in the mycelial dry weight from third day to 15th day can be attributed to presence of nutrients in the medium and the fungus showed gradual increase in the weight by utilizing them to the maximum extent. The decrease in the dry mycelial weight from 17th day onwards may probably be due to autolysis of the mycelium and exhaustion of nutrients in the medium. This remark is not an exception to the investigation made by Singh and Shukla (1986) and Laxman (2006).

In the present study, the maximum mycelial dry weight was recorded on 15th day. Hence, to obtain maximum fungal growth of *C. truncatum*, 15 days of incubation appears to be optimum.

5.3.1.2 Growth and sporulation of *C. truncatum* on different solid media

Among the various media used for growth and sporulation of *C. truncatum* potato dextrose agar proved to be the best for good growth followed by oat meal agar. Excellent sporulation was seen on both these media. Semi-synthetic media supported better growth due to the presence of some vitamins, which are essential for growth and development of organism (Mathur *et al*., 1950). Similar type of observations were also made by earlier workers (Kenchaiah, 1975; Mesta, 1996; Ekbote *et al*., 1997 and Laxman, 2006). Fungus made the least growth and poor sporulation in potato carrot agar which may be attributed to quality of sugar present in them. Agnihotri and Prasad (1971) reported that sugar supplemented media supported good growth of *C. capsici* than the minimal media used as control.

In the present study, the potato dextrose agar recorded maximum growth and excellent sporulation. Hence, this medium can very well be used for obtaining maximum fungal growth as well as for the excellent sporulation of *C. truncatum*.

5.3.1.3 Growth and sporulation of *C. truncatum* on different liquid media

Several liquid media were evaluated for growth and sporulation of *C. truncatum*. It was found that Richard's medium supported maximum mycelial dry weight (203.17 mg) followed by Czapeck's medium (168.30 mg) and the excellent sporulation was observed in Richard's medium and potato dextrose broth. The ability of a fungus to grow more on Richard's medium indicated the requirement of nutrients present in that medium for *C. truncatum*. Similarly, Ekbote (1994), Mesta (1996), Shirshikar (1995), Angadi (1999) and Varaprasad (2000) reported that maximum growth and sporulation was observed on Richard's medium.

In the present study, the Richard's medium showed maximum growth and excellent sporulation of the fungus. Hence, Richard's medium was used for obtaining maximum growth and sporulation of *C. truncatum*.

5.3.2 Physiological studies

5.3.2.1 Effect of temperature on the growth and sporulation of *C. truncatum*

Temperature plays an important role in infection and disease development. An effort was made to know the optimum temperature for the growth and sporulation. In the present

study, it was observed that maximum fungal growth and sporulation was recorded at 30^0C followed by 25^0C and it was least at 40^0C. The present results are in agreement with the results obtained by Chowdhury (1957) and Wong *et al.* (1983). Similarly, Singh and Shukla (1986) and Laxman (2006) reported that optimum temperature for growth and sporulation of *C. truncatum* was 25^0C to 30^0C.

In the present study, the excellent fungal growth and sporulation was observed at 30^0C followed by 25^0C. Hence, the temperature range of 25 to 30^0C can be recommended to obtain excellent fungal growth and sporulation of *C. truncatum*.

5.3.2.2 Effect of relative humidity on the growth and sporulation of *C. truncatum*

Relative humidity is another important epidemiological factor for influencing fungus as well as the outbreak of the disease. It plays a vital role in development of the disease into an epidemic form. The maximum growth of fungus was noticed at 95 per cent RH (85.05 mm) whereas the minimum mycelial growth was found at 65 per cent RH (60.78 mm). The sporulation was excellent at 85 and 95 per cent RH levels, which play a key role during the disease initiation and dispersal of disease. Similarly, Shirshikar (1995), Varaprasad (2000) and Laxman (2006) reported that maximum growth of fungus was observed at 85 and 95 per cent RH levels.

5.3.2.3 Effect of pH levels on dry mycelia weight and sporulation of *C. truncatum*

Any living organism requires a particular medium for the growth and development. The pH of the media should be optimum for growth. A wide range of pH supported the growth of *C. truncatum*. Good growth was found at a range of 5.5 to 7.0 pH. The sporulation was also influenced by the pH and is known to play a crucial role. In present investigation, the excellent sporulation was found in 6.0 and 6.5 pH and good sporulation was noticed at 5.5 and 7.0 pH. The optimum pH range was obtained towards acidic pH side and sudden decline were observed towards basic pH side which indicated that fungus was acid tolerant. Cochrane (1958) and Bilgrami and Verma (1978) also opined that in contrast to bacteria and actinomycetes fungi are relatively more tolerant to acid ions (H^+) than basic ions (OH^-). The observations are in agreement with those of Singh and Shukla (1986), Shirshikar (1995), Mesta (1996) and Laxman (2006).

In the present study, the maximum dry weight was recorded at 6.5 pH. However, optimum growth of fungus was also observed at pH of 5.5 to 7.0. Hence, it can be recommended that to obtain good growth of *C. truncatum*, the pH level from 5.5 to 7.0 can be maintained in the culture.

5.3.2.4 Effect of light intensities on the growth and sporulation of *C. truncatum*

Light is also playing an important role in disease development. In the present study, the results revealed that alternate cycles of light and darkness, as well as the continuous darkness supported maximum growth and excellent sporulation of the pathogen. However, maximum fungal growth was recorded in the treatment of alternate cycles of light and darkness. The present findings are in agreement with the studies conducted by Wong *et al.* (1983), Sinclair (1988), Shirshikar (1995), Mesta (1996) and Varaprasad (2000). Based on the findings, it can be recommended that for obtaining maximum fungal growth and sporulation, *C. truncatum* culture should be exposed to alternate cycles of 12 hr day light under day light tubes and 12 hr darkness.

5.4 Epidemiological studies

5.4.1 Aerobiology

Studies on aerobiology of the pathogen is important in order to forecast the occurrence of disease and in devising supervisory management practices. In the present investigation, the atmospheric conidial load of *C. truncatum* was studied. In order to find out the conidial load, several techniques have been used from simple glass slides (Mehta, 1940) to Bukard volumetric spore trap (Kulkarni and Ramakrishnan, 1977).

In the present study, the spore trap for stationary slide was used to understand the prevalence of air borne conidia of anthracnose of greengram caused by *C. truncatum* under different weather parameters and primarily to understand the epidemiology of the disease.

5.4.4.1 Effect of weather factors on spore load of *C. truncatum*

The study on spore load of *C. truncatum* was carried out at ARS, Bidar and presented on weekly average in terms of standard week with meteorological data.

Air sampling carried out during 2006 indicated that, the first appearance of spore load in the atmosphere was recorded after 13 days of sowing and maximum spore counts were observed during last week of July and first week of August. While, during 2007, the first appearance of spore was noticed at 12 days of sowing and maximum number of conidia were also recorded during last week of July and first week of August as observed in 2006. The favourable weather conditions such as temperature, relative humidity and rainfall prevailed during these periods. Hence, there was increase in the spore load. These studies are in accordance with Thakur and Khare (1991), they reported that maximum trapping of spores (*C. lindemuthianum* and C. *dematium*) was recorded when there was moderate temperature (26 – 29^{0}C), RH (91 – 96%) and rainfall (0 – 21.6 mm). Highest spore trap coincided with these conditions prevalent on July 30th. Similar results were reported by Ashok Kumar *et al.* (1999).

Temperature was significantly and negatively correlated with weekly spore load during both the years and in pooled analysis also. Higher temperatures are always detrimental for production and germination of spores as it leads to desiccation (Agrios, 1997). Relative humidity and rainfall have positive correlation with weekly spore load. Rain promotes the release of many fungal spores and is of particular significance in release of *Colletotrichum* spores as they are embedded in a gelatinous substance in the acervuli (Hirst and Steadman, 1963). Further, Sinclair and Backman (1989) reported that anthracnose disease developed in soybean after prolonged period of high humidity and that the conidia of the fungus germinated and formed appressoria at temperatures below 35^{0}C when the plant surface was wet. Similarly, Zaumeyer and Thomas (1957) reported that anthracnose fungus required cool temperature ranges for its growth, infection and development.

The variation in disease appearance and development in different localities may also be attributed to the prevalence of favourable weather conditions. The dissemination of conidia is almost exclusively by wind (Hirst, 1953). Thakur (1988) reported that conidia of anthracnose were dispersed very efficiently by air. Air borne concentration follows the pattern of field disease incidence and can be used to assess the severity.

The multiple linear regression equation was fitted to the data and the equation arrived for all the weather parameters during 2006 is, $Y = 344.10 – 2.42 X_1 – 8.32 X_2 – 0.83 X_3 + 0.07 X_4 – 0.07 X_5$ and for 2007 is, $Y = 95.95 + 3.89 X_1 – 12.39 X_2 + 0.52 X_3 + 0.36 X_4 – 0.15 X_5$. The pooled equation of 2006 and 2007 is $Y = 135.85 – 2.55 X_1 – 2.18 X_2 + 0.09 X_3 – 0.04 X_4 – 0.05 X_5$. Hence, it is evident from the data that, all the weather factors influenced the spore load of *C. truncatum* for 2006 to the extent of 90 per cent and 93.00 per cent for 2007 and the pooled years of 2006 and 2007, it has influenced to the extent of 81.00 per cent.

5.4.4.2 Effect of weather parameters on development and spread of the disease

Environmental factors decide the epidemic of anthracnose of greengram. The environmental factors like temperature, relative humidity and rainfall are important for disease development and these environmental factors are being used to forecast disease severity. Further, the knowledge of weather conditions for the development and spread of disease are important to organize Agro Advisory Services for the farmers to take up timely management practices.

During 2006, the anthracnose symptoms were first observed on 25th standard week when the crop was at 19 DAS. The severity increased slowly and reached the incidence of 68.50 per cent during 32nd standard week. During the previous week, maximum temperature of 26.56^{0}C and minimum temperature of 20.57^{0}C with morning relative humidity of 96.00 per cent and evening relative humidity of 79.14 per cent was followed by 173.2 mm rainfall. During 2007, the anthracnose symptoms were first observed on 25th standard week when the crop was at 18 DAS. The rainfall of 80.1 mm with minimum temperature of 20.54^{0}C and maximum temperature of 30.09^{0}C, morning relative humidity of 94.00 per cent and evening relative humidity of 60.71 per cent prevailed during previous week. Severity increased slowly and reached maximum incidence of 70.12 per cent during 32nd standard week. The favourable conditions such as maximum temperature of 29.03^{0}C and minimum temperature of

20.77^{0}C with 91.86 per cent morning relative humidity and 63.86 per cent evening relative humidity and rainfall of 6.6 mm were noticed during that standard week.

In general, frequent rains with moderate temperature (26 – 30^{0}C) and high relative humidity (85 – 96%) are essential for initiation and spread of the disease. The findings are in conformity with Ashok Kumar *et al.* (1999), who reported that maximum anthracnose disease development was noticed after mid July to Mid August, when weather variables *viz.*, temperature (19 – 20^{0}C), relative humidity (74 – 77%), rainfall (0.2 – 166.9 mm) and frequency of rains (2 – 5 days) were congenial in kidneybean. Two consecutive days of rain accompanied by cloudiness and high humidity are necessary for infection of *C. truncatum* in bean (Chambers, 1969). Further, Thakur and Khare (1991) reported excessive spore production when there was moderate temperature (26 – 29^{0}C), RH (91 – 96%) and rainfall (0 – 21.6 mm). Further, they noticed maximum increase in lesion size of greengram anthracnose when the relative humidity was 100 per cent followed by temperature 27^{0}C.

The analysis was made to establish the relationship between weather factors and per cent disease index of anthracnose in highly susceptible variety Chinamung through correlation and multiple linear regression analysis. The relationship between anthracnose PDI and weather factors during 2006 indicated higher negative correlation between maximum and minimum temperature but positive correlation with morning and evening relative humidity and rainfall. During 2007, also similar trend was noticed except rainfall which showed negative correlation. The results are in agreement with Chambers (1969), Thakur (1988), Thakur and Khare (1991) and Ashok Kumar *et al.* (1999).

The multiple linear regression equation was fitted to the data and the equation arrived for all the weather parameters during 2006 is, $Y = 1349.88 + 16.37\ X_1 – 75.27\ X_2 – 2.72\ X_3 + 0.92\ X_4 + 0.09\ X_5$ and for 2007 is, $Y = -1885.52 + 34.81\ X_1 – 21.93\ X_2 + 15.15\ X_3 + 0.02\ X_4 – 0.90\ X_5$. The pooled equation of 2006 and 2007 is, $Y = 579.69 – 2.42\ X_1 – 22.06\ X_2 + 0.69\ X_3 + 1.01\ X_4 + 0.01\ X_5$. Hence, the weather factors influence the disease incidence for 2006 to the extent of 98 per cent and for 2007 to the extent of 89 per cent, then for the pooled data, to the extent of 57 per cent.

5.4.4.3 Disease prediction

Weather factors play an important role in the disease development when the vulnerable host and virulent pathogen coincide in a situation. Since, weather parameters influence the development and further spread of disease. Hence, an attempt was made to predict the severity of anthracnose using 2nd degree polynomial function model. The 2nd degree polynomial function model for the *kharif* 2006 and *kharif* 2007 are as under.

$Y = -4.90 + 5.37\ X + 0.33\ X^2$ for *kharif* 2006 with $R^2 = 0.99$

$Y = -7.60 + 7.87\ X + 0.11\ X^2$ for *kharif* 2007 with $R^2 = 0.99$

The models had highest coefficient of determination values with 99 per cent during 2006 and 2007. Since, the coefficient of determination is 99 per cent, it is appropriate to employ the 2nd degree polynomial function model for estimating the development of anthracnose infection. Similar type of prediction method for development and progress of *Alternaria* blight of sunflower was developed by Mesta (2006).

5.4.5 Effect of date of sowing on incidence of anthracnose

Alternation of the date of sowing of crop always plays an important role in disease escape due to unfavourable weather conditions for infection. Field experiment was undertaken to determine the effect of sowing time and corresponding weather factors on anthracnose severity. The results obtained during both years revealed that, the crop sown during 4th June to 11th June recorded lesser incidence of anthracnose which reflected on obtaining more grain yield of greengram compared to the crop sown during 18th June and subsequent weeks. The late sown crop suffered more because of coincidence of the favourable period like moderate temperature coupled with higher humidity and frequent rains with stage of the crop. Chambers (1969) reported that amount of rain was found to be of less importance than prolonged wetness with high humidity which are necessary for infection by *C. truncatum* in bean. The results are similar to Mittal (1998), who reported that early sown blackgram crop suffered least due to low inoculum potential and unfavourable weather conditions for pathogen, whereas late sown crop suffered more because of ready availability of inoculum build up in early sown crop. Similar observations were made by Naidu and

Chandrika (1997) in case of leaf spot of groundnut and Das (2005) in foliar diseases of greengram.

In the present study, the crop sown on first fortnight of June recorded minimum disease severity compared to rest of the dates of sowings. This clearly indicated that crop sown during this period suffers less, which may be due to low inoculum potential, whereas the late sown crop suffers more because of the readily available inoculum in the early sown crops. Low disease severity in first fortnight sowing may be attributed to the non-congenial weather factors (higher temperature coupled with lower humidity) for the development of the disease.

5.4.6 Survival of *C. truncatum* in debris and seed

5.4.6.1 Survival of conidia of *C. truncatum* under different storage conditions

As a part of epidemiological study, the survival of pathogen in host debris was studied. The study was conducted to know the survival ability of the conidia at different storage conditions. The present study revealed that conidia survived for 90 days in infected leaflets under field or natural conditions (28 – 30^{0}C) and upto 120 days in glasshouse conditions (25 – 28^{0}C). However, the conidia survived for a maximum of 360 days under freeze (4 – 5^{0}C) conditions. Germination of conidia was observed upto 210 days at room temperature (20 – 25^{0}C) and 240 days under tree shade (18 – 22^{0}C). However, no germination was observed after 360 days of storage. This finding is in accordance with Chona and Nariani (1954) and Varaprasad (2000).

Further, per cent viability of conidia decreased with increase in storage period in all the conditions tested. The viability of conidia was observed upto 120 days of storage at glasshouse (25 – 28^{0}C) condition and upto 210 days at room temperature (20 – 25^{0}C) and 240 days under tree shade. Whereas under freeze (4 – 5^{0}C) conditions, the viability of conidia was upto 360 days but under field conditions, it was least upto 90 days. These findings are in line with Tu (1983), who reported that longevity of *C. lindemuthianum* varied greatly depending on environmental conditions. Moisture had a profound effect on its longevity. The fungus survived at least five years in infected pods of bean that were air dried and kept in storage at 4^{0}C. Under natural field condition, there is rapid decrease in viability of conidia in wet conditions which may be attributed to the loss of the mucilaginous water soluble matrix of the condia.

Germination of infectious propagules is an important process in life cycle of pathogenic fungi and disease development, as host penetration and infection depends on this process. More and quick germination also plays a vital role in faster development and spread of the disease. This has a significant role in epidemiology of the disease.

5.4.6.2 Survival of *C. truncatum* on greengram seed

The survival of *C. truncatum* in infected greengram seeds was tested by periodic sampling of greengram seeds starting from 30 days onwards upto 360 days. The samples were assayed by standard blotter technique for detection of *C. truncatum* infection level in each sample. It was observed that *C. truncatum* fungus recorded 23.50 per cent survival in the 30 days old seed sample. Later, there was a gradual decrease in the survivability of fungus. The lowest per cent survivability of 7.25 was recorded at 360 days old sample indicating the survival of *C. truncatum* even in 360 days old seeds. The germination percentage gradually increased with the increase in the storage period and reached upto 87.00 per cent after 360 days of storage. These studies were in accordance with Rajkumar *et al.* (1989) and Deeksha and Tripathi (2002b), wherein they reported the decrease in survivability of the *C. capsici* through blackgram seeds with increase in storage period and a corresponding increase in germination of seeds with time. Decline in survivability may be due to factors like depletion of nutrients and moisture in seeds as well as poor saprophytic ability of the pathogen. Increase in germination of seeds with time might be a result of decline in survivability of fungus with increase in storage period.

The present observations indicate the potentiality of greengram seed as a carrier of primary inoculum. This study suggests that since *C. truncatum* can survive for longer period, the care is needed to be taken while exchanging the greengram seeds for sowing purpose.

5.4.7 Effect of age of plant in relation to infection of *C. truncatum*

Age of the plants is important for development of disease. In the present study the per cent disease severity on 5 day old seedlings was to the tune of 3.40, later increased gradually. However, the maximum disease severity of 68.92 per cent was observed on 40 day old plants, followed by 35 days (62.05%) and 45 days (52.20%), further, the disease severity decreased to 22.87 per cent on 50 day old plants. Earlier work carried out by Fuse *et al*. (1981), Sinclair (1982), Rahman and Fakira (1985), Shirshikar (1995) and Madhusudhan (2002) have indicated that the soybean plant is more susceptible to *C. truncatum* in its early growth stage to bloom stage, which is also evident in the present investigation.

The experimental findings of this study emphasizes the need of protecting greengram plants against *C. truncatum* during flowering stage and also indicated the suitability of flowering stage for artificial screening of the greengram genotypes.

5.4.8 Host range studies

The host range study was conducted to know the ability of *C. truncatum* to survive on different hosts in the absence of main host. This would help to locate source of initial inoculum of pathogen in the disease cycle and also in developing suitable control measures. Occurrence of *C. truncatum* and other *Colletotrichum* sp. on the minor pulses like blackgram, greengram and horsegram has been reported (Rath and Routray, 1978; Saxena and Gupta, 1979 and Rangaswami *et al*., 1991). The present study conducted on pathogenicity of *C. truncatum* obtained from greengram, to other pulses like blackgram, soybean, horsegram, cowpea, redgram, bengalgram, frenchbean and clusterbean revealed that only two pulses *viz*., blackgram and horsegram showed the positive reaction and all other pulses showed negative reaction to the greengram isolate of *C. truncatum* indicating its host specificity. Bharadwaj and Singh (1986) in their studies isolated *C. truncatum* from soybean, greengram, blackgram and horsegram and observed pathogenic behaviour of these four isolates on six leguminous crops *viz*., greengram, blackgram, horsegram, soybean, cowpea and adzukibean. Based on the pathogenicity test, they concluded that all these four isolates differed from each other in their pathogenic behavour and this differed pattern in their pathogenicity could be attributed to the existence of different pathogenic variants of *C. truncatum* in nature and their specific adoption to host species. Manandhar *et al*. (1988) have reported that variation exists among the isolates of soybean *C. truncatum* fungus. Sinclair and Backman (1989) also suggested that *C. truncatum* vary considerably in pathogenicity. Similarly, Madhusudhan (2002) reported that *C. truncatum* obtained from soybean could infect frenchbean, weeds like *Amaranthus viridis, Physalis minima, Solanum nigrum* and *Acalypha indica*.

In the present study, the *C. truncatum* obtained from greengram could infect blackgram and horsegram. Thus, they are the hosts of *C. truncatum* infecting greengram. These findings are in agreement with Bharadwaj and Singh (1986) and Varaprasad (2000).

5.4.9 Cross inoculation study

In the present cross inoculation study, two hosts *viz*., blackgram and horsegram showed positive reactions. The anthracnose pathogen was cross inoculable from greengram to those hosts and vice-versa. This has proved their role as alternative hosts of *C. truncatum* infecting greengram. These findings are in line with McLean and Roy (1988) studies where in they collected weed samples *viz*., prickly sida and spotted spurge and smooth pig weed infected with *C. truncatum* from soybean field and the cross inoculation study revealed that isolate of *C. truncatum* from prickly sida and spotted spurge were pathogenic to soybean seedlings, but the isolate from smooth pig weed failed to infect soybean. They opined that smooth pig weed isolate may be considered as sub-species of *C. truncatum*. Simialr views were expressed by Hartman *et al*. (1986). It can thus be concluded that, the perennation of *C. truncatum* is by way of infected collateral hosts, which might serve as primary source of inoculum.

5.5 Screening of genotypes against anthracnose and biochemical factors of resistance

5.5.1 Screening of greengram genotypes under artificial conditions against anthracnose

In the present investigation, 30 greengram genotypes were screened to find out the sources of resistance against *C. truncatum*. The studies indicated that none of the genotypes

screened showed immune reaction against the pathogen. Only two genotypes *viz*., TM-96-2 and TARM-18 were found resistant, three genotypes *viz*., BGS-9, TM-98-50 and TM-97-55 showed moderately resistant reaction. Two genotypes *viz*., Pusa baisaki and Sel-4 were found susceptible and 23 genotypes showed highly susceptible reaction. Attempts have been made in the past in India and abroad by various workers (Khare and Chacko, 1983; Wong *et al*., 1983; Manandhar *et al*., 1988; Madhusudhan, 2002) to locate the source of resistance against *C. truncatum*. These studies have revealed that very few genotypes showed resistance against *C. truncatum*.

The present study has revealed that the cultivars TARM-18 and TM-96-2 have shown resistant reaction to the pathogen. These cultivars may be further evaluated for their yielding ability and may be released for cultivation or may be utilized in the resistance breeding programme.

5.5.2 Screening of promising varieties against anthracnose under field conditions

The management of disease through host plant resistance is important component in all the crop improvement programmes. Utilization of resistant cultivars in farming is the most simple, effective and economical method in the management of diseases. Besides these, the resistant cultivars conserve natural resources and reduce the cost, time and energy when compared to other methods of disease management. In the present study out of six promising varieties of greengram screened, only one variety *viz*., TARM-18 was found resistant in both 2006 and 2007, whereas TM-96-2 was found resistant in 2006 but showed moderately resistant reaction in 2007. Agarwal (1989) reported that the reaction of blackgram and greengram genotypes against the anthracnose pathogens is not consistent. Whereas, BGS-9 variety showed moderately resistant reaction in both years, others showed highly susceptible reaction. Thakur and Khare (1989) evaluated 27 cultivars of greengram against anthracnose in the field and found that Pusa 109 was highly resistant, while ML-33 showed resistant reaction and other showed susceptible reaction. Similarly, Rathaiah and Sharma (2004) reported that in greengram the cultivars *viz*., MLTG-2 and TARM-18 were highly resistant to *C. truncatum*.

The present study has revealed that the cultivars TARM-18, TM-96-2 and BGS-9 have shown resistant to moderate resistant reaction which may be used by breeders to evolve high yielding greengram varieties.

5.5.3 Biochemical factors of resistance to anthracnose of greengram

The common biochemical constituents like chlorophyll, sugars and phenols are important in imparting resistance to the crop plants. Sometimes, host plant is induced to synthesize these compounds upon infection. In the present study taking two genotypes from each category of resistant, moderately resistant and three genotypes from susceptible groups, an attempt has been made to find out the role of some biochemical constituents present in greengram plants in imparting resistance.

5.5.3.1 Chlorophyll content

In the present study, chlorophyll ‘a’, ‘b’ and total chlorophyll were recorded in higher amounts in resistant cultivars than susceptible cultivars. Their relative concentration was also decreased as a result of disease, but the rate of decrease was higher in susceptible varieties than resistant varieties. Abnormalities in the form and destruction of chloroplasts are common features of diseased tissue in plants infected with pathogens, which usually exhibit reduced photosynthetic rate, phosphorylation, hill reaction and carbon dioxide assimilation (Bawden, 1999). These changes may be partially or completely accounted by reduction in chlorophyll content. In the susceptible cultivars more reduction in chlorophyll content may be due to death of more leaf tissues due to infection of *Colletotrichum*. Also in the present study, chlorophyll ‘b’ was found more sensitive than chlorophyll ‘a’ as it recorded higher reduction percentage after inoculation of *C. truncatum*.

The results are in conformity with Benagi (1995) in leaf spot of groundnut. Rajiv Kumar and Singh (1996), Amaresh (2000) and Mesta (2006) in *Alternaria* blight of sunflower.

5.5.3.2 Sugar content

In general, the infection by some pathogens bring about changes in respiratory pathway and photosynthesis, which are important vital processes taking place inside the plant

leading to wide fluctuations in sugars (Farkars and Kiraly, 1962 and Klement and Goodman, 1967). In susceptible varieties, disease development was more whereas the mean sugar content came down at later part of the crop growth. This indicated the utilization of these sugars by the invaded pathogens for their nutrition. Such nutritional utilization of sugars by the invading pathogens has been reported earlier also (Krog *et al*., 1961).

In the present study, total sugar, reducing sugar and non-reducing sugar contents were higher in healthy leaves of susceptible genotypes than of resistant ones. Further, in diseased leaves their amount decreased in both resistant and susceptible genotypes. The results are in conformity with Sindhan and Parashar (1996) in leaf spot of groundnut and Sindhan *et al.* (1999) in leaf spot of mungbean. The reduction in carbohydrate contents after infection may be due to rapid hydrolysis of sugars during pathogenesis through enzymes secreted by pathogens and subsequent utilization by pathogens for their development (Jaypal and Mahadevan, 1968). The present study revealed that the low amount of carbohydrates in resistant genotypes may be responsible for resistance of genotypes against anthracnose of greengram.

5.5.3.3 Phenols

A definite correlation exists between the resistance of plants to pathogens and the state of their phenolic complex (Hare, 1966 and Kosuge, 1969). For realization of their protective action, phenolic compounds must be liberated from inactive forms, since they are precisely in the free state (Friend, 1979). Among all the biochemical components in imparting resistance to several plant diseases, high concentration causes an instant lethal action by a general tanning effect while, low concentration causes gradual effect on the cellular constituents of the parasite. If the concentration does not occur in toxic level, the inhibition will be obviously slow. Besides, the pathogens readily detoxify low concentrations of the toxicant rather than high concentrations (Dasgupta, 1988).

In the present investigation, the healthy leaves of resistant and moderately resistant genotypes contained higher amount of total phenol and O.D. phenol than susceptible one. In diseased leaves their amount increased in all groups of genotypes, but this increase in phenol content was at higher rates in resistant and moderately resistant genotypes while it was at lower rates in susceptible genotypes. Concentration of phenolic compounds was usually higher in resistant genotypes than susceptible genotypes of different crop plants (Arora and Wagle, 1985 and Saini *et al*., 1988). The results of present investigation on phenols are in agreement with the findings of Gupta *et al.* (1985), Benagi (1995), Sindhan and Parashar (1996) and Sindhan *et al.* (1999), who reported that total phenols increased due to infection by *Cercospora personatum* in resistant varieties as compared to susceptible varieties of groundnut. The higher amount of phenolic compounds in diseased leaves may be due to either enhancement of synthesis or translocation of phenolics to the site of infection or hydrolysis of phenolic glycosides by fungal glycosides to yield free phenols, which helped in arresting the spread of the pathogen (Sharma *et al*., 1983). The present study revealed that high amount of phenolic compounds in resistant genotypes may be responsible for resistance of genotypes against anthracnose of greengram.

5.6 Disease management

5.6.1 *In vitro* evaluation of fungicides

In vitro evaluation of fungicides provides useful and preliminary information regarding efficacy of fungicides against pathogen within a shortest period of time and therefore, serves as a guide for field testing. In the present investigation, eight systemic fungicides and five non-systemic fungicides were tested at three concentrations. Among the eight systemic fungicides propiconazole, carbendazim, thiophanate methyl and benomyl were the best in inhibiting (100%) the growth of *C. truncatum* at all the three concentrations (0.05, 0.1 and 0.15%) and next best were hexaconazole and tricyclazole (100%) at 0.1 and 0.15 per cent. Efficacy of these fungicides was previously reported by Varaprasad (2000), Madhusudhan (2002) and Laxman (2006).

Among the five non-systemic fungicides tested carbendazim + mancozeb (SAAF) was found to be effective in inhibiting the growth of mycelium upto 100 per cent at all three concentrations (0.1, 0.2 and 0.25%). The next best was chlorothalonil (100%) at 0.25 per cent. These results are in accordance with Mesta (1996), Hegde (1998) and Madhusudhan (2002).

5.6.2 *In vitro* evaluation of botanicals

At present, plant extracts are gaining importance in plant disease management practices. These are the cheaper and safer means of disease management which reduce not only toxicity hazards but also present ecofriendly approach in nature.

In the present investigation though the complete inhibition of the fungus was not observed in any of the ten botanicals used, considerable amount of inhibition of growth was noticed in some of the botanicals. Herbal products *viz*., azadirachtine was found to be effective followed by eucalyptus oil and to some extent garlic bulb extract. The fungicidal spectrum of neem (*Azadirachta indica*) has already been investigated by Singh and Pande (1966) and reviewed in detail by Praveen and Alam (1993). Further, Shivapuri *et al.* (1997), found neem, garlic and *Datura stramonium* most effective against *C. capsici*. Similarly, Angadi (1999) reported that nimbicidine and NSKE showed considerable amount of inhibition of *C. capsici*. Later, Laxman (2006) observed the effectiveness of eucalyptus oil, garlic and neem against *C. truncatum* in greengram.

5.6.3 *In vitro* evaluation of bioagents

Biological control through the use of antagonistic microorganisms is a potential non-chemical means of controlling plant disease by reducing inoculum levels of the pathogens. Such a management would help in preventing the pollution and also health hazards.

In the present investigation, the antagonistic effect of different bioagents was assessed against *C. truncatum* by dual culture technique. Among the different bioagents evaluated *Trichoderma harzianum* has inhibited the growth of fungus with maximum extent followed by *Gliocladium virens* and *T. koningii*. Gupta *et al*. (1991) reported that *Gliocladium virens*, *T. harzianum* and *T. viride* significantly inhibited growth of *C. lindemuthianum in vitro*. The present investigations are in agreement with Varaprasad (2000), who found effectiveness of *Trichoderma* sp. against *Colletotrichum dematium*, whereas Laxman (2006) against *C. truncatum*. This could be obviously due to several possibilities of existence of microbial interactions such as stimulation, inhibition, mutual intermingling of growth of antagonistic isolate over test pathogen *etc.* have been enumerated by many workers (Porter, 1924, Ghaffar, 1969 and Naik and Sen, 1995).

5.6.4 *In vitro* evaluation of ITK's

Indigenous knowledge reflects the body of knowledge evolved in local environment that was creative and experimental in nature and involved minimum risk to rural families and conserved local resources. In the present investigation, five different ITK's are evaluated against spore germination of *C. truncatum* at different concentrations. All the ITKs used have shown antifungal activity but the extent of activity varied with different ITK dilutions and their interactions. However, the maximum inhibition of spore germination was observed in cow urine followed by fermented butter milk and panchagavya at 1:2 dilution. Whereas, cow urine and butter milk treatments have shown on par relation with 1:5 and 1:10 dilutions.

The results of the present study was supported by Priya and Kuruchcve (2005) who studied the effect of animal excrements on conidial germination of *Cercospora personata*. Sapre and Verma (2006) reported that cow urine and butter milk reduced mycelial growth, number and size of sclerotia of *Rhizoctonia bataticola*. Sumangala and Patil (2007) reported that panchagavya an organic formulation evaluated *in vitro* for its antifungal activity against *Curvularia lunata* in rice, resulted in 86.30 per cent inhibition of mycelial growth and 95.9 per cent of spore germination. Similarly, Mahantesh (2008) reported the effectiveness of cow urine against *Phakopsora pachyrhizi* in soybean.

5.6.5 Integrated disease management

The fungicides and biorationals which were found effective in the laboratory condition were evaluated under field conditions. In the present investigation, it is evident that, foliar spray of propiconazole and foliar spray of hexaconazole followed by benomyl seed treatment + foliar spray of benomyl and carbendazim seed treatment + foliar spray of carbendazim were effective in minimizing the per cent disease index and getting higher grain and stalk yields. Deeksha and Tripathi (2002c) and Laxman (2006) reported propiconazole, hexaconazole and carbendazim were effective fungicides against anthracnose of blackgram and greengram,

respectively. Madhusudhan (2002) observed that either benomyl or carbendazim seed treatment along with two foliar applications was found effective in controlling the soybean anthracnose. There were similar reports regarding effectiveness of benomyl and carbendazim in controlling *Colletotrichum* sp. (Bharadwaj and Thakur, 1991; Shirshikar, 1995; Singh *et al*., 1999 and Varaprasad, 2000).

The biorationals were found less effective in controlling the disease as compared to chemical and did not enhance the yield. Among the biorational, the least incidence of anthracnose was noticed in *Trichoderma harzianum* seed treatment + foliar spray of azadirechtine. The present findings are in accordance with Chandrasekaran *et al.* (2000), Varaprasad (2000) and Laxman (2006), who reported the ineffectiveness of plant products and bioagents over the fungicides in controlling anthracnose of soybean, chickpea and greengram, respectively. The low effectiveness of the bioagents might be attributed to the low level of relative humidity prevailing in the field (Belanger *et al*., 1994).

In the present investigation, as far as disease control and yields are considered, triazoles *viz*., propiconazole and hexaconazole performed better as compared to conventional fungicides. These findings are in conformity with those of Deeksha and Tripathi (2002c), Madhusudhan (2002) and Laxman (2006) in case of anthracnose of blackgram, soybean and greengram, respectively. From the practical point of view, the chemical, which gives the maximum returns is more important rather than the control of the disease. So calculation of benefit:cost ratio gives an information on whether the technology could be adopted in the farmers fields or not. Hence, benefit:cost ratio is an important parameter for recommendation of any treatment for successful control of plant disease. In the present study, though the treatment containing foliar spray of propiconazole gave significant control of anthracnose, maximum cost:benefit ratio of 23.10 was realized in treatment containing foliar spray of hexaconazole followed by carbendazim seed treatment + foliar spray of carbendazim (22.62), foliar spray of propiconazole (14.16) and benomyl seed treatment + foliar spray of benomyl (12.73). This clearly indicated that one foliar spray hexaconazole (0.1%) was more useful not only in reducing the cost of protection but also gave higher benefits as compared to other treatments and can be recommended for the management of greengram anthracnose. This is followed by carbendazim seed treatment + foliar spray of carbendazim, foliar spray of propiconazole and benomyl seed treatment + foliar spray of benomyl treatments. Similar types of findings were observed by many workers (Bharadwaj and Thakur, 1991; Shirshikar, 1995; Madhusudhan, 2002 and Laxman, 2006). Hence, spraying of hexaconazole (0.1%) could be considered as an effective management practice to manage anthracnose of greengram. Integration of moderately resistant genotypes (BGS-9, TM-98-50 and TM-97-55) coupled with hexaconazole spray will be effective in reducing the disease pressure and enhancing the yields of greengram.

5.7 Future line of work

1. Survey and surveillance of the disease have to be undertaken every year in future to observe rhythmic changes in the severity of the disease and also the status and regional severity of the disease.
2. Collateral hosts need to be critically observed during off season for the survival and perpetuation of conidia of *C. truncatum*.
3. Qualitative and quantitative estimation of spore load of *C. truncatum* and validation of forecasting models through air sampling by using Burkard volumetric spore trap.
4. The epidemiological studies can be continued for at least 7 – 8 years, so that a software package can be developed for prediction of disease at any given time using input variables. The different prediction models can be tested over a period of time for validity.
5. There is need for studies on variability in *C. truncatum*.
6. Utilization of resistant entries in disease resistant breeding programme and molecular characterization of genes responsible for resistance in the resistant varieties.

6. SUMMARY AND CONCLUSIONS

Greengram (*Vigna radiata* L. Wilczek) is an important pulse crop of India. Anthracnose caused by *Colletotrichum truncatum* (Schw.) Andrus and Moore is the most important fungal disease which is found world wide. The disease is prevalent in greengram growing areas of Karnataka and is considered as a limiting biotic factor for successful cultivation of greengram. In recent past, it has become a major menance for greengram cultivation in India in general and northern Karnataka in particular. Hence, systematic studies on survey and surveillance, cultural and physiological characters of the pathogen, crop loss assessment, epidemiology and management aspects of the disease were carried out and results thus obtained are summarized hereunder.

The disease was characterized by the appearance of reddish brown lesions on lower surface, later they turned to deep dark brown and became chlorotic forming the lesions on both surfaces. At later stage, the same lesions appeared on the petioles, stem and also on the pods. In severe case premature defoliation occurs.

The mycelium was septate, branched, brown and 3 to 4 µ in thickness. The acervuli were black in colour, oval to conical in shape and measuring 171.5 × 248.0 µm. Setae were longer than conidiophore and measured 78.0 to 201.0 × 5.0 to 7.1 µm. Conidia were single celled, smooth walled, hyaline, curved and measured 20.0 to 23.3 × 3.5 to 4.0 µm in size.

The disease is widely distributed in Karnataka. Survey conducted during *kharif* 2006 and *kharif* 2007 revealed that the disease severity was found more in Bidar district (49.43%) followed by Gulbarga (48.12%) and Gadag (44.59%) and least in Bijapur (23.86%). With respect to individual talukas, Humnabad taluka (59.52%) of Bidar district recorded highest disease severity followed by Chincholli taluka (55.54%) of Gulbarga district. Indi taluk in Bijapur district recorded least disease severity (23.02%) in both the years.

The observations on loss estimation revealed that comparatively lower disease index with increase in grain and stalk yield and also maximum BCR was recorded in plots receiving two sprays of carbendazim. The average grain yield loss of 40.18 per cent and stalk yield loss of 46.90 per cent were noticed due to anthracnose in unsprayed plots. Yield loss models were developed using PDI as input variable and are given below.

Y = 15.46 – 0.24 PDI with r = 0.84 (*Kharif*, 2006)

Y = 14.83 – 0.21 PDI with r = 0.89 (*kharif*, 2007)

Pooled yield loss model obtained as Y = 15.14 - 0.23 PDI with r = 0.87

The growth phase study on *C. truncatum* revealed that the fungus produced maximum dry mycelial weight of 214.13 mg on 15th day in potato dextrose broth, beyond which autolysis occurred.

Cultural studies conducted revealed that among the solid media, potato dextrose agar was best followed by oat meal agar for the growth and excellent sporulation of the *C. truncatum*. Among the various liquid media used, maximum dry mycelial weight and excellent sporulation was observed in Richard's medium followed by Czapeck's medium.

The optimum range of temperature and pH levels for the fungus were 25^{0}C to 30^{0}C and 5.5 to 7.0, respectively. However, maximum growth and sporulation of fungus was recorded at 30^{0}C temperature and 6.5 pH.

In studies on relative humidity and light requirements for growth and sporulation of *C. truncatum* in culture, it was found that relative humidity of 85 to 95 per cent with alternate cycles of 12 hr day light under day light tubes and 12 hr darkness supported good growth and sporulation.

The aerobiological studies on effect of weather factors on the development of spore load of *C. truncatum* indicated that more conidial counts were observed during last week of July and first week of August, which coincided with the critical stages of infection.

The correlation studies between spore load and weather parameters indicated a negative correlation with temperature, while relative humidity and rainfall have positive correlation with spore load. Through pooled analysis of *kharif* 2006 and 2007, the multiple linear regression model obtained as $Y = 135.85 – 2.55 X_1 – 2.18 X_2 + 0.09 X_3 – 0.04X_4 – 0.05 X_5$ with R^2 of 0.81.

The studies on effect of weather factors on development of disease revealed that per cent disease index (PDI) was progressing at linear rate throughout the plant growth and it was negatively correlated with temperature, while positively correlated with relative humidity and rainfall. The multiple regression model developed for PDI is $Y = 579.69 – 2.42 X_1 – 22.06 X_2 + 0.69 X_3 + 1.01 X_4 + 0.01 X_5$ with R^2 value of 0.57.

The 2nd degree polynomial function models development for *kharif*, 2006 and *kharif* 2007 are as under.

$Yx = -4.90 + 5.37 X + 0.33 X^2$ for *kharif* 2006 with $R^2 = 0.99$.

$Yx = -7.60 + 7.87 X + 0.11 X^2$ for *kharif* 2007 with $R^2 = 0.99$.

The models had highest coefficient of determination values with 99 per cent during 2006 and 2007.

The study on date of sowing revealed that the greengram crop sown on 4th June showed least disease severity followed by crop sown on 11th June. Whereas, crop sown during 18th June and subsequent weeks, suffered a lot due to anthracnose. Temperature was negatively correlated with disease severity, while relative humidity and rainfall were positively correlated. The coincidence of the favourable period with stage of the crop led to considerable boost in disease incidence.

Viability of conidia of *C. truncatum* was observed upto 360 days in freeze condition followed by tree shade condition (240 days) and room temperature (210 days). Under glasshouse condition, viability was noticed upto 120 days and at field condition, it was upto 90 days.

The survival of *C. truncatum* in greengram seeds was studied by drawing seed samples at regular intervals starting from 30 days upto 360 days. It was observed that 30 days old seed samples had 23.50 per cent survival when compared to 360 days old samples which recorded the survival of 7.25 per cent, indicating the survival of fungus upto 360 days. The germination percentage gradually increased with the increase in the storage period and reached upto 87.00 per cent after 360 days of storage.

The relation of greengram plant age to anthracnose disease severity revealed that, flowering stage was found susceptible to the disease development. Maximum disease severity of 68.92 per cent was recorded on 40 days old seedlings.

Out of nine pulses tested for host range, two hosts *viz*., blackgram and horsegram developed symptoms and acted as hosts of *C. truncatum*. Cross inoculation studies indicated that, inoculum from greengram, infected blackgram and horsegram and vice-versa. These hosts may act as collateral hosts and help in perpetuation of the pathogen.

Screening of thirty genotypes under greenhouse condition revealed only two genotypes *viz*., TM-96-2 and TARM-18 as resistant, three genotypes *viz*., BGS-9, TM-98-50 and TM-97-55 as moderately resistant reaction, two genotypes *viz*., Pusa baisaki and Sel-4 as susceptible and 23 genotypes as highly susceptible to *C. truncatum*. Whereas, in screening of six promising varieties under field condition indicated that TARM-18, TM-96-2 and BGS-9 were resistant to moderately resistant.

The biochemical studies indicated that the chlorophyll and sugar content was found to decrease due to the infection of *C. truncatum* and the rate of decrease was more in susceptible genotypes than resistant genotypes. However, phenol content was found increased due to infection. Also, the rate of increase was higher in resistant genotypes than in susceptible genotypes. Comparatively lesser sugar and higher phenol content were observed in resistant and moderately resistant varieties than in susceptible variety.

Out of eight systemic fungicides tested *in vitro* against *C. truncatum*, propiconazole, carbendazim, thiophanate methyl and benomyl were best in inhibiting (100%), the growth of fungus at all the three concentrations tested (0.05, 0.1 and 0.15%) and next best was hexaconazole (100%) at 0.1 and 0.15 per cent. Carbendazim + mancozeb (Saaf) was found to be superior among the non-systemic fungicides by inhibiting 100 per cent at all three concentrations (0.1, 0.2 and 0.25%).

Among ten botanicals evaluated *in vitro* azadirachtin (63.34%), eucalyptus oil (60.62%) and garlic (59.44%) were found most promising ones which showed higher mycelial growth inhibition at 10 per cent concentration.

In vitro evaluation of bioagents revealed that, *Trichoderma harzianum* inhibited the growth of fungus with maximum extent followed by *Gliocladium virens* and *T. koningii*. Among ITKs, maximum inhibition of spore germination was observed in cow urine followed by fermented butter milk and panchagavya at 1:2 dilution.

Studies on integrated approach for the management of greengram anthracnose disease under field conditions revealed that out of 17 treatments, foliar spray of propiconazole was found to be effective in control of the disease and increased the grains and stalk yields but foliar spray of hexaconazole showed maximum BCR followed by carbendazim seed treatment + foliar spray of carbendazim than propiconazole and was also found to be effective in managing anthracnose of greengram and increased the grain and stalk yields.

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** Originals not seen*

APPENDIX

Appendix I: Weekly meteorological data recorded at ARS, Bidar during *kharif*, 2006

Standard weeks	Month and Date	Temperature (^{0}C)		Relative humidity (%)		Rainfall (mm)
		Maximum	Minimum	Morning	Evening	
23	June 4 – 10	33.37	22.29	84.86	40.71	1.36
24	June 11 – 17	34.51	23.60	77.43	37.14	4.4
25	June 18 – 24	32.49	22.37	89.57	55.86	20.2
26	June 25 – 01	31.26	21.91	92.86	58.00	18.4
27	July 02 – 08	30.23	21.66	90.71	62.43	8.7
28	July 09 – 15	31.46	21.94	86.86	51.71	1.5
29	July 16 – 22	32.34	21.86	88.14	49.00	8.8
30	July 23 – 29	28.74	21.14	94.86	71.00	83.4
31	July 30 – 05	26.56	20.57	96.00	79.14	173.2
32	August 06 – 12	29.03	20.69	95.86	63.71	17.9

Appendix II: Weekly meteorological data recorded at ARS, Bidar during *kharif*, 2007

Standard weeks	Month and Date	Temperature (^{0}C)		Relative humidity (%)		Rainfall (mm)
		Maximum	Minimum	Morning	Evening	
23	June 4 – 10	36.34	21.66	86.14	41.57	11.5
24	June 11 – 17	34.69	22.97	86.29	41.29	36.2
25	June 18 – 24	32.14	21.63	90.00	66.71	129.7
26	June 25 – 01	27.77	20.24	95.00	70.14	63.0
27	July 02 – 08	30.03	21.37	87.71	60.71	0.0
28	July 09 – 15	30.43	21.31	88.14	52.86	3.0
29	July 16 – 22	30.43	21.31	90.00	66.29	22.4
30	July 23 – 29	31.00	21.26	88.86	59.57	21.0
31	July 30 – 05	30.09	20.54	94.00	60.71	80.1
32	August 06 – 12	29.03	20.77	91.86	63.86	6.6

EPIDEMIOLOGY AND INTEGRATED MANAGEMENT OF ANTHRACNOSE OF GREENGRAM

SUNIL A. KULKARNI 2009 Dr. V. I. BENAGI
Major Advisor

ABSTRACT

Anthracnose caused by *Colletotrichum truncatum* (Schw.) Andrus and Moore is one of the major diseases of greengram. Survey work revealed that, the disease severity was found more in Bidar (49.43%) and least in Bijapur (23.86%) districts.

The observations on loss estimation revealed that comparatively lower disease index and maximum BCR was recorded in plots receiving two sprays of carbendazim. The average grain yield loss of 40.18% and stalk yield loss of 46.90% was noticed due to anthracnose in unsprayed plots.

The maximum mycelial weight was observed after 15 days of incubation. Solid medium like potato dextrose agar, liquid medium like Richard's medium, temperature of 30^0C, RH of 95%, pH 6.5 and alternate cycles of light and darkness were found best for the growth and sporulation of *C. truncatum*.

The spore load was maximum during last week of July and first week of August. The correlation studies between spore load, PDI and weather parameters indicated a negative correlation with temperature, while positively correlated with relative humidity and rainfall.

The greengram crop sown on 4^{th} June showed least disease severity. *C. truncatum* was survived upto 360 days in infected seeds and also in infected leaves stored under freeze condition. The flowering stage was found susceptible to the disease development. In host range studies only blackgram and horsegram had shown infection.

The genotypes TM-96-2 and TARM-18 were found resistant to anthracnose. Further, comparatively lesser sugar and higher phenol content were observed in these varieties than in susceptible variety.

In vitro evaluation revealed that, propiconazole, carbendazim, thiophanate methyl, benomyl, hexaconazole and carbendazim + mancozeb were found effective. Among botanicals azadirachtin was effective. Among bioagents and ITKs *Trichoderma harzianum* and cow urine respectively were most effective. In the management study hexaconazole was found to be effective in control of the anthracnose with maximum BCR