STUDIES ON VARIATION, SURVIVAL AND MANAGEMENT OF *Alternaria solani* Sorauer., THE INCITANT OF EARLY BLIGHT OF TOMATO (*Lycopersicon esculentum* Mill.)

MAHADEVAIAH P. G.

DEPARTMENT OF PLANT PATHOLOGY
UNIVERSITY OF AGRICULTURAL SCIENCES
BANGALORE
1993

STUDIES ON VARIATION, SURVIVAL AND MANAGEMENT OF *Alternaria solani* Sorauer., THE INCITANT OF EARLY BLIGHT OF TOMATO (*Lycopersicon esculentum* Mill.)

MAHADEVAIAH P. G.

Thesis submitted to the
University of Agricultural Sciences, Bangalore
in partial fulfilment of the requirements
for the award of the Degree of
MASTER OF SCIENCE (Agriculture)
in
PLANT PATHOLOGY

BANGALORE JULY 1993

Dedicated to

My Beloved Parents

and

Rao Bhadur Gubbi Thotadappa

Department of Plant Pathology
University of Agricultural Sciences
Bangalore

C E R T I F I C A T E

This is to certify that the thesis entitled **"STUDIES ON VARIATION, SURVIVAL AND MANAGEMENT OF *Alternaria solani* Sorauer., THE INCITANT OF EARLY BLIGHT OF TOMATO (*Lycopersicon esculentum* Mill.)"** submitted in partial fulfilment of the requirements for the degree of **MASTER OF SCIENCE(AGRICULTURE)** in **PLANT PATHOLOGY** to the University of Agricultural Sciences, Bangalore, is a record of research work carried out by **Mahadevaiah,P.G.**, under my guidance and supervision, and that no part of the thesis has been submitted for the award of any other degree, diploma, associateship, fellowship or other similar titles.

(M.N.L.Sastry)
Associate Professor of Plant Pathology

Bangalore
July 9 , 1993

APPROVED BY:

Chairman: (M.N.L.Sastry)

Members : 1. (H.B.Reddy)

2. (A.N.A.Khan)

3. (K.T.Shivashankar)

4. (K.Sampangi Ramaiah)

A C K N O W L E D G E M E N T

It gives me a great pleasure to express my profound sense of gratitude and indebtedness to **Dr.M.N.L.Sastry,** *Associate Professor, Department of Plant Pathology, University of Agricultural Sciences, Bangalore and chairman of my advisory committee for suggesting the problem, valuable guidance, scientific excellence, critical review of the manuscript and unfailing interest throughout the period of investigation and compilation of this thesis.*

I wish to express my sincere thanks to **Dr.H.R.Reddy,** *Professor and Head, Department of Plant Pathology, University of Agricultural Sciences, G.K.V.K., Bangalore, for providing necessary facilities to conduct research in the Department. I would also like to thank him for having critical gone through the manuscript and valuable suggestions for the improvement of the thesis as a member of my advisory committee.*

I also owe my grateful thanks to **Dr.A.N.A.Khan,** *Associate Professor of Plant Pathology,* **Dr.K.Sampangi Ramaiah,** *Assistant Professor of Plant Pathology and* **Mr.K.T.Shivashankar,** *Associate Professor of Horticulture, U.A.S., G.K.V.K., Bangalore, for having served as member of my advisory committee and for the constructive criticism and valuable suggestions for the improvement of this thesis.*

I am grateful to Mr.Subramanyam and Mr.Ganeshan, for having provided the land and necessary materials during the course of field trial. I also thank Mr.Chandrappa, Research Assistant, Department of Soil Science, for providing necessary facilities to carryout the investigation on Melanin.

I whole heartedly thank my friends S.Onkarappa, Chandrashekar,B., Krishna and other P.G. students for their assistance and co-operation during the course of the present investigation. I also take this opportunity to express my sincere thanks to all the faculty members and supporting staff of the Department of Plant Pathology who have helped me in one way or the other during the course of my investigation.

I wish to express my appreciation to **Mr.N.V.Srinivasa**, Stenographer, for the fine printout of the manuscript on their Computer System.

Above all, I owe my success to my loving parents, beloved brother and sister for having been a source of inspiration throughout and for their constant encouragement, it would not have been possible to complete this study successfully.

Bangalore
July 9 , 1993

[MAHADEVAIAH, P.G.]

C O N T E N T S

LIST OF TABLES

LIST OF FIGURES

LIST OF PLATES

INTRODUCTION

I. INTRODUCTION

Tomato (Lycopersicon esculentum Mill.), a succulent plant belonging to the family Solanaceae is known for its acid fruit (Atherton and Rudich, 1986). It is being cultivated as an annual fruit vegetable. Among the vegetables, tomato ranks next to potato and sweet potato in its world acreage; and as a processing crop it ranks first (Yawalkar, 1969). Both raw and ripe fruit of tomato are used for culinary purposes. It is an abundant sources of vitamins A, B, C and minerals. Easy adaptability to a wide range of climatic and soil conditions enables its world wide cultivation (Bose and Som, 1986). Tomato occupies an area of 2883 million hectares with the production of 69145 thousand metric tons in the world (Anon., 1991). India is also one of the leading countries in tomato production with an area of 300 thousand hectares and a production of 3100 thousand metric tons (Anon., 1991). In Karnataka, it occupies an area of 36,201 hectares with a production of 6,64,023 metric tons (Anon., 1991). Many factors operating in the successful cultivation of tomato of which diseases play an important role.

There are several diseases on tomato caused by fungi, bacteria, viruses and nematodes (Balancard, 1992).

Among the fungal diseases, early blight also known as target spot disease incited by Alternaria solani Soraueris one of the world's most catastrophic diseases. It causes loss upto 50-86% in fruit (Mathur and Shekhawat, 1986). The causal organism is air borne and soil inhabiting and is responsible for early blight, collar rot and fruit rot of tomato (Datar and Mayee, 1981).

In the preliminary field survey of the disease during 1991-92 in southern districts of Karnataka viz., Tumkur, Bangalore, Mandya and Kolar, a mild to severe form of Alternaria infection on tomato was recorded, indicating the presence of pathogenic variation in the organism. This study has not been made earlier.

Spores of the fungi are one of the important means of dissemination and also used in the identification and classification of the organism. Not much information is available on the factors responsible for spore production of the fungus and necessitates its study.

It is well established that the tomato early blight fungus could survive on the infected seeds for several days. But, it is still speculative whether the

seed-borne inoculum of A.solani serves as a source for triggering primary infection in the next season (Neergaard, 1945). The ability of the pathogen to survive for a long time in the diseased plant parts, soil and on alternate hosts in the absence of the main host, determine the ability of the pathogen to perpetuate (Moore and Thomas, 1942; Basu, 1971 and Rands, 1917). As not much light has been shed on these aspects of the pathogen, thorough knowledge on the survivability of the pathogen is needed to evolve suitable control measures.

Spraying of broad spectrum fungicides like Dithane M-45, Captan and Duter has been recommended for the control of early blight of tomato by several workers (Ramakrishnan, 1971 and Stevenson, 1977). While, the number of applications of these chemicals are more, they are less persistent on the foliage (Thind and Jyooty, 1982). Thus, the control achieved by these chemicals is inadequate. One of the reasons attributed for the low sensitivity of A.solani to fungicides mentioned above is the production of dark brown to black pigment called melanin by the fungus, which enhance the survival and competative abilities of the pathogen under certain environmental conditions (Bell and Wheeler, 1986). Therefore, in the present study it is thought worthwhile

to test the efficacy of more chemicals like Iprodione and Captafol against early blight fungus and also the role of melanin in the fungicidal efficacy against a fungus.

The objectives of the investigations are as below:

1. To survey and collection of A.solani isolates from different localities to study the pathogen variations.

2. To study the factors affecting the production and germination of spores of A.solani in the laboratory.

3. To study the survivability of A.solani in seed, soil and plant debris.

4. To screen the promising varieties of tomato for resistance against A.solani.

5. To screen some fungicides against A.solani under laboratory and field conditions.

6. To study the melanin pigment from the organism and to study its role in the effectiveness of fungicides.

REVIEW OF LITERATURE

II. REVIEW OF LITERATURE

Early blight of tomato caused by Alternaria solani Sorauer is an important and widely distributed disease through out the world and it occurs in mild to severe form in different parts of the country. Large number of reports are available in literature on different aspects of the disease. In the present review only those references relevent to the aspects investigated have been included.

1. SYMPTOMATOLOGY

According to Locke (1949) the early blight on tomato was characterized by the appearence of brown to dark leathery necrotic spots first on the leaflets producing target-board effect. Walker (1952) reported that, the spots were oval or angular in shape upto 0.3 or 0.4 cm diameter and there was usually a narrow chloratic zone around the spot which later faded into the normal green colour. Older leaves of tomato were affected first as a rule and the disease progressed upward. Finally the leaves dried up and drooped down. Ramakrishnan (1971) observed cankerous spots on tomato stems. They were specially injurious when they occured at the juncture of the stem and side branches. Collar

rot, another symptom on tomato occured as a stem lesions on seedlings at the soil line extending above and below that point to form cankers, which resulted in girdling of the plants (Basu, 1971; and McCarter et al., 1976). Datar and Mayee (1981) showed that A.solani could attack fruits in the green and ripe stages at the stem end, growth cracks and other wounds.

2. CAUSAL ORGANISM

Early blight of tomato caused by A.solani was first recorded in 1882 in New Jersey, USA (Bose and Som, 1986). The mycelium consisted of septate, branched, light brown hyphae which turned darker with age. The conidiophores were short, 50 to 90 µm and dark coloured: conidia were 120-296 x 12-20 µm in size, beaked, muriform, dark coloured and borne singly. However in culture they formed short chains. According to Singh (1987), the conidia contained 5-10 transverse septa and 1-5 longitudinal septa.

3. PATHOGENICITY

Andrus et al. (1945) confirmed the pathogenicity on tomato by using mycelial fragments of A.solani as inoculum. Locke (1949) used blended mycelial fragments of A.solani for puncture inoculation. Brock(1950) and

Henning and Alexander (1959) used the suspension of mycelial fragments of A.solani to inoculate the leaves of field or green house grown plants. Barksdale (1968) and Dhiman et al. (1980) used suspension containing 20,000 spores/ml distilled water for proving pathogenicity of early blight of tomato caused by A.solani. Further, they atomized the culture suspension on three leaf stage seedlings at the rate of 30 ml per seedling for successful inoculation..

4. CULTURAL STUDIES

Several reports in the literature showed that potato dextrose agar as a good medium for the growth and sporulation of A.solani (Bonde, 1929; Neergaard, 1945 and Rotem, 1966). Barksdale (1968) showed that lima bean agar and potato dextrose agar were the best media for growth and sporulation of A.solani. Kaul and Saxena (1988) reported cultural variability of A.solani isolates on potato dextrose agar and classified them into four distinct cultural groups based on types of growth, colony colour, colour of the substrate and growth rate.

5. PHYSIOLOGICAL STUDIES

5.1. Temperature

The temperature requirement for A.solani was found to be in the range of 5-35^{o}C (Bonde, 1929; Verma,

1970 and Gemawat and Ghosh, 1979). Kaul and Saxena (1988) reported that the maximum growth of five isolates of A.solani at 25°C followed by 20, 15, 10 and 5°C with the least growth at 35°C. A.solani germinated most rapidly in darkness when ambient temperature was near 25°C (Stevenson and Packer, 1988).

5.2. Light

According to Lukens (1963) the conidia of A.solani normally formed after 6th hour in the dark. Prasad and Dutt (1971) found maximum sporulation in six-days old culture with 24 hours of exposure to sunlight for 6 min. than culture exposed to incandescent electric light or infrared light. Growth and sporulation of A.solani was sparse in Czpek's synthetic medium but the same was high in case of semi-synthetic and natural carrot leaf media when exposed to light (Fencelli and Kimati, 1990).

6. SPORULATION

Rands (1917a) induced sporulation in A.solani by growing the fungus on potato-dextrose agar for 10-12 days. McChallan and Chan (1944) induced sporulation in A.solani by mycelial wounding on PDA and then irradiated

the cultures with UV light. Charton (1953) and Ludwig et al. (1962) reported sporulation of A.solani on different media due to dehydration. Padhi and Rath (1973) described the effect of sunlight, nutrition, pH and temperature on sporulation of A.solani. Shahin and Shepard (1978) reported that A.solani produced spores on water agar plus $CaCO_3$ at 18°C. Vakolounakis (1983) recorded that large masses of conidia formed when mycelial discs of A.solani inoculated onto sterile solanaceous leaf discs placed on water agar plates at 20°C. Zhu et al. (1985) showed that A.solani sporulated profusely on corn meal agar when illuminated with fluorescent lamp for 8 hours at 18°C.

7. SURVIVABILITY

Robert and Boothroyd (1972) showed that A.solani, the pathogen causing early blight of tomato and potato survived as long as 18 months in dry diseased leaves. A.solani inoculum persisted in field for several months (Basu, 1971). Alternaria solani overwintered as conidia, chlomydospores and mycelium on plant debris and in the soil (Dorozhkin and Ivanyuk 1979). Mahabaleswarappa (1981) recorded that A.carthami Chowdhury survived as conidia for about four months and as mycelium for five

months, when diseased leaves were kept between folds of blotting papers.

According to Kvasnyuk (1986) conidia of A.solani remained viable on over wintering potato litters every year and conidia were killed by wet warm weather in autumn and spring or by frequent thaws in winter followed by sharp drop in temperature. Patterson (1991) reported that chlamydospores persisted in soil for 12 months and played a role as primary inoculum for A.solani. Chlamydospores placed at depth of 4.8 and 12 cm in soil initiated infection and collar rot on tomato.

8. HOST RANGE

Nagaraj and Ponnappa (1970) reported a wide host range for Alternaria species and showed that it infected the members of Amaranthaceae, Amaryllidaceae, Brassicaceae, Cucurbitaceae, Euphorbiaceae, Fabaceae, Malvaceae, Pedalaceae, Porreae and Solanaceous families.

9. VARIETAL REACTION

According to Basu (1974a) the tomato variety, mini rose found to be resistant among the nine tomato cultivars tested against A.solani in field trials. Carrasco et al. (1977) found that Cuba C-I was the best

cultivar producing the highest yield among eleven cultivars screened against A.solani, the incitant of early blight in tomato. Vakalounakis (1982) found that meltin and nemato tomato varieties were slightly less susceptible to A.solani than the other tomato varieties tested in glasshouse. Sridhara and Naik (1983) observed that four cultivars showed some resistance to A.solani among the 38 tomato cultivars evaluated in the field. Reda et al. (1985) noticed that fruits of Red stone, Roma VF and marmandi-early were more susceptible to rot caused by A.solani in tomato. Ismail (1987) reported some resistance in var. NY-59 and desires among 16 var. and clones of tomato screened against A.solani. Madalageri et al. (1988) recorded resistance in 19 genotypes against early blight among 58 genotypes obtained from India, USA, Taiwan and Netherland by screening against A.solani under epiphytotic condition. Sharif et al. (1988) studied the reaction of 35 tomato varieties to A.solani under artificial conditions and found that dombito and monte-carlo were susceptible to A.solani. Choulwar et al. (1990) studied the reaction of 5 hybrids, 20 lines and 28 varieties to A.solani and none showed complete resistance. While, varieties bonny best, mangala, sheetal, vaishali, somrudhi and hybrids HY 9, Hy 10, Hy 13, Hy 14 and Hy 15 were found to be moderately resistant.

10. CHEMICAL CONTROL

Kodmelwar et al. (1973) reported that copper-based fungicides gave the best control of A.solani in in vitro. Lodha and Prasad (1975) found that Dithane Z-78 very effectively checked the growth of A.solani followed by crystalline actione in pot trials and in vitro. In the field trials of fungicides against A.solani, Dithane M-45 performed better than the other fungicides such as Dithane M-22, Bavistin, Vitavax, Chlorothalonil, Captofol, Benlate and Difolaton (Abol-Wafa and Kamara, 1975; Stevenson, 1977 and Rajagopal and Vidyashekaran, 1983). Ramakrishnan and Kandaswamy (1978) from their field trials in 1972 and 1974 reported that A.solani on tomato was controlled and yield also increased with the application of Dithane M-45 at 0.2 per cent followed by Difolaton and Benlate. Bashi (1979) showed that Daconil at 3 kg/ha twice a week reduced incidence of A.solani on foliage of potato. But according to Quinn and Johnson (1979) weekly high volume captafol spray applications against A.solani were more effective. Mora (1978) obtained best results in controlling early blight of tomato caused by A.solani with the application of Daconil, Duter and Difolaton at intervals of 7, 10 and 13 days from the begining of flowering stage. Neto and

Oliveira (1980) stated that Maneb and Propineb were most toxic effect against A.solani among the seven fungicides tried.

In field tests, the inorganic copper fungicides such as cupric hydroxide provided good control of tomato early blight when applied at low and ultra low dosages (Mabbett and Phelps, 1985). Dithane M-45 proved best for controlling early blight disease of potato and tomato (De and Chattopdyaya, 1984 and Datar and Mayee, 1985). Maeso (1986) evaluated efficacy of fungicides for the control of early blight of tomato for two years and recorded good control of the disease by sprays of captofol (Difolaton), Chlorothalonil (Daconil) and Fentin acetate (Breston) during 1981-82 and chlorothalonil Dconil, Fentin acetate+maneb (Brome), copperoxychloride+maneb+zineb (Cuprosan), copper salt+mancozeb (Trimiltox forte) and mancozeb alone (Dithane M-45) during 1982-83. Achuta et al. (1988) found that among five fungicides tested carbendazim, captan and moncozeb were more effective in controlling early blight and increasing the yield in tomato.

11. MELANIN PIGMENT

According to Pigman (1957) the spore walls and dark hyphal walls of soil fungi contained melanin. Zink

and Fengal (1989) studied that hyphae of two blue-stained fungi Ceratocystis fimbriata Elli and Hals and A.alternata (Fr.) Keissler by scanning and transmission electron microscopy, and identified the colouring matter as melanin deposited in the walls of the two fungi. Dahiya (1989) reported that melanin production in the light was higher than dark. Lockwood (1960) noted that the dark hyphae of Helmenthasporium sativum Pam.King and Bakke and A.solani were more resistant to lysis than the hyaline hyphae. Polysaccharide hydrolyzing enzymes excreated by mycolytic organism, lysed hyaline fungal structures to varying extents but fungi that produced melanized spores like sclerotia and vegetative hyphae were resistant to lysis (Portgieter and Alexander, 1966; Bloom Field and Alexander, 1967 and Chu and Alexander, 1976). Kuo and Alexander (1967) reported that the resistance of Aspergillus nidulans (Eidom) Wint. hyphae to lysis by a β-(1-3) gluconase-chitinase resulted from the presence of melanin in the fungal wall. Several antipenetrant fungicides such as Tricyclazole pyroquilon, chlorobenthiazone and Rabcide compounds had no curative effects against melanin producing fungi such as Pyricularia oryzae Breda de Haan (Fryoyd et al., 1978; Inoue et al., 1983 and Schwinn et al., 1979).

MATERIAL AND METHODS

III. MATERIAL AND METHODS

The present investigation on early blight disease of tomato was conducted during 1991-92 at the College of Agriculture, GKVK, Bangalore. The material used and the methods followed are described below.

1. SURVEY AND DISTRIBUTION

Random survey was conducted during 1991-92 around Bangalore, Tumkur, Mandya, Doddaballapur and Kolar to know the occurrence and severity of the early blight disease at different growth stages of tomato. In each field visited, 15 plants were randomly selected in 30x30 mt^2 area and by selecting 5 leaves randomly in each plant, the severity of the disease was assessed by using 0-9 scale (Mayee and Datar, 1986), and per cent disease index worked out using the formula (Wheeler, 1969).

$$\text{Per cent disease index (PDI)} = \frac{\text{Sum of individual ratings}}{\text{No. of leaves examined} \times \text{Maximum disease grade (9)}} \times 100$$

2. ISOLATION AND IDENTIFICATION

Standard tissue isolation technique was followed to obtain A.solani culture. For this purpose tomato plants showing typical early blight symptoms were

collected from different places viz., Jakkur (Bangalore), Gubbi (Tumkur), Bodanur (Mandya), Kolar and Doddaballapur (Bangalore). The leaves were cut into small bits measuring about 2 mm and surface sterilized in 0.5 per cent sodium hypochlorite solution for 2 min. Such bits were transferred into Petridishes containing 15 ml Potato Dextrose Agar (PDA) and incubated at 28±1°C for 7 days. Pure culture of the fungus was obtained by hyphal tip isolation method. The conidial morphology of the fungus is studied by using micrometer. Thus, the isolates collected from various places were designated as follows.

Sl.No.	Location	Designation
1	Bangalore	Isolate AS1
2	Doddaballapur	Isolate AS2
3	Kolar	Isolate AS3
4	Mandya	Isolate AS4
5	Tumkur	Isolate AS5

3. MAINTENANCE OF CULTURES

The five isolates of A.solani were maintained at 5°C in the refrigerator and subcultured periodically at an interval of 30 days during the course of this study.

4. PATHOGENICITY TEST

Pusa ruby Tomato seedlings were grown in earthen pots filled with sterilized soil. Thirty days old plants were

sprayed with sterile distilled water before inoculating the plants and then covered with a polythene hood for 24 hours. Spore and mycelial suspension ($2x10^4$ spores/ml) was prepared in sterile water from ten-day-old culture and then spot inoculated onto leaves. Such inoculated plants were again covered with polythene hoods, the inner walls of which were lined with the moist absorbant cotton to ensure high humid conditions. After 48 hours of incubation, polythene bag was removed and the plants were kept in green house. Controls were maintained by inoculating the plants with only sterile distilled water. Observations were made for symptom development periodically. Reisolations were made from infected plants and the cultures thus obtained were compared with original cultures to confirm the identity of the pathogen.

5. CULTURAL STUDIES

The cultural characters of the five isolates of A.solani were studied on the following solid media viz.,

1. Potato dextrose agar
2. Richard's agar
3. Czapek's dox agar
4. Oat meal agar

5. Bean meal agar

6. Host extract agar.

The composition and preparation of different media are given below (Tuite, 1969).

Potato dextrose agar

Potato..... 200 g

Dextrose..... 20 g

Agar..... 20 g

Distilled water...1000 ml

Potato slices were boiled in 500 ml distilled water for 20 minutes and filtered through muslin. Agar was melted in another 500 ml distilled water, both the solutions were mixed and volume was made upto 1000 ml and sterilized.

Richard's agar

Potassium nitrate..... 10 g

Potassium monobasic phosphate..... 5.0 g

Magnesium sulphate..... 2.5 g

Ferric chloride..... 0.02 g

Sucrose..... 50.0 g

Distilled water..... 1000 ml

Czapek's dox agar

Sodium nitrate	3.0 g
K_2HPO_4	1.0 g
$MgSO_4.7H_2O$	0.5 g
KCl	0.5 g
$FeSO_4.7H_2O$	0.1 g
Sucrose	30.0 g
Agar	15.0 g
Distilled water	1000 ml

Oat meal agar

Rolled oats	60.0 g
Agar	15.0 g
Distilled water	1000.0 ml

Rolled oat was boiled in 500 ml distilled water for 30 minutes, filtered through muslin, agar was melted in another 500 ml distilled water. Two solutions were mixed and the volume was made upto 1000 ml and then sterilized.

Bean meal agar

Green fresh bean pods	200.0 g
Agar	15.0 g
Distilled water	1000.0 ml

Sliced and washed bean pods were boiled in 500 ml distilled water for 20 minutes and filtered through muslin. Agar was melted in another 500 ml distilled water, both the solutions mixed and the volume was made upto 1000 ml and sterilized.

Host extract agar

Healthy tomato leaves.....	200.0 g
Agar.....	20.0 g
Distilled water.....	1000.0 ml

Tomato leaves were boiled in 500 ml of water for 30 minutes. Extracts was collected by filtering through muslin. The agar was melted in 500 ml of water. Both the solutions were mixed and the volume was made upto 1000 ml and sterilized.

Twenty ml of each medium listed above was poured into 75 mm diameter Petriplates. After solidification 5 mm disc from the respective A.solani isolates were cut using a gel cutter and a single disc was placed at the centre of the plate. Each set of experiment was replicated thrice and the plates were incubated at $26\pm1^{o}C$ for seven days. The colony diameter in the culture plates and cultural characters such as, colony colour,

type of margin, substrate colour and sporulation were recorded.

6. SPORE GERMINATION STUDIES

Spore germination of five isolates of A.solani was studied using standard "hanging drop" technique in cavity slides. A drop of spore suspension of each isolate prepared in tap water, glucose 2% solution, sucrose 2% solution, host extract, lima bean extract and Czpek's solution was transferred separately onto cover slips which were later inverted and placed over a cavity slide. The edges of the cover slips were sealed by using veseline. The slides were placed in petriplates lined with moist absorbent cotton at room temperature. Germination counts were recorded after eight hours of incubation. The spore germination was calculated by taking the average number of spores germinated out of the total number of spores present in ten microscopic fields, under low power objective and expressed as percentage of germination.

7. PHYSIOLOGICAL STUDIES

7.1. Effect of temperature on the growth and sporulation of A.solani

The different temperature tried for growth and sporulation of each of the five isolates were 15, 20, 25,

30 and 35°C. Fifteen ml of sterilized PDA was poured into 75 mm diameter petriplates and inoculated aseptically with five mm disc of the five isolates of A.solani from a seven day old culture. Petriplates were incubated at different temperatures and each treatment was replicated thrice. At the end of seventh day of incubation, observations on colony diameter and sporulation were recorded.

7.2. Effect of light on the growth and sporulation of A.solani

The effect of light on growth and sporulation on five isolates of A.solani was studied on potato dextrose agar by exposing the pure cultures to continuous light, continuous dark and alternate cycles of 12 hours complete light and 12 hours complete darkness. The inoculation of culture to petriplates containing PDA was done as explained earlier. The plates were incubated at 26±1°C for eight days, observations on colony diameter and sporulation were recorded.

8. PATHOGENIC VARIATION OF ISOLATES

Pathogenic studies of five isolates of A.solani were carried out on Pusa ruby variety of tomato grown in polythene bags (30x15 cm). Thirty days old seedlings

were inoculated by spray method with the spore suspension ($2x10^4$ spores/ml) of each five isolates of fungus separately and replicated five times. Immediately after inoculation seedlings were covered with polythene bags to increase relative humidity. After 48 hours, the polythene bags were removed and the seedlings were placed in green house. Observations were recorded 15 days after inoculation for number of spots per leaflet and lesion size.

9. SURVIVAL OF A.solani

9.1. Survivability of the pathogen in infected seeds and plant debris

This study was carried out to know the period of viability of the pathogen in the infected seeds and plant debris. For this purpose tomato leaves infected with Alternaria were collected and stored in the laboratory. Tomato seeds collected from fruits infected with A.solani were used to study the survivability of fungus in seed. Viability of A.solani in seeds was determined by planting thirty such seeds on potato dextrose agar in petriplates at 20 days intervals. The study was continued till the least recovery of the pathogen in seeds. The percentage of seeds yielding the fungus was recorded and tabulated.

Periodic isolations at 20 days interval were made from the plant debris stored under laboratory conditions. Standard tissue isolation technique was followed for this purpose. Recovery percentage of A.solani from infected debris was recorded. This study was continued for a period till the least recovery of A.solani was made from the host debris.

9.2. Survivability in soil

The study on the survivability of A.solani in soil was made on both sterilized and unsterilized soils. Soils made sick with 5 per cent inoculum of A.solani(As3) were filled to plastic cups (3"x2") separately at the rate of 100 g of soil per cup. Different moisture levels viz., 10, 30, 40, 50, 70 and 90 per cent moisture holding capacity were maintained throughout the experiment, by regular watering at 24 hour intervals on weight basis. The survivability of A.solani was studied based on collar rot infection (Patterson, 1991). From each treatment three cups were drawn at 25 days interval and ten healthy tomato seeds were sown in each cup. Twenty days after sowing, the collar rot infection was recorded. This study was continued over a period at which no collar rot infection was noticed.

The collar rot infection was confirmed by observing the infected tissues microscopically for conidial production and finally by tissue isolations.

10. HOST RANGE STUDIES

Different host plants of cereals, legumes, oilseeds, pulses, vegetables and weeds listed below were included in host range studies. Spores and mycelial suspension of A.solani prepared in sterile distilled water by using eight-day-old culture ($2x10^4$ spores/ml) was used for spot inoculation of plants with and without wounding. Two to three drops of inoculum suspension was placed in the centre of the leaves. The plants were kept under humid chamber for 48 hours and then transferred to glass house. All plants sprayed with sterile distilled water in place of spore suspension served as control. The time taken for the development of symptoms on each host was compared with time taken for the development of symptoms on tomato.

LIST OF HOSTS

Family	Scientific name	Common name
Amaranthaceae	Amaranthus paniculatus L.	Amaranthus
Apiaceae	Coriandrum sativum L.	Coriander
	Dacus carota L.	Carrot

Family	Scientific name	Common name
Asteraceae (=compositae)	Carthamus tinctorias L.	Safflower
	Helianthus annuus L.	Sunflower
	Tagetes erecta L.	Marigold
Brassicaceae	Brassica oleracea var. botrytis L.	Cauliflower
	Brassica oleraceae var. capitata L.	Cabbage
Cruciferae	Raphanus sativus L.	Radish
Cucurbitaceae	Lagenaria siceraria L.	Bottle gourd
	Momordica charantia L.	Bitter gourd
Euphorbiaceae	Ricinus communis L.	Caster
Fabaceae	Cajanus cajan (L.) Mill	Redgram
	Crotolaria juncea L.	Sunnhemp
Leguminaceae	Pisum sativum L.	Peas
	Vicia faba L.	Broad bean
Amaryllidaceae (=liliaceae)	Allium cepa var. cepa	Onion
Malvaceae	Abelmoschus esculantus L.	Bhendi
	Gossypium herbaceum L.	Cotton
Pedaliaceae	Sesamum indicum L.	Sesamum
Solanaceae	Capsicum annuum L.	Chilli
	Lycopersicon lycopersicum L. (=L.esculentum Mill)	Tomato
	Nicotiana glutinosa L.	Tobacco
	Solanum melongena L.	Brinjal
	Solanum tuberosum L.	Potato

(According to Holm et al., 1981)

11. VARIETAL SCREENING STUDIES

The experiment was conducted under glass house conditions to study the reaction of 21 popular tomato varieties and hybrids to A.solani.

The tomato plants of different varieties and hybrids were grown in polythene bags. Thirty days old seedlings were artificially inoculated by spraying the homogenized mycelial and spore suspension ($2x10^4$ spore/ml) of 12 days old culture of A.solani. Such inoculated plants were kept in humid chambers for 48 hr and then removed to glass house. Five replications were maintained for each variety. Proper controls were maintained for each variety/hybrid without inoculations with the fungus but maintaining all other conditions. Observation on the reaction of each varieties/hybrid to infection was recorded 15 days after inoculation. The disease grading was done as follows (Mayee and Dater, 1986).

Disease grading

Scale	Description
0	No symptoms on the leaf.
1	Small, irregular spots covering less than 1% of the leaf area.
3	Small, irregular, brown spots with concentric ring covering 1-10% of the leaf area.
5	Lesion enlarging, irregular, brown with concentric rings cover 11-25% of the leaf area.
7	Lesions coalesce to form irregular brown patches with concentric rings. Covering 26-50% of the leaf area. Lesion also on stem and petioles.

9 Lesions coalescing to form irregular, dark brown patches with concentric rings covering 51% or more of the leaf area. Lesions on stem and petioles.

The per cent disease index was calculated by using the formula (Wheeler, 1969).

$$\text{Per cent disease index (PDI)} = \frac{\text{Sum of individual rating}}{\text{No. of leaves examined}} \times \frac{100}{\text{Maximum disease grade (9)}}$$

The data on per cent disease index was analysed statistically on the basis of disease index and the varieties were classified into following 6 categories.

Category

Sl.No.	PDI	Reaction
1	<1%	Immune
2	2-10	Resistant
3	11-15	Moderately resistant
4	16-20	Moderately susceptible
5	21-25	Susceptible
6	26-30	Highly susceptible

12. IN-VITRO EVALUATION OF FUNGICIDES

The poisoned food technique (Falck, 1907) was followed to evaluate the efficacy of fungicides in laboratory against A.solani at concentrations of 100, 250, 500 and 1000 ppm with three replications each. The different fungicide tested were:

Common name	Chemical name
Captan	N-trichloromethyl thio-4-cyclohexene-1, 2-dicarboximide.
Captafol	Cis-N-(1,1,2,2-tetro chloro ethyl thio-4 cyclohexene 1,2-dicarboximide).
Carbendazim	Methyl-2-benzamidazole carbomate.
Chlorothalonil	Tetra chloro isophthalonitrile.
Iprodione	3-(3,5-dichlorophenyl)-N-isopropyl-carbomayl-2, 4-dioxoimidozolidine-1-carboxamide.
Mancozeb	Manganese ethelene bis dithiocarbamate.
Carboxin	5,6-dihydro-2-methyl-1, 4-oxythin-3-carboxanilide.

Molten sterilized potato dextrose agar was used as nutrient medium and required quantity of each fungicide was added separately so as to get a requisite concentration of that fungicide. The fungicides were thoroughly mixed by stirring and about 15 ml poisoned medium was poured to each of the 75 mm petridishes and allowed for solidification. The actively growing periphery of the seven day old culture of A.solani was carefully cut using a gel cutter end transferred aseptically to the centre of each petridish containing the poisoned solid medium. Suitable control was maintained by growing the cultures on PDA without the fungicides. The plates were incubated at $28\pm1^{o}C$ for

eight days and the colony diameter was recorded after that period.

13. FIELD EVALUATION OF FUNGICIDES

The field experiment was laid out in Randomised Complete Block Design (RCBD) with 8 treatments and 4 replications during 1990-91 and 1991-92 at Jakkur, near University of Agricultural Sciences, Bangalore.

Healthy Pusa Rubi tomato seedlings were raised in nursery beds and 21 days old seedlings were transplanted into the field with 75 cm inter and 60 cm intra row spacing in plots measuring 3x3 mt. All other cultural and pest control practices were followed as recommended in package of practice (Anon., 1991).

The fungicides - Mancozeb 0.2%, Captafol 0.2%, Carbendazim 0.1% and Iprodione at 0.1%, 0.2%, 0.3% and 0.4% were tested as 4 sequential sprays at an interval of 15 days. The first spray was carried out as soon as the first symptom of early blight was seen in the field.

Ten plants were selected randomly in each plot and observations such as severity of the disease on the foliage, per cent Alternaria fruit rot and per cent fruit stem infection were recorded under different treatments

one day before each spray and 15 days after the final spray. The intensity of disease on ten randomly selected leaves per plant was recorded as described in varietal screening.

The fruit yield in each plot was recorded and the economics of fungicide spray schedule was calculated on the basis of prevailing market rates of tomato fruits and fungicides.

14. EXTRACTION OF CRUDE MELANIN

The production of melanin pigment by A.solani was studied at different growth stages of the fungus viz., 5, 7, 9, 10, 13, 15, 17, 19 ans 21 days after inoculated into the media by following the procedure given by Ellis and Griffith (1974). The fungus was grown on 100 ml potato dextrose broth taken in 150 ml conical flasks. Thirty grams of mycelium harvested at each interval, was homogenized in 150 ml of 1M KOH for 5 minutes in a mixer. The cellular debris was extracted after treating with hot alkali (1M KOH at 100^{o}C) and filtered through Whatman's No.1 filter paper. The black residue was discarded while the dark brown to black filtrate was acidified to pH 2. The resistant dark brown black precipitate was collected by centrifugation, washed in distilled water and

redisssolved in 1M KOH at room temperature to obtain a 'crude' alkali melanin. The intensity of colour was read at 600 nm in spectrophotometer. Similarly, blanks were prepared which were used to adjust the light transmission to 100 per cent. This preparation was used for further studies.

The role of melanin on the efficacy of fungicides

The role of melanin of A.solani in the efficacy of a fungicide was studied by making use of an hyaline fungus - F.oxysporium f.sp. cubens (E.F.Smith) Snyd. and Hons. as a test organism. An experiment was set up with melanin taken in different quantities viz., 1, 2, 3 and 4 ml and then mixed with 250 ppm of Mancozeb (The conc. of the fungicide at which total inhibition of the test fungus can be obtained) in 100 ml of molten sterilized PDA. About 15 ml of medium was poured into each of the 75 mm petridishes and allowed for solidification. The actively growing periphery of the seven days old culture of the test fungus was carefully cut using a gel cutter and then transferred aseptically to the centre of each petridish and incubated at $28\pm1^{o}C$. Suitable controls were maintained by growing the fungus in 1, 2, 3 and 4 ml melanin alone (without the fungicide), 250 ppm Mancozeb

alone (without melanin) and on PDA (without either of melanin or fungicide). The observations on cultural characters, colony diameter and per cent inhibition of growth over control of the fungus were recorded after 7 days incubation by using the formula given by Vincent (1927).

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

where, I = per cent inhibition

C = growth in control

T = growth in treatment.

EXPERIMENTAL RESULTS

IV. EXPERIMENTAL RESULTS

The results of the experiments conducted on various aspects of early blight of tomato caused by Al ternaria solani with reference to symptamotology, incidence, variation in the pathogen, survivability and management of the disease are presented hereunder.

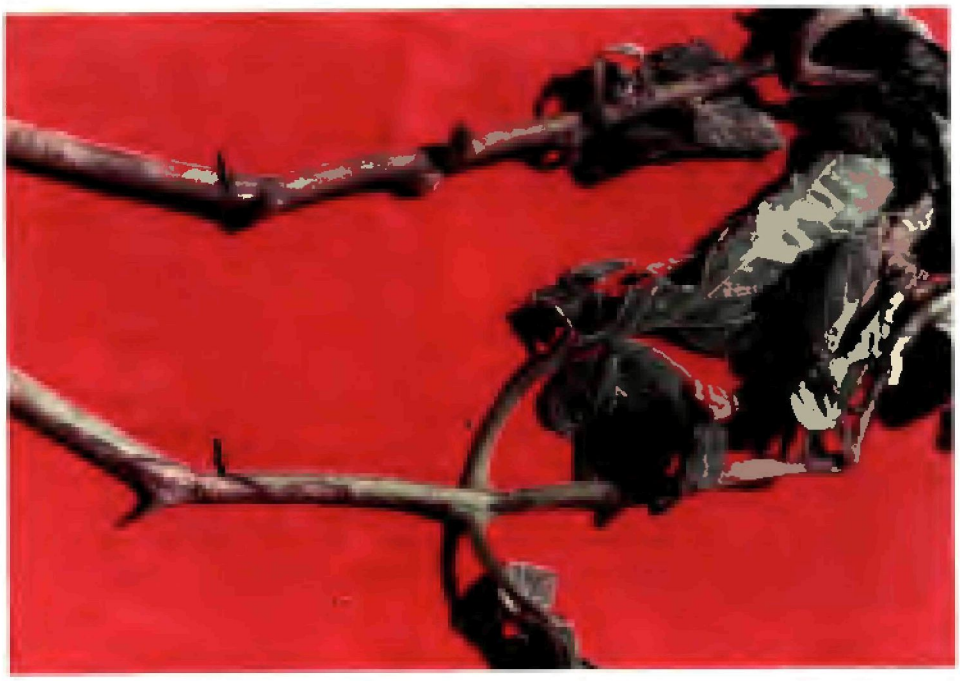
4.1. SYMPTOMOTOLOGY

The disease symptoms appeared on all the plant parts viz., leaf, stem, petioles, calyx and fruit. First symptom appeared on older leaves (Plate 1) as minute dark brown usually round necrotic spots of one to two mm in diameter. Later the spots enlarged with characteristic concentric rings at the centre to produce a target-board effect and the colour of the spots changed from brown to dark brown. The adjacent spots eventually coalesced to form large irregular spots ultimately leading to defoliation when plants were 60 days old. Symptoms also appeared on stem (Plate 2) and petioles (Plate 3) as brown to dark brown elongated cankerous target board type spots. These spots enlarged and covered the entire stem and petioles leading to withering of the plants. Symptom also developed on calyx (Plate 4) and flower buds in the form of minute brown to dark brown spots which enlarged

b) Diseased leaf a) Healthy leaf

1 Photograph showing typical symptoms due to Alternaria solani on tomato leaves

2 Photograph showing typical symptoms due to Alternaria solani on tomato stem



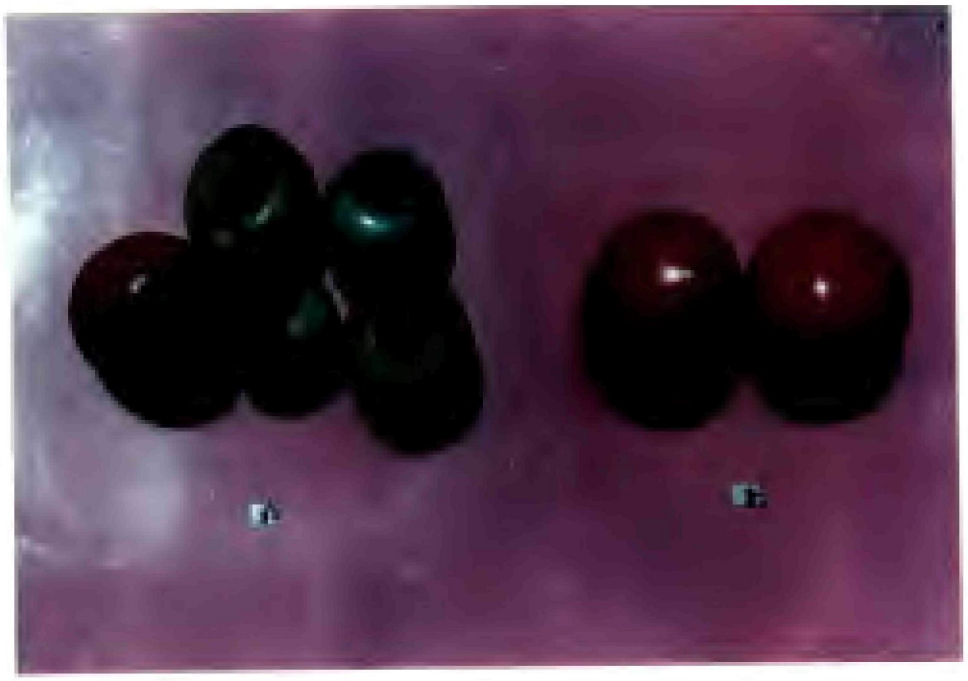
3 Photograph showing typical symptoms due to Alternaria solani on tomato petioles

4 Photograph showing typical symptoms due to Alternaria solani on tomato calyx

later and spread to sepals and fruit stems resulting in pre-matured dropping of fruits. The symptoms on fruits appeared (Plate 5) first at stem end as black or brown sunken spots both on green and ripe fruits which enlarged within eight days involving most of the fruits. Finally the fruits decayed.

4.2. SURVEY AND DISTRIBUTION

The data on early blight severity collected from major tomato growing localities of southern Karnataka during 1991 are presented in Table 1. The disease severity ranged from 0 to 96.15 per cent. The maximum disease severity was recorded on varieties Rupali, Sonali, Pusa ruby, Naveen and Rasmi with per cent disease index of 96.15, 92.06, 61.88, 60.12 and 55.12 respectively, in fields around Bangalore, Tumkur, Mandya, Kolar and Doddaballapur. The varieties Rajani, Arka varieties and local varieties recorded least disease, with PDI ranging from 0-35.16 in the fields of Mandya, Hessaraghatta and Doddaballapur. Regarding the susceptible age of the plants for the disease, it was found that the plants which were more than 40 days old after transplanting were found to be more susceptible to the disease than the plants which were less than 40 days old.



a) Diseased fruits b) Healthy fruits

5 Photograph showing typical symptoms due to Alternaria solani on tomato fruits

Table 1. Severity of early blight of tomato in southern parts of Karnataka during 1991

Location	Taluk (district)	Variety	Age of the crop (in days)	Per cent disease index
Pinnenahalli	Gubbi(Tumkur)	Sonali	b	92.66
Gollahalli	"	Local	c	10.32
Thippur	"	Local	b	16.11
Hosakere	"	Rupali	c	78.52
Herehalli (CPCRI)	"	Rasmi	a	55.12
		Naveen	a	15.62
Doddabale	Hebbal Bangalore	Rupali	c	96.15
Jakkur	"	Naveen	b	60.12
Kengeri	"	Pusa ruby	c	61.88
Hessaraghatta (IIHR)	"	Arka Vikas	b	33.16
		Arka Vishal	a	11.12
		Arka Authi	c	6.82
Mandya	Mandya	Rajani	a	0.00
Budanur	"	Rasmi	b	25.16
Kolar	Kolar	Rupali	b	31.88
		Rajani	b	32.11
Doddaballapur	Doddaballapur	Roma	a	30.12
		Local	a	0.00

a=Below 40 days; b=40-50 days; c=Above 50 days.

4.3. ISOLATION AND IDENTIFICATION OF THE PATHOGEN

Standard tissue isolation technique was followed to obtain A.solani from the tomato leaves showing typical early blight symptoms collected from Jakkur (Bangalore), Gubbi (Tumkur), Budanur (Mandya), Kolar and Doddaballapur. Repeated isolations yielded a species of Alternaria. The description of the fungus isolated from different localities is as follows. The conidiophores formed singly or in groups, straight or flexous brown to olivaceous brown. The conidia were solitary, stright or slightly flexuous, oblong or ellipsoidal tapering to a beak pale or olivaceous brown, length 150-300 µm and 15-20 µm thick in the broadest part with 8-10 transverse and 0-4 longitudinal septa (Plate 6). The beaks were flexous, pale and sometimes branched. The description of this fungus agreed with the description given for A.solani by Commonwealth Mycological Institute, Kew, Surrey, England (Ellis, 1971). Thus the pathogen causing early blight of tomato has been identified as A.solani Sorauer.

4.3. PATHOGENICITY TEST

Pathogenicity test was carried out as described in 'Material and Methods' by inoculating with spore

6 Photograph showing typical conidia of *Alternaria solani*

syspension ($2x10^4$ spore/ml) and homogenized mycelial bits of A.solani (Isolate As_3) on foliage of 35 days old Pusa ruby variety of tamato. Fifteen days after inoculation the symptom appeared on inoculated leaves as brown, round to oval necrotic spots with concentric rings. The reisolated cultures from these artificially infected leaves leaves were similar to that of original culture. Plants which were not inoculated with the fungal spore suspension did not show any symptoms of the disease.

4.5. CULTURAL STUDIES

The cultural characters of all five isolates of A.solani were studied on six solid media as described in 'Material and Methods'. The results of the growth and sporulation of each of the five isolates are presented in Table 2 and Fig.1.

Growth: The five isolates of the fungus showed difference in their growth. The radial growth of all five isolates of A.solani was maximum of potato dextrose agar and oat meal agar with the colony diameter of 67.00 mm and 58.73 mm respectively (Plate 7a and 7b) followed by host extract agar (52.86mm) and Czpek's agar (52.13 mm). Least radial growth of the fungus was recorded on Lima bean agar (33.18 mm) and Richard's agar (46.39 mm).

Plate 7a. PDA

Plate 7b. Oat meal agar

7a&b Photograph showing growth characters of five isolates of Alternaria solani on PDA and oat meal agar

Table 2. Colony diameter (mm) and sporulation of the five isolates of *Alternaria solani* on different culture media after 8 days of incubation at 26±1°C

Media / Isolates	Colony diameter (mm) and sporulation+					
	AS1	AS2	AS3	AS4	AS5	Mean
Lima bean agar	22.00 ***	43.33 **	44.00 **	40.33 *	60.00 *	33.13
Host extract agar	39.00 -	55.00 -	55.33 -	55.00 -	60.00 -	52.86
Richard's agar	45.00 ****	43.33 **	42.00 **	60.00 **	41.66 **	46.39
Potato dextrose agar	70.00 *	70.00 *	60.00 -	65.00 -	70.00 -	67.00
Oat meal agar	43.66 -	60.00 *	55.00 *	65.00 -	65.00 -	58.73
Czpek's agar	50.00 -	55.00 -	55.00 -	48.66 -	52.00 -	52.13
Mean	44.94	54.44	51.88	55.66	58.11	51.70

	S.Em	CD(5%)
Isolates	2.94	5.26
Media	2.68	5.76
Isolate x media	6.58	7.90

+Note:
****=Excellent sporulation=20 and above spores/microscopic field
***=Good sporulation=15 to 19 spores/microscopic field
**=Moderate sporulation=10 to 14 spores/microscopic field
*=Poor sporulation=1 to 9 spores/microscopic field
-=No sporulation

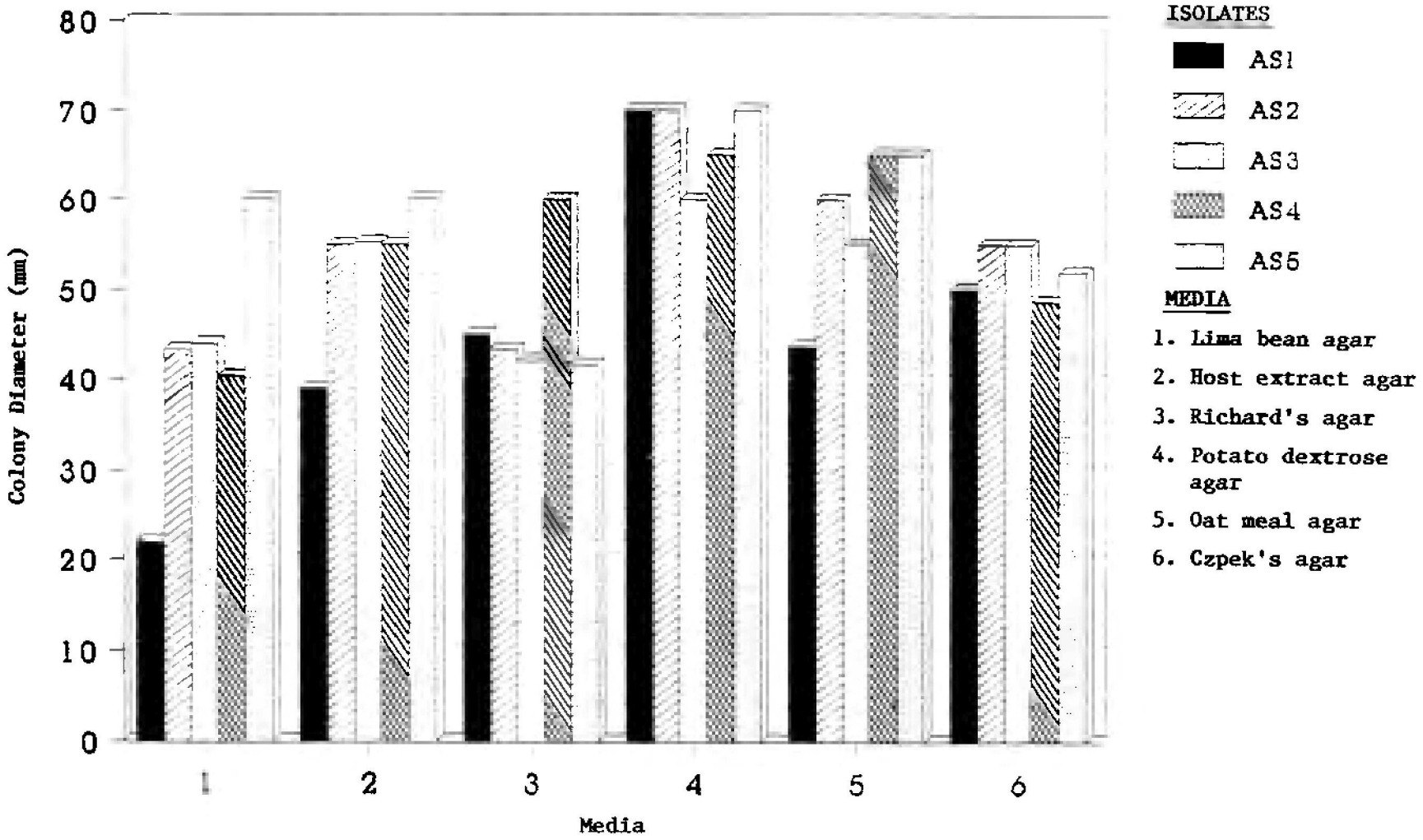


Fig.1: Effect of different solid media on growth of five isolates of Alternaria solani

Among the five isolates, maximum growth was noticed in isolate AS_5 (58.11 mm) which did not show significant difference over isolate AS_4 (55.66 mm) and AS_2 (54.40 mm), but significantly differed with the growth of isolate AS_3. Least radial growth was recorded in isolate AS_1 (44.94 mm).

Sporulation: All isolates of A.solani recorded moderate sporulation in Richard's agar. However, isolate AS_1 showed excellent sporulation. Isolates AS_1, AS_2 and AS_3 showed moderate sporulation on lima bean agar, but the isolates AS_4 and AS_5 produced poor sporulation on the same medium. Further, poor sporulation was recorded by isolate AS_1, AS_2 and AS_3 on potato dextrose agar and oat meal agar, while isolates AS_4 and AS_5 did not sporulate at all on them. All the isolates failed to sporulate on Czpek's and host extract agar.

Cultural characteristics: The morphological variation among five isolates of A.solani on different media are presented in Table 3. All the five isolates of A.solani produced dark brown to light white coloured colonies on potato dextrose agar. However, the growth on host extract agar was in the fashion of submerged mycelial

Table 3. Cultural characteristics of five isolates of [redacted] on different culture media after 8 days of incubation at 26±1°C

Isolates	Cultural characteristics		
Medium	Lima bean agar	Host extract agar	Richard's media
AS1	Mycelial colour white colour Substrate colour:Light greyish Margin:Irregular	Mycelial colour:Light grey Substrate colour:Greyish Margin:Irregular	Mycelial colour Cottony white Substrate colour:Light yellow Margin:Smooth
AS2	Mycelial colour Dirty white Substrate colour:Light yellow Margin:Smooth	Mycelial colour Light yellow Substrate colour:Light greyish Margin:Smooth	Mycelial colour:Dirty white Substrate colour:Light yellowish Margin.Smooth with brown coloured
AS3	Mycelial colour:Dirty white Substrate colour:Light yellow Margin:Smooth	Mycelial colour.Light yellow Substrate colour:Light greyish Margin:Smooth	Mycelial colour:Light brown Substrate colour:Light yellow Margin:Smooth
AS4	Mycelial colour:Dirty white Substrate colour:Brown Margin:Smooth	Mycelial colour:Greyish Substrate colour.Light greyish Margin:Smooth	Mycelial colour:Dirty white Substrate colour:Brown Margin:Smooth
AS5	Mycelial colour Brown Substrate colour:Light yellow Margin:Irregular	Mycelial colour.Light brown Substrate colour:Light brown Margin:Smooth	Mycelial colour.Light brown Substrate colour:Yellowish Margin:Smooth

Table 3 (Contd.)

Isolates	Cultural characteristics		
Medium	PDA	Oat meal agar	Czpek's media
AS1	Mycelial colour:Dark brown Substrate colour:Greyish Margin:Smooth	Mycelial colour.Light white Substrate colour:Greyish Margin:Smooth	Mycelial colour:Dirty white Substrate colour:Greyish Margin:Smooth
AS2	Mycelial colour:Dark brown Substrate colour:Brown Margin:Smooth	Mycelial colour:Light grey Substrate colour:Dirty greyish Margin:Smooth	Mycelial colour:Dirty white Substrate colour:Dirty greyish Margin:Smooth
AS3	Mycelial colour:Light brown Substrate colour:Light greyish Margin:Irregular	Mycelial colour:Dirty white coloured Substrate colour:Dirty greyish Margin:Smooth	Mycelial colour:Dark brown Substrate colour:Light yellow Margin:Smooth
AS4	Mycelial colour:White Substrate colour:Light greyish Margin:Irregular	Mycelial colour:White coloured Substrate colour:Light yellow Margin:Smooth	Mycelial colour:Dirty white Substrate colour:Light yellow Margin:Smooth
AS5	Mycelial colour:Dark brown Substrate colour:Greyish Margin:Smooth	Mycelial colour:Dirty white Substrate colour:Greyish Margin:Smooth	Mycelial colour:Dark brown Substrate colour:Dirty greyish Margin:Smooth

growth with yellow coloured substratum. On Richard's agar and lima bean agar the isolates of the fungus produced dull white colonies with entire margin. Whereas, on oat meal agar and Czpek's agar the mycelium was light brown colour with concentric rings at the centre of the colony. While the isolates AS_3 and AS_4 produced irregular colony margin on PDA, the isolates AS_1 and AS_5 produced such a colony margin on lima bean agar.

4.6. SPORE GERMINATION

The conidial germination of five isolates of A.solani were studied on different media viz., Tap water, Glucose 2%, sucrose 2%, Host extract, lima bean extract and Czpek's solution by employing 'hanging drop' technique as described in 'Material and Methods'. The results of the study are presented in Table 4.

Maximum spore germination was recorded in 2% glucose solution (93.90%) followed by germination in 2% sucrose (84.26%). Least germination percentage of spores were recorded in Czpek's solution (18.92%). Tap water and lima bean extracts recorded significantly more germination of spores over Czpek's solution. Maximum per cent spore germination was noticed in isolate

Table 4. Per cent spore germination of five isolates of *Alternaria solani* on different media after 8 hoiurs of incubation at 26±1°C

Medium / Isolates	Spore germination (per cent)					
	AS1	AS2	AS3	AS4	AS5	Mean
Lima bean	57.42	42.69	38.47	29.79	26.74	39.02
Glucose 2%	91.00	98.10	92.12	94.20	94.10	93.90
Sucrose 2%	81.20	79.80	84.10	89.40	86.84	84.26
Host extract	32.58	29.81	10.09	16.01	17.15	20.73
Czpek's broth	22.17	10.30	17.98	30.34	13.79	18.92
Tap water	58.42	77.98	48.40	47.35	68.15	60.06
Mean	57.13	56.11	48.52	51.18	46.67	52.81

	S.Em.	CD(5%)
Media	0.295	0.578
Isolates	0.330	0.647
Media x Isolates	0.660	1.294

AS_1 (57.13%) in all media tested, followed by isolate AS_2 (56.11%) and isolate AS_4 (51.18%).

4.7. PHYSIOLOGICAL STUDIES

4.7.1. Effect of temperature on growth and sporulation of A.solani

All the five isolates of A.solani were inoculated on 15 ml of potato dextrose agar in petriplates and incubated at 15, 20, 25, 30 and 35°C for 8 days. The average radial growth and sporulation of five isolates were recorded and presented in Table 5 and Fig.2.

All the five isolates gave maximum growth at 25°C (49.60 mm), followed by the growth at 20°C (40.53 mm) and 30°C (37.86 mm). Least radial growth was observed at 35°C (26.46 mm) which differed significantly over the growth at other temperatures. Among the five isolates of A.solani. Isolate AS_5 recorded maximum growth (42.80 mm), followed by isolate AS_3 (41.06 mm) and AS_4 (40.06 mm). Least radial growth was noticed in isolate AS_1 (27.19 mm).

Excellent sporulation of all the isolates except AS_2 was recorded at 30°C. However, the sporulation of all the isolates was found to be good at 35°C. Isolates AS_1 and As_2 recorded poor sporulation at 25°C, whereas,

Table 5. Mean colony diameter and sporulation of five isolates of Alternaria solani at different temperature after 8 days of incubation

Temperature(°C)	Mean colony diameter(mm) and sporulation+					
Isolates	AS1	AS2	AS3	AS4	AS5	Mean
15	24.00 -	27.66 -	31.00 -	30.00 -	30.00 -	28.53
20	24.66 -	35.00 -	42.00 -	52.00 -	49.00 -	40.53
25	35.00 *	41.00 *	55.00 -	51.00 -	65.00 -	49.00
30	32.00 ****	31.00 **	46.33 ****	41.00 ****	39.00 ****	37.86
35	19.33 ***	22.00 ***	31.00 ***	29.00 ***	31.00 ***	26.46
Mean	27.19	31.33	41.06	40.06	42.80	36.47

	S.Em.	CD(5%)
Isolates	0.363	0.711
Temperature	0.363	0.711
Isolate x Temperature	0.811	1.589

+Note:
****=Excellent sporulation=20 and above spores/microscopic field
***=Good sporulation=15 to 19 spores/microscopic field
**=Moderate sporulation=10 to 14 spores/microscopic field
*=Poor sporulation=1 to 9 spores/microscopic field
-=No sporulation

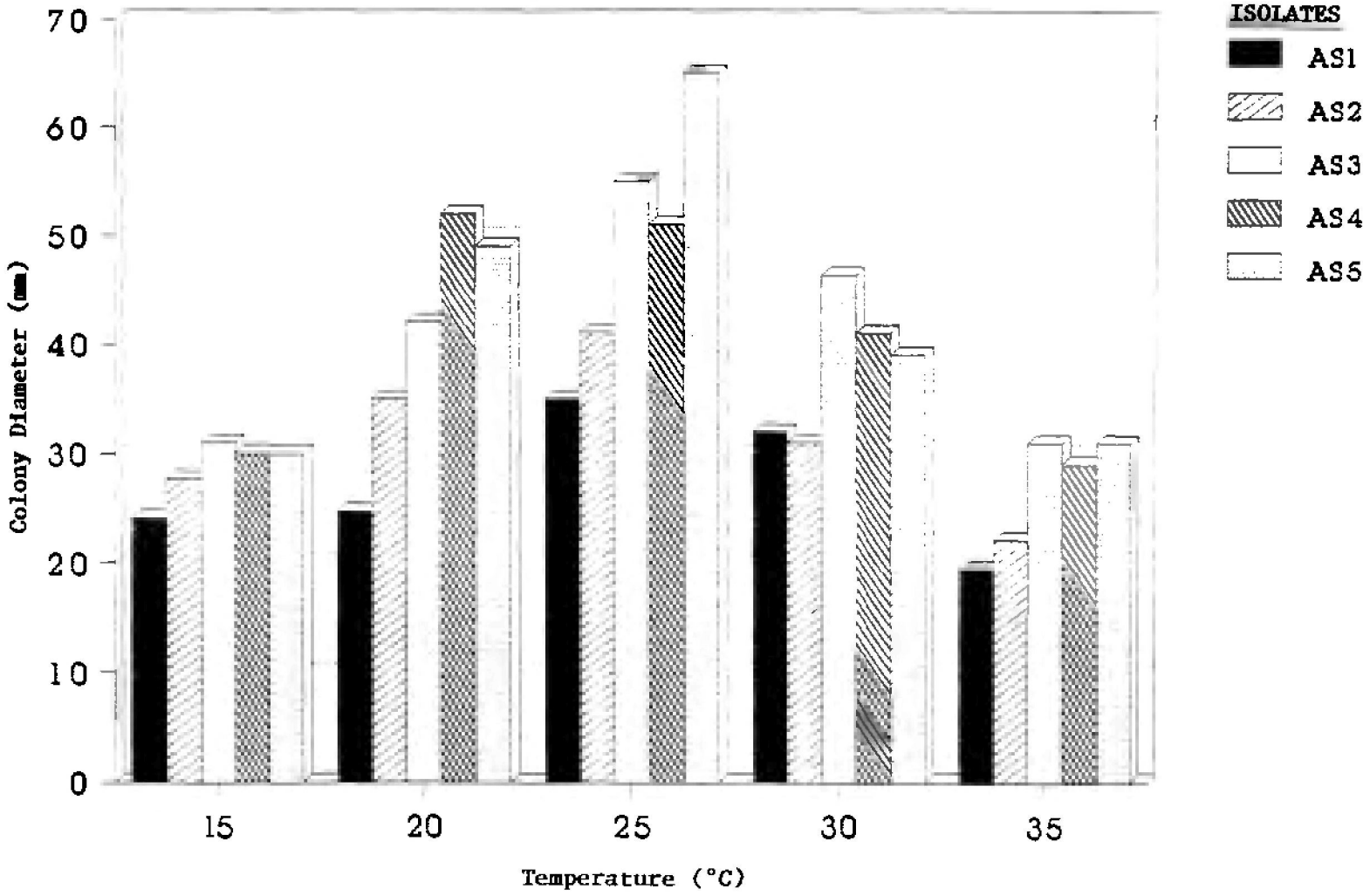


Fig.2: Effect of temperature on five isolates of Alternaria soloni

the other isolates did not sporulate at 25°C. None of the isolates sporulated at 20° and 15°C.

4.7.2. Effect of light on growth and sporulation of A.solani

This experiment was conducted to study the effect of light on the growth and sporulation of five isolates of A.solani. The isolates were inoculated to 15 ml of PDA in petriplates as described in 'Material and Methods' and exposed to alternate cycles of 12 hours light 12 hours of darkness and continuous light and continuous darkness for 24 hours for 8 days. The results are presented in Table 6 and Fig.3.

All the five isolates of A.solani gave maximum growth (59.1 mm) when inoculated petridishes were exposed to alternate cycles of light and darkness or continuous darkness. But, there was significantly reduced mycelial growth of the isolates, when inoculated plates were exposed to continuous light (Plate 8a and 8b).

Among the five isolates of A.solani, maximum growth of 64.33 mm was noticed in isolate AS_2 which differed significantly over all other isolates. Least significant radial growth of 37.83 mm was recorded in isolate AS_5.

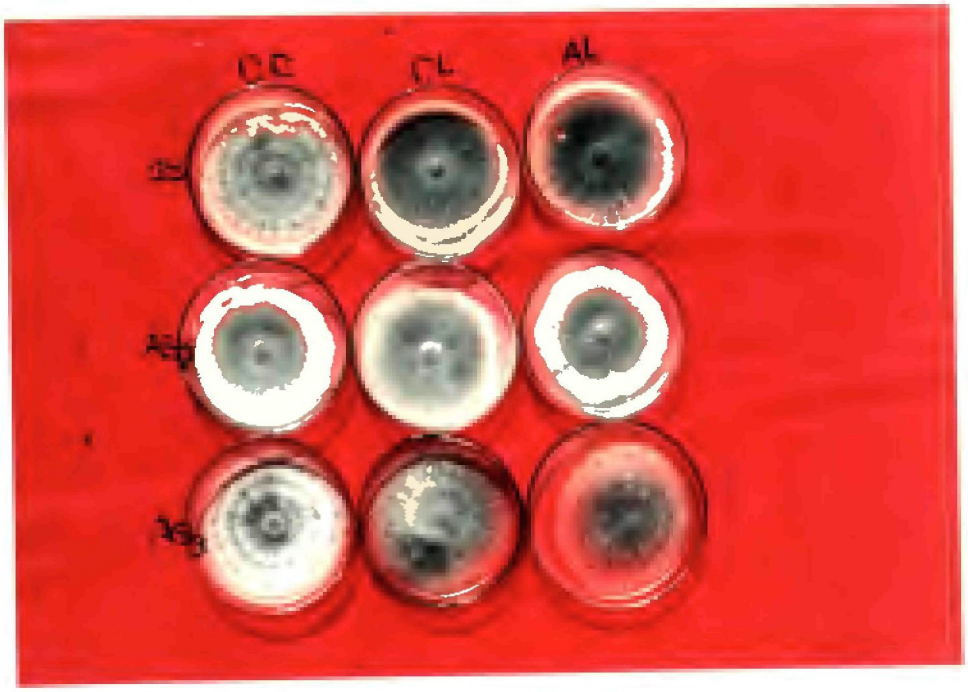


Plate 8a

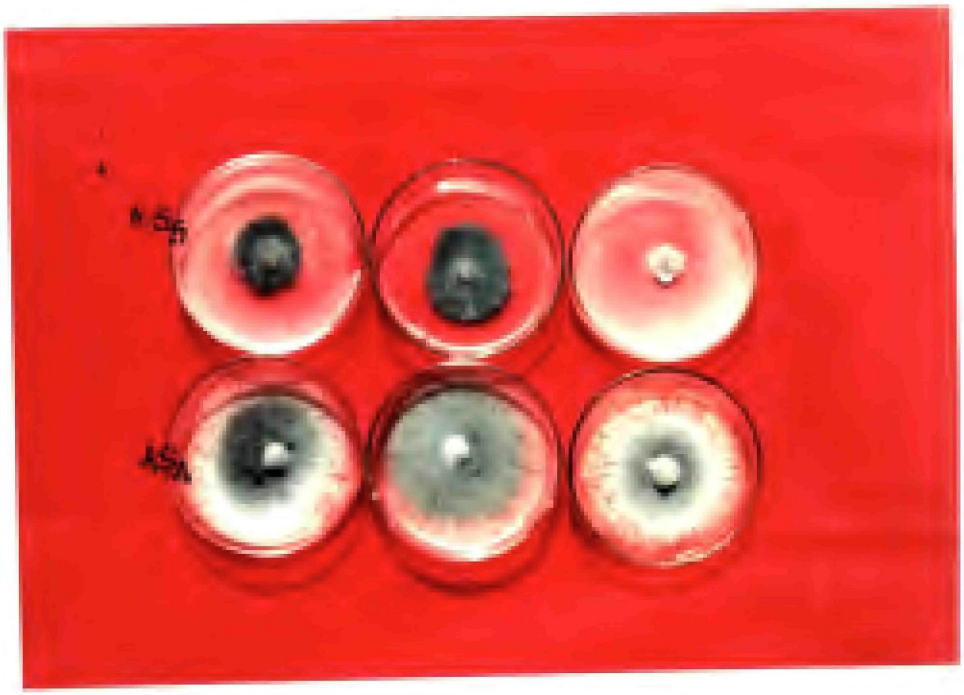


Plate 8b

8a&b Photograph showing the effect of light on growth of five isolates of Alternaria solani

Table 6. Mean colony diameter and sporulation of five isolates of Alternaria solani at different light intervals after 8 days incubation at 26±1°C

Isolates	Colony diameter(mm) and sporulation+			
Light interval(h)	12h light+ 12h dark	24h light	24h dark	Mean
AS1	39.00 ****	38.00 *	65.00 *	47.33
AS2	65.00 ****	63.00 *	65.00 *	64.33
AS3	61.50 ***	61.00 *	50.00 *	57.50
AS4	60.00 ***	50.00 *	40.00 *	50.00
AS5	70.00 ***	61.50 *	45.00 -	37.83
Mean	59.10	51.70	59.00	42.08

	S.Em.	CD(5%)
Isolates	0.714	1.399
Light	0.922	1.596
Isolate x Light	1.59	3.12

+Note:
****=Excellent sporulation=20 and above spores/microscopic field
***=Good sporulation=15 to 19 spores/microscopic field
**=Medium sporulation=10 to 14 spores/microscopic field
*=Poor sporulation=1 to 9 spores/microscopic field
-=No sporulation.

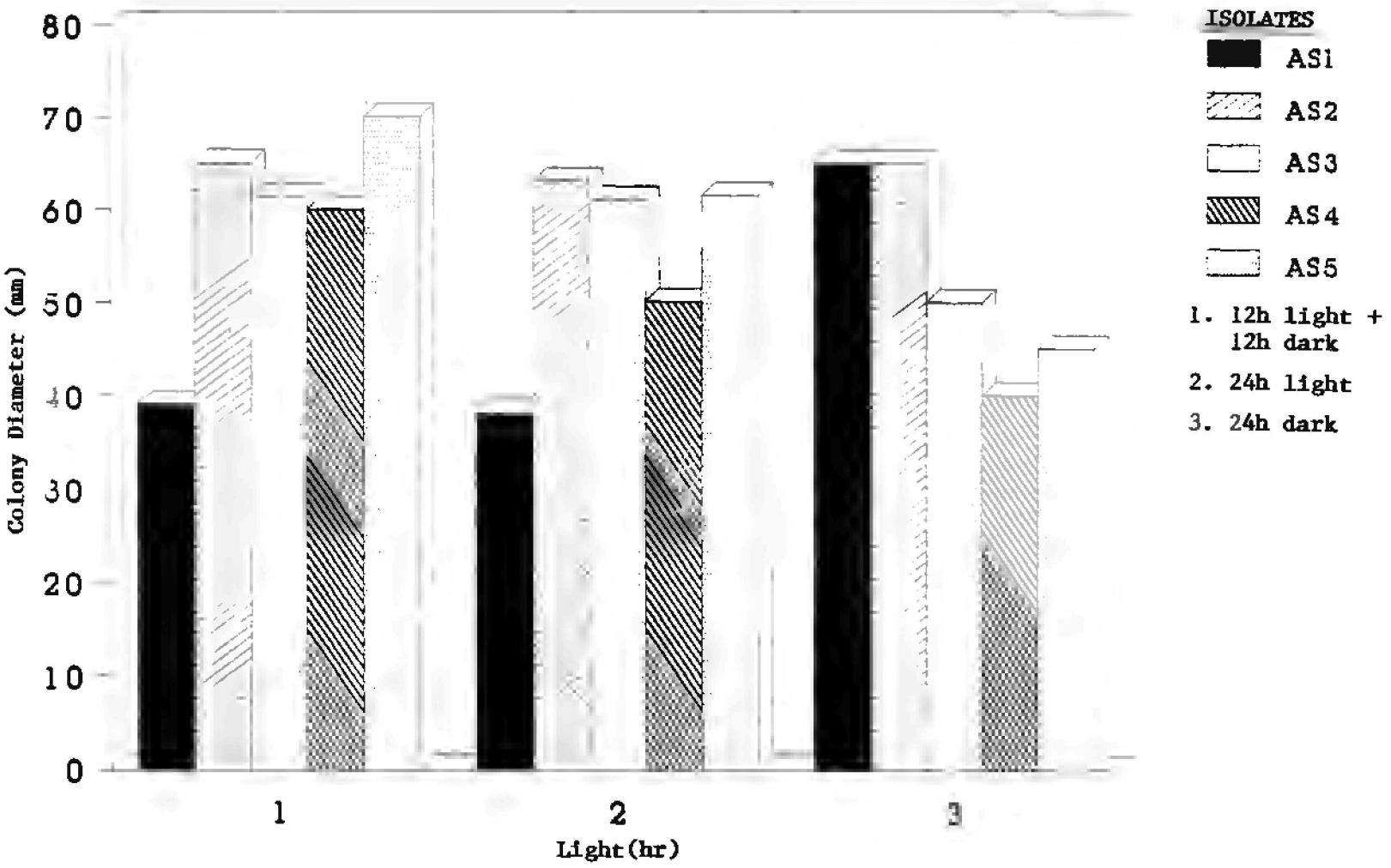


Fig.3: Effect of light on five isolates of Alternaria solani

All the five isolates of A.solani recorded excellent to good sporulation when exposed to alternate cycles of light and darkness and poor sporulation was recorded when exposed to continuous darkness and continuous light.

5. PATHOGENIC VARIATION

Pathogenic variation among five isolates of A.solani was studied on 30 days old Pusa Ruby tomato variety by artificial inoculation. The results of the experiments giving the number of spots per leaflet and lesion size are presented in Table 7.

Maximum number of spots per leaf was recorded by isolate AS_3 (6.02) followed by isolate AS_2, AS_1, AS_5. The least number of spots per leaflet was noticed in isolate AS_4 (1.83). However, there was wide variation in lesion size among five isolates of A.solani. Maximum lesion size of 17.23 mm on leaves was noticed by isolate AS_2, followed by isolate AS_5 and isolate AS_3 with lesion sizes of 14.23 mm and 11.70 mm respectively. Least lesion size on leaves was recorded in isolate AS_4 (10.66 mm).

Table 7. Pathogenic variation of five isolates of *Alternaria solani* on Pusa Ruby variety of tomato

Isolates	No.of spots/ leaflet	Lesion size (mm)
AS1	5.77	11.01
AS2	5.85	17.23
AS3	6.02	11.70
AS4	1.83	10.66
AS5	5.04	14.23
S.Em.	1.029	3.18
CD(5%)	3.356	4.37

6. SURVIVABILITY

6.1. Survivability of A.solani in seeds

Seeds collected from infected tomato fruits were made use for the study of survivability of A.solani in them. Seeds thus collected were plated on water agar (1%) at an interval of 20 days and observed for the growth of fungus on them as explained in 'Material and Methods' and the per cent seeds showing the fungus are presented in Table 8 and Fig.4.

Maximum recovery of infected seeds (88.66%) was recorded in case of freshly infected tomato seeds. As the test continued to 160 days at an interval of 20 days, the recovery of A.solani from infected seeds drastically reduced from 82.23% to 18.45%. No recovery of the fungus was recorded beyond that period. The fungal growth completely covered the seeds without allowing it to germinate.

6.2. Survivability of A.solani in host debris

The data on the recovery of A.solani in the isolations from the infected host debris over a period of 180 days at an interval of 20 days is presented in Table 8 and Fig.4.

Table 8. Survivability of *Alternaria solani* in infected plant debris and seeds under laboratory conditions

Intervals (days)	Survivability (%)	
	Seeds	Plant debris
0	88.66	100.00
20	83.23	100.00
40	80.88	100.00
60	78.47	79.59
80	76.14	68.29
100	67.27	51.00
120	55.58	34.88
140	34.21	21.69
160	18.45	9.95
180	-	5.02
S.Em.	0.71	0.67
CD(5%)	2.13	2.03

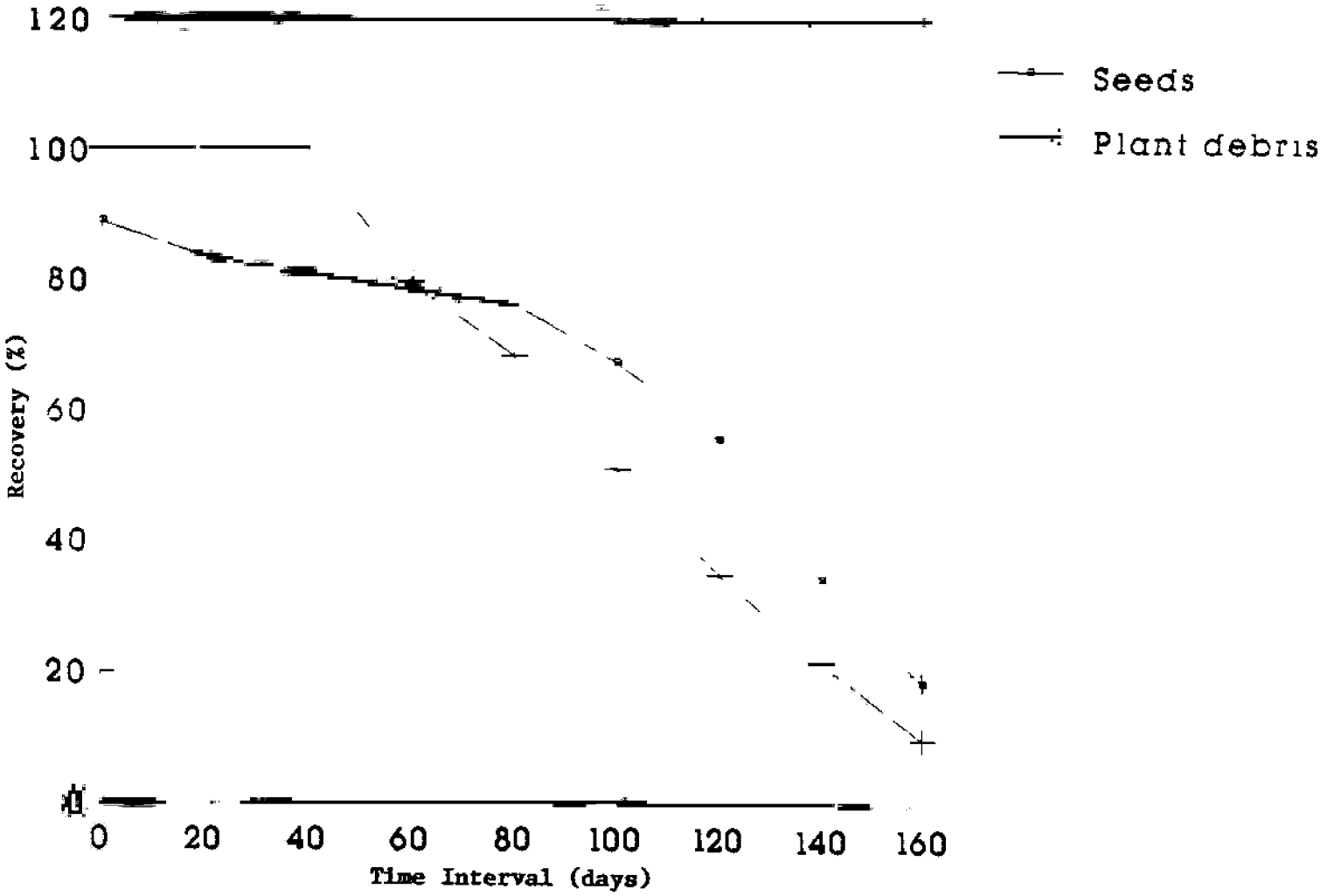


Fig.4: Survival of Alternaria solani in infected seeds and plant debris under laboratory condition

Maximum recovery (100%) of the fungus was recorded when the isolations were made from the freshly infected plant debris. As the isolations were continued at an interval of 20 days, a decline in recovery percentage of pathogen from the host debris was recorded. At the end of 180 days the recovery percentage of the fungus from the infected plant debris was only 5.02 per cent.

6.3. The survivability of A.solani in soil

The studies on the survival of A.solani was carried out in both sterilized and unsterilized soils, at different moisture levels. The extent of survival of A.solani was determined based on the development of seedling collar rot symptoms (Plate 9), when the tomato seeds were sown over a period at an interval of 25 days after the pathogen was inoculated to soil. The data on per cent collar rot infected tomato seedling are presented in Table 9.

Collar rot infection of over 90% was recorded in case of a trial wherein, the seeds were sown simultaneously as soon as the soils (both sterilized and unsterilized) were inoculated with the pathogen. As the experiment was continued out over a period of 200 days at

9 Photograph showing collar rot infection on tomato due to *Alternaria solani*

Table 9. Effect of moisture levels on the survivability of Alternaria solani in sterilized and unsterilized soil

Interval(days)	Seedling collar rot (per cent)									
	Sterilized soil MHC(%)					Unsterilized soil MHC*(%)				
Moisture level(%)	10	30	50	70	90	10	30	50	70	90
0	100.00	100.00	90.00	100.00	91.00	100.00	100.00	90.00	100.00	100.00
20	100.00	100.00	88.12	90.00	90.00	100.00	100.00	90.00	81.00	91.00
40	94.12	98.14	88.00	90.00	76.14	90.00	100.00	79.00	76.00	72.00
60	94.66	98.11	86.12	85.13	28.00	81.00	93.00	68.00	69.14	32.00
80	91.00	95.00	86.00	68.00	-	76.14	86.86	44.33	38.44	-
100	48.16	89.19	53.18	44.18	-	39.86	71.43	36.43	21.00	-
120	22.88	58.26	32.14	22.18	-	-	59.16	18.28	5.04	-
140	-	34.00	32.14	12.12	-	-	53.81	13.12	-	-
160	-	26.68	31.15	-	-	-	53.81	-	-	-
180	-	12.66	10.58	-	-	-	-	-	-	-
200	-	8.82	-	-	-	-	-	-	-	-

*MHC=Moisture holding capacity.

an interval of 25 days, there was decline in the seedlings of tomato developing collar rot infection. However, this decline in collar rot infection varied with moisture level of soils. Maximum survivability of the pathogen over a period of 200 days in case of sterilized soil (Fig.5) and 160 days in case of unsterilized soil (Fig.6) was recorded at 30% moisture level. Least survivability of the fungus was noticed at 90% moisture level in both sterilized and unsterilized soils.

7. HOST RANGE STUDIES

The ability of A.solani to survive on different hosts in the absence of the main host was studied by inoculating the spore suspension of the fungus ($2x10^4$ spore/ml) to host plants belonging to different species as explained in 'Material and Methods'. Observation on the development of symptoms was made 15 days after inoculation and the reaction of the pathogen on different hosts is presented in Table 10. Species belonging to Solanacea family such as Capsicum annuum Lycopersicon lycopersicum, Nicotiana glutinosa, Solanum melongena and Solanum tuberosum, produced mild to moderate symptoms when they were inoculated by the fungus without wounding and severe symptoms produced by wound inoculation. However, mild to moderate symptoms were

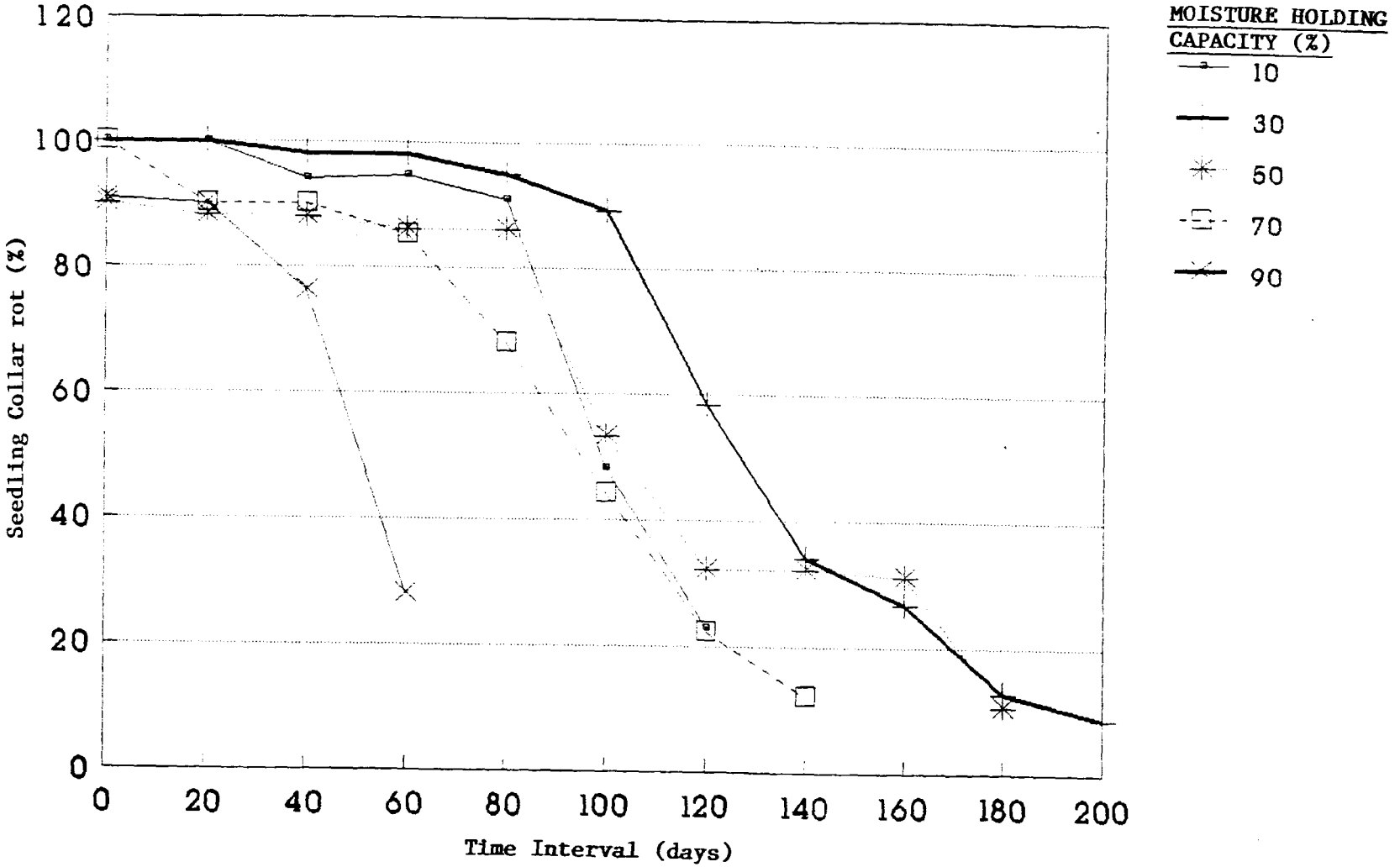


Fig.5: Survival of Alternaria solani in sterilized soil at different moisture level

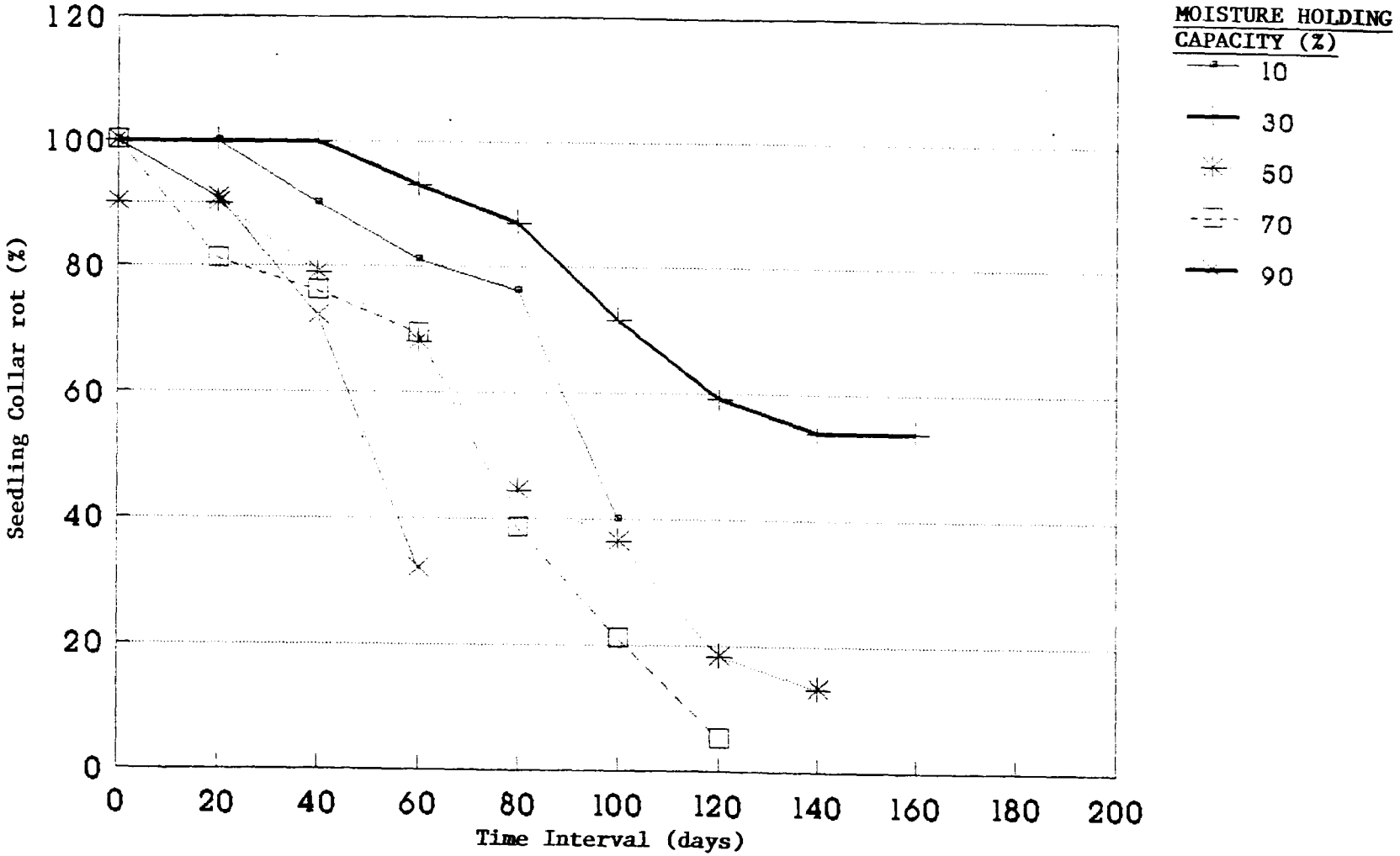


Fig.6: Survival of Alternaria solani in unsterilized soil at different moisture levels

Table 10. Reaction of different host plants to artificial inoculation with *Alternaria solani*

Family	Scientific name	Symptoms obtained	
		Uninjured	Injured
Amaranthaceae	*Amaranthus paniculatus* L.	-	+
Apiaceae	*Coriandrum sativum* L.	-	-
	Dacus carota L.	-	-
Asteraceae	*Carthumus tinctorus* L.	-	-
	Helianthus annuus L.	-	-
	Tagetes erecta L.	-	-
Brassicaceae	*Brassica oleracea* var. *capitata* L.	-	+
	Raphanus sativus L.	-	-
Cucurbitaceae	*Lagenaria siceraria* L.	-	+
	Momordica chorantia L.	-	++
Euphorbiceae	*Ricinus communis* L.		++
Pabaceae	*Cajanus cajan* (Linn.) Mill sp.		++
	Crotolaria juncea L.	-	+
	Pisum sativum L.	-	+
	Vicia faba L.		+
Amaryllidaceae	*Allium cepa* var. *cepa* L.	-	-
Malvaceae	*Abelmoschus esculentus* L.	-	++
	Gossypium herbaceum L.	-	++
Pedaliaceae	*Sesamum indicum* L.	-	+
Solanaceae	*Capsicum annum* L.	++	+++
	Lycopersion lycopersicum Mill.	++	+++
	Nicotiana glutinosa L.	+	++
	Solanum melongena L.	++	+++
	Solanum tuberosum L.	+	+++

Note: -=No symptoms; +=Mild symptoms; ++=Moderate symptoms; +++=Severe symptoms.

recorded in case of hosts viz., Amaranthus paniculata, Lagenaria siceraria, Crotolaria juncea, Pisum sativum, Vicia faba, Sesamum indicum, Momordica chorantia, Ricinus communis, Cajanus cajan, Abelmoschus esculentus and Nicotiana glutinosa, when they were inoculated by the fungus only after the hosts were injured/wounded.

8. VARIETAL REACTION

Twenty one popular tomato varieties were screened against A.solani in the green house as explained in 'Material and Methods' and the results on varietal reaction against the pathogen are given in Table 11.

The results indicated that varieties Sonali, Vishali, IAHS 881, IAHS 882, Rajani and all Arka varieties produced a resistant reaction against the fungus. Whereas, the varieties viz., Roma, Punjab Kesari, Rasmi and Naveen showed moderately resistant reaction. The variety Pusa Ruby showed highly susceptible reaction and the varieties Safed, Rupali and Punjab Choura showed moderately susceptible reaction. No variety was immune to the fungus.

9. CHEMICAL CONTROL

In vitro evaluation of fungicides on the growth of A.solani

The in vitro evaluation of different fungicides against A.solani was made at 100, 250, 500 and 1000 ppm

Table 11. Reaction of tomato varieties to artificial inoculation with *Alternaria solani* in glass house after 15 days of inoculation

Reaction	Per cent disease index	Varieties
Immune(leaf area infected)	0	-
Resistant	1-10	Arka alok Sonali Arka authi Vishali Arka Sourabh IAHS 881 IAHS 882 Arka abha Arka Vishali Arka Vikas Rajani Arka Ashish
Moderately resistant	11-15	Roma Punjab Kesari Rasmi Naveen
Moderately susceptible	16-20	Safal Rupali Punjab Choura
Susceptible	21-25	Pusa Ruby dwarf
Highly susceptible	>26	Pusa Ruby

by adopting "poisoned food technique". The results are presented in Table 12. The effect of different fungicides at different concentrations and their interactions were found to be statistically significant. The fungal growth was totally inhibited in all the four concentrations of Mancozeb and Iprodione. Captafol gave better inhibition of fungus (10.00 mm) at 500 ppm than the other fungicides viz., Captan (20.00 mm), Carboxin (17.66) and Chlorothalonil (17.66), when the growth in control (without fungicides) plates was 70.16 mm. Least inhibition of the fungus was recorded in case of Carbendazim (Plate 10a and 10b).

10. FIELD EVALUATION OF FUNGICIDES AGAINST A.Solani

Fungicides which gave good inhibitory effect against A.solani under laboratory conditions were evaluated in the field against early blight of tomato during 1991 and 1992 at a farm in Jakkur near University of Agricultural Sciences, GKVK Campus, Bangalore.

The experimental design, the fungicides and the number of spray treatments given against the disease are given in 'Material and Methods'.

10.1. 1991 Cropping season

The fungicides viz., Iprodione (at 0.1, 0.2, 0.3 and 0.4%), Mancozeb (0.2%), Captafol (0.2%) and

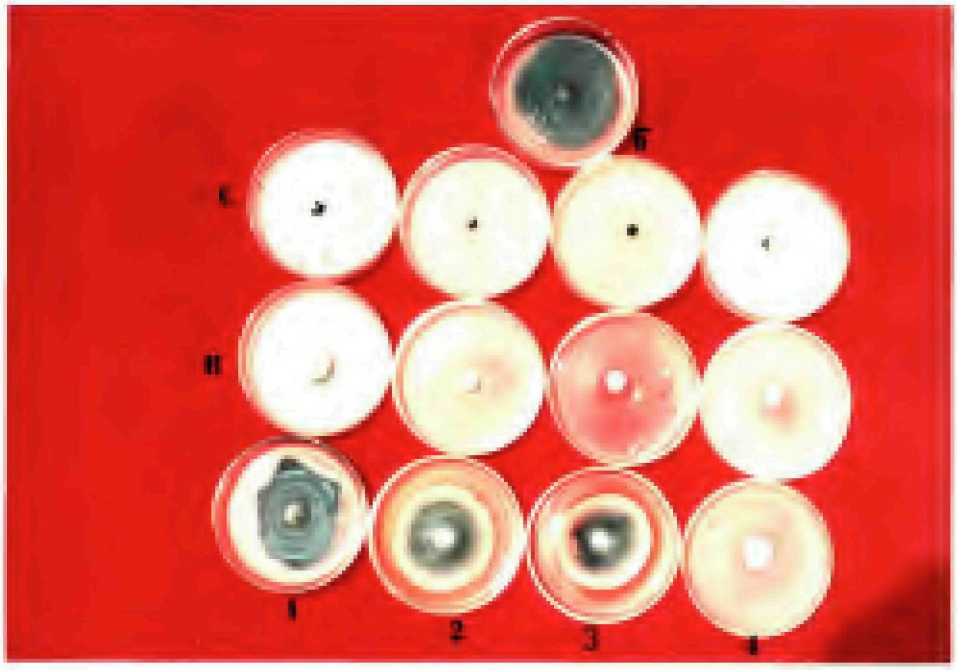


Plate 10a

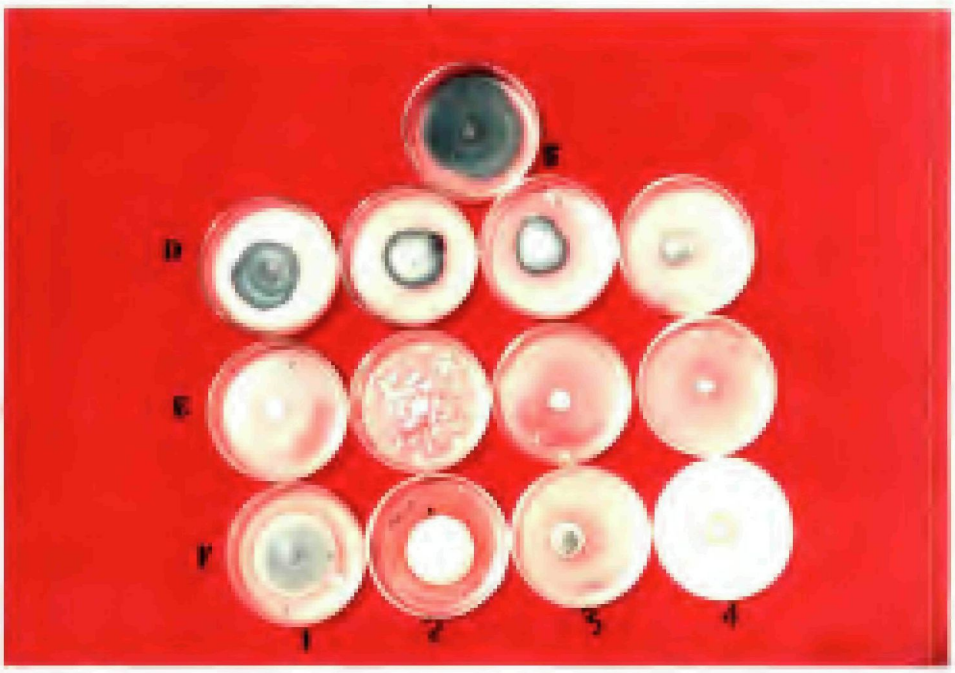


Plate 10b

10a&b Inhibition on mycelial growth of Alternaria solani by different fungicides at different concentrations

Table 12. Inhibition of mycelial growth of *Alternaria solani* by different fungicides after 8 days of incubation

Fungicide	Mycelial growth(mm)				
Concentration(ppm)	100	250	500	1000	Mean
Carbendazim	46.00 (6.81)	32.00 (5.70)	30.00 (5.55)	26.60 (5.05)	44.89 (5.78)
Captafol	22.33 (3.48)	14.00 (3.13)	10.00 (2.91)	0.00 (2.96)	11.58 (3.12)
Chlorothalonil	41.33 (6.43)	29.00 (5.42)	17.66 (4.24)	15.00 (4.41)	26.74 (5.13)
Captan	33.00 (5.78)	23.00 (4.84)	20.00 (4.48)	11.66 (3.47)	21.91 (4.64)
Iprodione	0.00 (0.70)	0.00 (0.70)	0.00 (0.70)	0.00 (0.70)	0.00 (0.70)
Mancozeb	0.00 (0.70)	0.00 (0.70)	0.00 (0.70)	0.00 (0.70)	0.00 (0.70)
Vitavax	34.33 (5.89)	30.00 (5.52)	17.66 (4.25)	11.00 (3.38)	23.24 (4.76)
Control	70.66 (8.43)	70.00 (8.43)	70.00 (8.43)	70.00 (8.43)	70.16 (8.43)
Mean	29.49 (4.74)	24.50 (4.25)	19.12 (3.83)	15.33 (3.31)	

	S.Em.	CD(5%)
Fungicides	0.087	0.171
Concentration	0.058	0.114
Fungicides x concentration	0.175	0.343

Data in parenthesis is angular transformed values.

Carbendazim (0.1%) were the spray treatments given in the field. Four sprays of the fungicides were given in both the years at an interval of 30, 45, 60 and 75 days after transplantating. The disease severity expressed as per cent disease index (PDI) recorded before the commencement of each spray and a final observation on 90th day (15 days after the final spray) is presented in the Table 13. The disease progress curve in different fungicidal treated plots and control plot are presented in Fig.7. Data revealed that the PDI was in the range 8.03 to 10.98 in the experimental plots before the first fungicidal spray was given. This range in the disease index did not differ significantly in the plots meant for different treatments. But in subsequent sprays all fungicidal treated plots recorded significantly less disease index over control.

On 90th day (i.e., 15 days after the final spray), maximum disease control with 25.76 PDI was recorded in the Iprodione (0.2%) treated plots, followed by control of the disease in Iprodione 0.3%, 0.4% and Mancozeb (0.2%) sprayed plots. The disease control in Iprodione (0.3 and 0.4%) did not significantly differ with Iprodione (0.2%) treated plots. Further, Iprodione 0.3 and 0.4% treated plots, the plants expressed

Table 13. Effect of foliar sprays of different fungicides on the disease severity of leaf blight, fruit stalk and fruit infection of tomato caused by Alternaria solani and fruit yield during 1991

Treatments	Per cent disease index (PDI)					Fruit stalk infection (per cent)	Fruit infection (per cent)	Fruit yield (tons/ha)
	30	45	60	75	90			
Iprodione 0.1%	9.66	17.87	30.20	34.67	36.96	7.33	0.00	17.80
Iprodione 0.2%	10.98	15.44	19.27	24.81	25.76	1.81	0.00	24.13
Iprodione 0.3%*	11.06	14.49	18.76	24.92	26.10*	1.57	0.00	18.86
Iprodone 0.4%*	10.62	15.96	17.76	22.25	26.05*	0.79	0.00	16.89
Mancozeb 0.2%	8.03	16.42	18.57	26.60	35.79	0.53	0.00	19.60
Captafol 0.2%	10.62	21.15	21.80	34.36	42.90	2.61	0.00	18.80
Carbendazim 0.1%	10.79	19.29	39.10	45.77	58.23	8.63	2.59	16.80
Control	10.49	28.48	18.95	55.66	79.44	10.35	3.90	16.21
S.Em.	1.12	1.56	1.61	1.43	1.71	0.5863		1.66
CD(5%)	3.20	4.47	4.62	4.09	4.90	1.7246		0.56

*Phytotoxic effect was noticed.

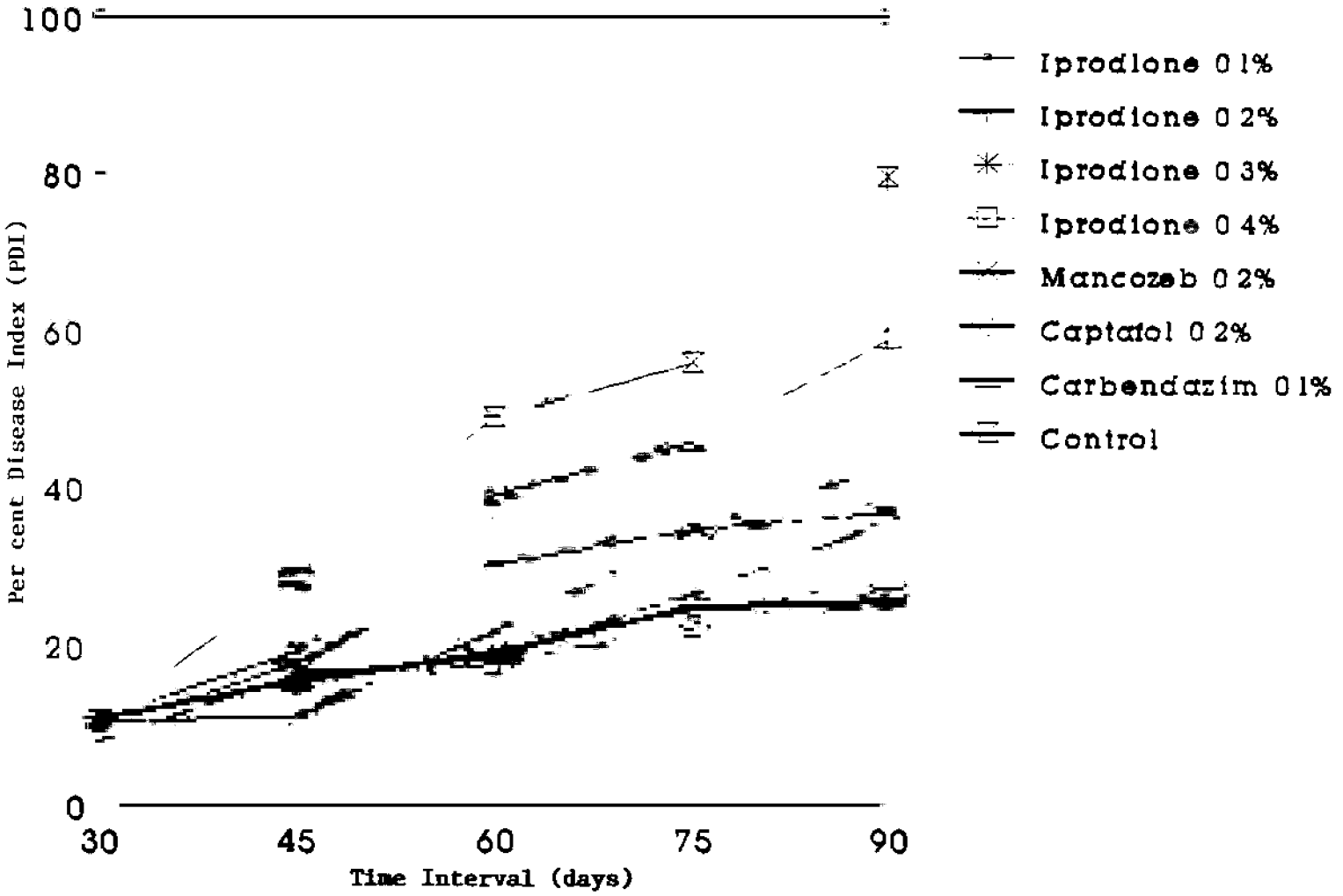


Fig.7: Effect of foliar spray of different fungicides on control of early blight of tomato during 1991

phytotoxic effect (Plate 11). Least reduction of the disease was recorded in Carbendazin 0.1% treated plots with PDI of 58.13. While, in PDI in control plots it was 79.44.

The data also revealed that in all treatments there was an increase in disease index from 30th day to 90th day. However, the rate of increase in per cent disease index was slow in case of fungicidal treated plots when compared to check plots.

10.2. Fruit stalk infection

The data on fruit stalk infection recorded during 1991 is presented in Table 13.

All fungicidal spray treatments showed significantly less fruit stalk infection (0.53 to 8.63%) over control (10.38%). However, maximum reduction in fruit stalk infection of tomato was recorded in 0.2% Mancozeb sprayed plots (0.53% infection), which was on par with the infection in 0.4% Iprodione treated plots. Least control of fruit stalk infection was noticed in 0.1% carbendazim sprayed plots (8.63% infection).

10.3. Fruit infection

There was no Alternaria fruit infection was noticed in fungicide sprayed plots except in 0.1%

a) Healthy leaf b) Leaf showing Iprodione phytotoxicity

11 Photograph showing phytotoxicity of Iprodione on tomato leaf

carbendazim treated plots, wherein 2.59 per cent fruit infection was recorded while, the fruit infection in control plots was 3.90% (Table 13).

10.4. Fruit yield (tons/ha)

The data on fruit yield recorded during 1991 is presented in Table 13.

There was significantly higher fruit yield in fungicidal spray treatments over control. However, maximum yield (24.13 tons/ha) was obtained in 0.2% Iprodione sprayed plot, followed by 0.2% Mancozeb and 0.3% Iprodione sprayed plots with the yield of 19.60 tons/ha and 18.86 tons/ha respectively. Least fruit yield (16.80 tons/ha) was obtained in 0.1% Carbendazim sprayed plots.

10.5. 1992 cropping season

During 1992 cropping season also the same fungicides evaluated during 1991 were tested in the same field. The data on their efficacy against the disease is presented in the Table 14 and the disease progress curve is presented in Fig.8.

The per cent disease index (PDI) in the experimental plots was in the range of 8.80 to 10.90

Table 14. Effect of foliar sprays of different fungicides on the disease severity of leaf blight, fruit stalk and fruit infection of tomato caused by Alternaria solani and on fruit yield during 1992

Treatments	Per cent disease index (PDI)					Fruit stalk infection (per cent)	Fruit infection (per cent)	Fruit yield (tons/ha)
	30	45	60	75	90			
Iprodione 0.1%	10.66	18.33	30.20	46.51	66.55	21.24	2.80	15.27
Iprodione 0.2%	8.85	10.10	20.25	23.85	28.15	5.14	1.90	23.99
Iprodione 0.3%	10.42	21.35	22.86	26.31	30.00*	4.48	1.50	17.71
Iprodone 0.4%	9.27	14.84	20.61	25.00	30.44*	3.89	0.70	15.52
Mancozeb 0.2%	9.76	15.97	24.44	29.03	42.20	2.58	0.40	19.88
Captafol 0.2%	10.90	15.69	24.97	39.75	52.99	10.58	1.90	16.77
Carbendazim 0.1%	8.60	18.63	37.14	48.29	68.36	26.82	3.20	14.38
Control	9.84	38.83	54.21	69.62	96.44	28.36	6.30	14.16
S.Em.	1.06	1.60	2.04	1.63	2.14	1.7577		1.53
CD(5%)	3.14	4.51	5.02	4.29	5.33	5.1704		0.43

*Phytotoxic effect was noticed.

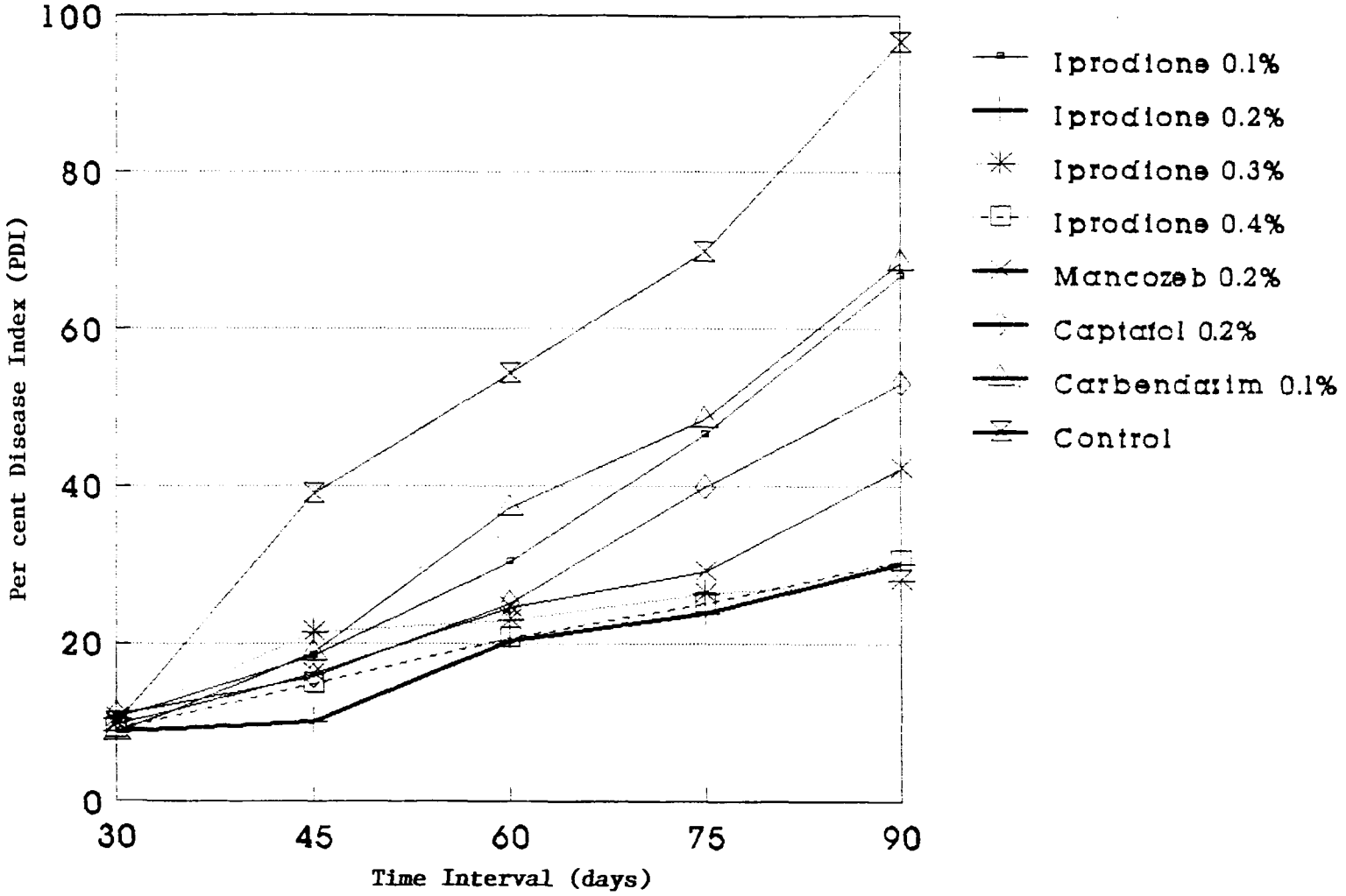


Fig.8: Effect of foliar spray of different fungicides on control of early blight of tomato during 1992

before the first spray was given which did not differ significantly. On 90th day (15 days after the final spray) maximum control of the disease with least PDI of 28.00 was recorded in 0.2% Iprodione treated plots followed by the disease control in Iprodione 0.3, 0.4% and Mancozeb (0.2%). However, the disease index in Iprodione 0.2%, 0.3% and 0.4% did not differ significantly with each other. The plants in Iprodione 0.3 and 0.4% treated plots showed phytotoxic symptoms (Plate 11). Least control of the disease was recorded in Carbendazim (0.1%) treated plots 68.36 PDI. Whereas in check plots was 96.44.

The data also showed that in all treatments there was increase in disease index from 30th day to 90th day. However, the rate of increase in PDI was slow in case of fungicide treated plots compared to PDI in controlled plots.

10.5.1. Fruit stalk infection

The data on fruit stalk infection was recorded during 1992 is presented in Table 14.

All fungicidal spray treatments showed significantly less fruit stalk infection (2.58 to 21.24%) over control (28.36%). However, least fruit stalk

infection was recorded in 0.2% Mancozeb sprayed plots (2.58%) which was on par with 0.4% Iprodione (3.89%). Maximum fruit stalk infection was noticed in Carbendazim sprayed plots (26.82%).

10.5.2. Fruit infection

The fruit infection was less in case of all thefungicide treated plots (0.4 to 3.2%) compared to control (6.3%). Maximum control of fruit infection (0.4%) was recorded by 0.2% Mancozeb. Least control of fruit infection was recorded in 0.1% Carbendazim sprayed plots with fruit rot infection of 3.2% (Table 14).

10.8. Fruit yield (tons/ha)

The data on fruit yield recorded during 1992 is presented in Table 14.

There was significant increase in fruit yield in fungicidal treated plots over control. However, maximum yield (23.99 tons/ha) was obtained in plots sprayed with 0.2% Iprodione, followed by 0.2% Mancozeb and 0.3% Iprodione sprayed plots with the yield of 19.88 tons/ha and 17.71 tons/ha respectively, while, the fruit yield in check plots was 14.16 tons/ha. Least fruit yield was obtained in 0.1% Carbendazim sprayed plots.

10.9. Economics of fungicidal sprays

Economics of 4 sprays of different fungicides against early blight of tomato during 1991 and 1992 is presented in Table 15.

Cost of fungicidal spray was maximum in case of 0.4% Iprodione (Rs.5620/ha), followed by 0.3% Iprodione (Rs.4290/ha) and 0.2% Carbendazim (Rs.3016.0/ha). The cost involved in four sprays of 0.2% Mancozeb is Rs.1151.2/ha which is least compared to all other fungicidal treatments.

Net return per rupee is highest in 0.2% Mancozeb treatment (Rs.7.30 and 14.90 during 1991 and 1992 respectively), followed by 0.2% Iprodione (6.68 and 9.96), 0.2% Captofol (Rs.3.27 and 3.71) and 0.1% Iprodione (Rs.2.43 and 2.05) during the years 1991 and 1992 respectively. Least net return per rupee was obtained by 0.1% Carbendazim treatment.

11. MELANIN EXTRACTION

The abilityy of A.solani to produce melanin pigment at different ages of the culture was studied by growing the fungus on potato broth as explained in 'Material and Methods'. The data are presented in Table 16.

Table 15. Cost-Benefit ratio of fungicidal sprays in the control of leaf blight, fruit stalk and fruit infection of tomato due to *Alternaria solani*

Fungicides	Dose in kg/ha	1991						1992			
		Quantity required for 4 sprays (kg)	Cost of fungicidal spray including labour charge for 4 sprays (Rs.)	Yield tons/ha	Yield increase over control (tons/ha)	Net return due to chemical spray	Net return per rupee	yield tons/ha	Yield increase over control (tons/ha)	Net return due to chemical spray	Net return per rupee
Iprodione 0.1%	0.7 kg a.i/ha	2.80	1630.00	17.80	1.59	3975.00	2.43	15.27	1.11	3330	2.05
Iprodione 0.2%	1.4 kg a.i/ha	5.60	2960.00	24.13	7.92*	19800.00	6.68	23.99	9.83	29490	9.96
Iprodione 0.3%	2.1 kg a.i/ha	8.40	4290.00	18.86	2.65	6625.00	1.54	17.71	3.55	10650	2.48
Iprodione 0.4%	2.8 kg a.i/ha	11.20	5620.00	16.89	0.68	1700.00	0.30	15.52	1.36	4080	0.72
Mancozeb 0.2%	1.4 kg a.i/ha	5.60	1151.00	19.60	3.39	8475.00	7.36	19.88	5.72	17160	14.9[illegible]
Captafol 0.2%	1.4 kg a.i/ha	5.60	2105.70	18.80	2.59	6475.00	3.07	16.77	2.61	7830	3.71
Carbendazim 0.1%	0.7 kg a.i/ha	2.80	3016.00	16.80	0.59	1475.00	0.48	14.38	0.22	660	0.21
Control	-	-	-	16.21	-	-	-	14.16	-	-	-

* Note:
Labour cost=Rs.25/day Tomato fruit=Rs.2.5/kg=1991
Iprodione =Rs.475/kg Rs.3.0/kg=1992
Mancozeb =Rs.152/kg
Carbendazim=Rs.485/kg
Captafol =Rs.322/kg

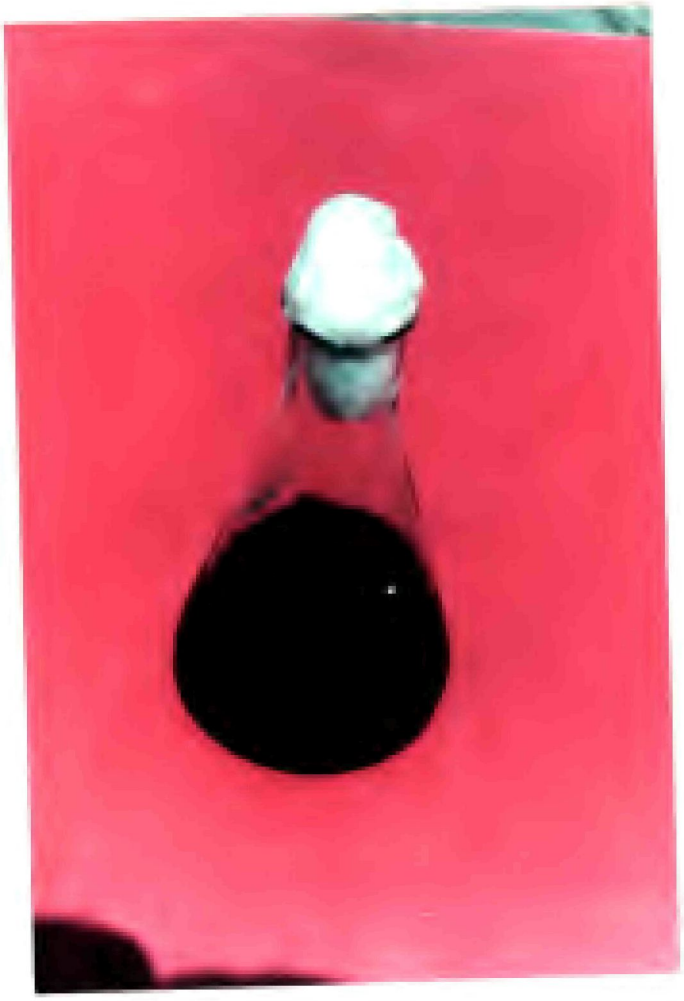
Table 16. Per cent absorption of light due to melanin pigment extracted from *Alternaria solani* at different ages

Age of the culture (days)	Per cent absorption
5	0.065
7	0.231
9	0.521
11	0.912
13	1.890
15	1.899
17	1.921
19	1.955
21	1.985

The data revealed an increase in the production of melanin with the increase in the age of the fungus. The least per cent absorption of light (0.065%) due to melanin pigment was recorded in 5 day old culture. However, the per cent absorption of light increased to 1.985% in 21 days old culture indicating the increased production of melanin with increased age of the fungus (Plate 12). At 5th day of incubation, the hyphae of A.solani were light brown, but mycelium turned to dark brown colour in 21 days old culture.

11.1. Interaction of melanin with fungicide

This experiment was undertaken to know the effect of melanin produced by A.solani on the efficacy of the fungicides against a fungus. For this purpose, a hyaline fungus like F.oxysorum f.sp. cubens was taken as a test fungus to test the efficacy of Mancozeb with or without a melanin. The experiment was carried out as explained in the 'Material and Methods'. The observation on colony diameter and cultural characteristics of the fungal growth in various treatments viz., melanin without fungicide, melanin with fungicide, fungicide without melanin and neither melanin nor fungicide. The data are presented in Table 17.



12 Photograph showing Melanin pigment produced by Alternaria solani

Table 17. Effect of melanin on the efficacy of mancozeb against the growth of *Fusarium oxysporum* f.sp. *cubens* after 8 days of incubation at 26±1°C

Treatments	Colony diameter (mm)	Per cent inhibition
1. Mancozeb(ppm)		
100	40.00	38.46
200	9.00	86.15
250	0.00	100.00
2. Mancozeb(ppm)+Melanin(ml)		
250+1	30.00	53.84
250+2	38.00	41.53
250+3	42.00	35.38
250+4	58.00	10.76
3. Melanin(ml)		
1	45.00	30.76
2	46.00	29.23
3	45.00	30.76
4	45.00	30.76
4. Control	65.00	-

The data showed that the growth of Fusarium was totally inhibited at 250 ppm concentration of Mancozeb. Whereas, when 250 ppm of Mancozeb was mixed with 4 ml of melanin the growth was 58 mm, which was on par with the growth of the fungus when melanin (4 ml) alone was added to culture medium and also neither melanin nor fungicide was present in the media. Similar results of increased growth of the fungus was recorded, when it was grown in medium containing 100 or 200 ppm of Mancozeb along with melanin at various concentrations (Table 17), indicating the reduction in inhibitory activity of the fungicide in the presence of melanin. However, there was not much variation in growth of the fungus at different quantities of melanin when compared to control. The test fungus produced dirty white, loose mycelium with yellow pigmentation at the under surface of the Fusarium colony in plates with 250 ppm of Mancozeb with 4 ml melanin whereas, in case of culture plates with 4 ml melanin alone it produced compact mycelium with yellow pigmentation at the under surface of the colony (Table 18).

Table 18. Morphology of colonies of Fusarium oxysporum f.sp. cubens due to effect of different concentrations of melanin and with mancozeb

Mancozeb			
100	200	250	Control
Colony dirty white, semi compact mycelium. Light yellow pigmentation on the under surface of the colony	Colony white, sparse mycelial growth	No growth	Colony white aerial cottony loose mycelium elevation of mycelium at centre sunflower like radiations under the surface of the colony
Mancozeb+Melanin			
250 ppm+1 ml	250 ppm+2 ml	250 ppm+3 ml	250 ppm+4 ml
Colony white, sparse radiating mycelial growth	Colony dirty white, loose radiating mycelial growth, dirty yellow pigmentation on the under surface	Colony white aerial cottony loose mycelium sunflower like radiation under the surface of the colony	Colony dirty white, loose mycelium, dome like at ceotre, yellow pigmentation under the surface of the colony
Melanin			
1 ml	2 ml	3 ml	4 ml
Colony white, semi compact, mycelium, depression at centre	Colony ditty white, compact mycelium, depression at centre	Colony white, aerial cottony loose mycelium, sunflower like radiation under the surface of the colony	Colony dirty white, compact mycelium, yellow pigmentation under the surface of the colony

DISCUSSION

V.DISCUSSION

The early blight of tomato caused by Alternaria solani Sorauer is an important disease leading to complete defoliation of the plant resulting in drastic reduction of fruit yield. In recent years, the disease has assumed serious proportions in Karnataka and several complaints have been reported from the farmers regarding the improper control of the disease by the recommended management practices made at present. Therefore, an investigation was carried out on various aspects of the pathogen and the disease viz., variation in the pathogen, host range, survivability, varietal reaction to pathogen, evaluation of chemicals against the disease both under laboratory and field conditions and studies on the production of melanin and its role in the efficacy of fungicides.

The symptoms on tomato plants in the field were first noticed on the older leaves as minute brown to black necrotic spots of one to two mm in diameter. Often these spots enlarged with concentric ridges to produce characteristic target board effect. Later upward progression of the disease was observed and leaves dried up and drooped down. Walker (1952) gave similar

description of symptoms on tomato due to A.solani. The disease on stems, petioles and calyx appeared as brown to dark brown, elongated to oval cankerous spots and on fruits at stem end as black or brown sunken lesions which later enlarged to the extent they involved the whole fruits which ultimately led to their decay. Datar and Mayee (1985) and Ramakrishnan et al. (1971) also recorded such symptoms on stems, petioles and fruits.

The severity of early blight of tomato expressed as per cent disease index varied from 0 to 96.15% as evident in the survey conducted during 1991 in various parts of southern Karnataka. Similar observations were made by Datar and Mayee (1981), who reported severe epidemics of A.solani having a coefficient of disease index of 71.66%. Further, the severity of the disease was maximum on varieties viz., Rupali, Sonali and Pusa ruby at different locations.

The fungus was isolated from leaves showing typical early blight symptoms by following standard tissue isolation technique. The pathogenicity tests were carried out by inoculating spore suspensions ($2x10^4$ spores/ml) with mycelial bits of fungus on 30 days old Pusa ruby tomato variety. Symptoms appeared on older

leaves 15 days after inoculation as brown necrotic spots with concentric ridges at the centre. Similar technique was followed by Andrus et al. (1945) to prove the pathogenicity of A.solani on tomato. The reisolated fungus yielded a species of Alternaria. The study on the characteristics of five isolates of Alternaria obtained from different localities indicated that the conidiophores of the fungus formed singly or in groups, straight or flexous, brown to olivaceous brown. The conidia were solitary, stright or slightly flexuous, oblong or ellipsoidal tapering to a beak-pale or olivaceous brown, smooth, 150-300 um in length and 15-20 um thick in the broadest part with 8-10 transverse and none or few (1 to 4)longitudinal septa. The beaks were flexous, pale and sometimes branched. The description of the fungus agreed with the description given for A.solani Sorauer by Commonwealth Mycological Institute, Kew, Surrey, England (Ellis, 1971). Thus the pathogen causing early blight of tomato has been identified as Alternaria solani Sorauer.

Growth on different media and the cultural characteristics are often been taken as a guide for differentiating strains of the pathogens in general. In the present investigation five isolates of A.solani were grown on six solid media to study the variation in growth

and cultural characteristics. Among six media used for growth and sporulation of A.solani, all the five isolates produced maximum growth on potato dextrose agar, followed by oat meal agar which may be attributed to complex nature of natural media supporting good fungal growth. In a similar experiment, Gemwat and Ghosh (1979) reported that among seven solid media tested for the growth of A.solani, potato dextrose agar showed good growth and excellent sporulation after 10 days of incubation. Bonde (1929) observed profuse production of aerial mycelium of A.solani on potato dextrose agar. Neergaard (1945) and Rotem (1966) also recorded potato dextrose agar as the best medium for A.solani.

Among the five isolates A.solani, isolate AS_5 recorded maximum growth, which did not differ significantly over isolates AS_4 and AS_2. However, isolate AS_3 showed significantly more growth over isolate AS_1.

Maximum sporulation of the pathogen was recorded on Richard's agar, lima bean agar and potato dextrose agar. Barksdale (1968) also noticed more sporulation of A.solani on lima bean agar and potato dextrose agar. Whereas, Gemwat and Ghosh (1979) recorded good growth and

excellent sporulation on Richard's media. Joshi (1981) and Mahabaleswarappa (1981) observed that potato dextrose agar and Richard's agar were good media for growth and sporulation of Alternaria spp. viz., A.gomphrena Togashi and A.carthami infecting Gomphrena globosa L. and Carthamus tinctorus L. respectively. Morphological variations such as colony colour, colony margin and substrate colour were noticed among the isolates of A.solani. Several workers notably, Bonde (1929), Henning and Alexander (1959), Rotem (1966) and Kaul and Saxena (1988) also observed differences in cultural characters like growth rate, type of growth, colony colour, colour of the substrate and sporulation among ninety two isolates of A.solani.

Cochrane (1958) has opined that any consideration of ecology or spread of economically important fungi must take spore germination into account. Alternaria spores are air borne and are thus obvious sources of secondary infection by the pathogen. An attempt was made to study the factors which determine the germination of the spores. In the present study, among the different media tested for spore germination of Alternaria solani maximum percentage germination of spores was recorded in glucose (2%) and sucrose (2%) solutions after eight hr.

of incubation indicating the preference of the fungus for sugars (mono and disaccharides) for the germination. Hanumanthaiah (1976) observed that the spores of A.tenuissima (Fries) Wiltshire showed maximum germination in many nutrient solutions and host extracts. Desai (1979) observed that germination of A.macrospora Zimm. was maximum in 2% sucrose solution.

Temperature is the most important physical environmental factor regulating vegetative and reproductive activity of the fungi. Studies on the influence of temperature on growth of five isolates of A.solani showed maximum growth at 25°C. Kaul and Saxena (1988) also reported the temperature of 25°C for the good growth of A.solani. Chowdhury (1944) observed maximum growth of A.carthami at 30°C and 25°C. Bonde (1929) studied that growth rate of A.solani ranged from 15-40°C and observed maximum growth of the fungus at 25°C. In the present study, among five isolates of A.solani AS_5 and AS_3 produced significantly more growth, than isolate AS_4, AS_2 and AS_1 at different temperatures.

Maximum sporulation of the fungus was observed at 30°C followed by the same at 35°C. Verma (1970) and Bonde (1929) reported a wide temperature range of 25-30°C

for good sporulation in Alternaria solani. Hemperson and Maude (1982) observed that the optimum temperature for sporulation in the temperature range of 18-24°C for A.brassicae (Berk.) Sacc. and 20-30°C for A.brassicola (Schw.) Wiltshire.

Light has profound effect on growth and sporulation of fungi, which has been amply demonstrated by many workers (Leonion, 1924; Drayton, 1937; Barnett and Lilly, 1950 and Timmick et al., 1951). The preliminary studies carried out in the present investigation with A.solani indicated a maximum growth and sporulation, when the inoculated plates were exposed to alternate light and dark conditions (12 hr. light alternated with 12 hr. dark) followed by continuous dark. Similar experiment conducted by Luckens (1963) on A.solani found that conidia of fungus normally begin to form sixth hours in the dark. Whereas Zhu et al. (1985) observed that A.solani sporulates profusely on cornmeal agar when illuminated with fluorescent lamp for 8 hours at 18°C. Fencelli and Kimati (1990) observed that maximum growth and sporulation of A.dauci (Khuhn.) Groves and Sholko when exposed to light. Among the isolates AS_2 and AS_3 gave maximum growth and sporulation than the other three isolates when exposed to alternate light

treatment. This may be due to the inducement of certain metabolic processes necessary for growth and sporulation of the fungus which usually does not occur in light. Similar type of observation has been reported by Lilly and Barnett (1951).

A study on the pathogenicity of the strains of A.solani on tomato revealed that considerable differences exist in their ability to infect tomato leaves. The five isolates used in these tests, were all pathogenic and they produced symptoms when inoculated on tomato plants. However, maximum number of spots per leaflet and lesion size was observed in isolate AS_3 and AS_2. Isolate AS_4 was less pathogenic causing only a few small lesions.

In the foregone studies, the five isolates of A.solani were subjected to various studies with a sole intension of understanding the extent of variability among them. The results gave an indication that the isolates varied significantly with regard to more than one parameter, when subjected to different media, temperature, light and relative pathogenicity tests indicating the existence of variations among the isolates.

The survivability of an organism depends on its capacity to survive in an adverse environment and its host range, so that it can subsist in the absence of the main host. A.solani was isolated from more than 80 per cent of the seeds, stored under laboratory condition (ambient temperature 26+1^{o}C) on 40th day. A slow and steady decrease of recovery percentage occurred in subseaquent isolations at 20 days intervals and on 160th day of storage, the fungus recovery was only 18.45 per cent from seeds. Seed-borne nature of A.solani has been reported by Groves and Sholka (1944), Neergaard (1945) and Walker (1952).

The fungus was isolated from more than 50 per cent infected host bits stored under laboratory condition on 100 day of storage. A slow and steady decrease of recovery percentage occurred in subsequent isolations at 20 days intervals and on 180th day of storage, the recovery percentage was only 5.02 per cent. The survivability of A.solani in host debris in soil has been reported by Basu (1974b), McCarter et al. (1976) and Moore and Thomas (1942).

Seedling collar rot technique was employed to determine the survivability of A.solani in sterilized and

unsterilized soil at different moisture levels. Collar rot developed on more than 90% of the tomato seedlings at the begining when soil contaminated with A.solani. The collar rot infection decreased when tested at 20 days planting interval, the pathogen able to infect tomato seedling upto 200 days at 30% moisture holding capacity, followed by 50% moisture holding capacity (140 days) conginial for A.solani to survive. Similar trend in survival were observed in both sterilized and unsterilized soil at 30% moisture level by Chambers and Elentje (1969) in case of Ophiobolus graminis Sacc. and Vidyasekaran (1971) while working with Helminthosporium nodulasum (Berk and Curt) Sacc. and H.tetramera McKenney. In the present study A.solani could not infect tomato seedlings in unsterilized soil after 160 days. However, pathogen was able to survive for longer period at 30% moisture holding capacity. This low survivability in unsterilized soil could be due to mutual antagonism in soil fungi (Rangaswamy and Vidyasekaran, 1963 and Vidyasekaran and Rangaswamy, 1969). Further, the low survivability at 90 per cent moisture holding capacity could be due to lack of aeration or due to accumalation of carbon dioxide and ammonia. Combe (1960) suggested that the high carbon dioxide content in soil leads to the elimination of the fungi from soil.

The cereals, legumes, oilseeds, pulses, vegetables and a few weeds have been reported by various workers such as Neergaard (1945), Rands (1917b) and Walker 1952) as host plant for Alternaria spp. In the present study 24 plant species belong to eleven families were inoculated to leaves with or without wounding to know whether these plants take infection. None of the plants produced symptoms without wound inoculation except species belonging to solanaceae. On wound inoculation, symptoms developed on almost all plants though differing in reaction. Mild to moderate symptoms were noticed on Capsicum annuum, Lycopersicon esculentum, Solanum melongena and Solanum tuberosum, when A.solani was inoculated to unwounded leaves. However, severe symptoms were noticed when the pathogen was inoculated to wounded leaves. Alternate and Collateral hosts play a role in perpetuation of plant pathogens. In the present study many plant species got infection on wounding and the fungus was recovered by tissue isolation again. Wounds on plants is a normal phenomenon in nature which occur on plants either mechanically or by the activity of insects, thus opening the door for infection. However, how far, such infected plants are going to serve as source of A.solani inoculum and for its perpetuation from year to year are to be investigated.

In an effort to find out the source of resistance, 21 popularly growing tomato varieties werescreened for their reaction against the disease. Out of these varieties, only twelve varieties were resistant (1-10 PDI) and four were moderately resistant (11-15 PDI) to the pathogen. While, the varieties Safed, Rupali and Punjab Chhoura were moderately susceptible (16-20 PDI), Pusa ruby was found to be highly susceptible (26-30 PDI) to the pathogen. The reaction of these varieties needs to be confirmed by field evaluation under heavy inoculum densities. Several workers such as Basu (1974a), Carrasco et al. (1977), Sridhara and Naik (1983) reported resistant varieties of tomato against A.solani.

Evaluation of fungicides in vitro is a handy tool to screen a larger number of fungicides. In the present studies, the in vitro evaluation of fungicides by 'food poison technique' revealed that Iprodione and Mancozeb were found to be highly effective in completely inhibiting the growth of A.solani. Rajagopal and Vidyasekaran (1983) reported Mancozeb and Captafol to be very effective against A.solani. Barris and Mariotto (1986) reported that Iprodione reduced mycelial growth at 1 ppm and inhibit the sporulation at 10 ppm or 100 ppm in Helminthos orium sativum.

Experiments were carried out in the field to control the early blight disease of tomato by chemical sprays during 1991 and 1992 at a farm in Jakkur near University of Agricultural Sciences, GKVK Campus, Bangalore. Chemicals viz., Iprodione (at 0.1, 0.2, 0.3 and 0.4%), Mancozeb (0.2%), Captafol (0.2%) and Carbendazim (0.1%) were the spray treatments given in the field.

Data on disease severity during 1991 showed that all the chemical sprayed plots significantly reduced the disease over control. However, maximum disease reduction was recorded in Iprodione (0.2%) with per cent disease index of 25.76, followed by Iprodione 0.3 and 0.4%, Mancozeb (0.2%) with per cent disease index of 26.10, 26.05 and 35.79 respectively, when the per cent disease index in control was 79.44 on 90th day. Iprodione when sprayed at 0.3 and 0.4% concentration phytotoxicity on leaves was noticed. Maximum control of fruit stalk infection and fruit infection was noticed in Mancozeb 0.2% sprayed plot with 0 and 0.53% of infection respectively, when fruit stalk and fruit infection in control was 10.35 and 3.90% respectively.

The data on fruit yield recorded during 1991 indicated that there was significant increase in fruit

yield in fungicides spray treatments over control. Maximum yield (24.13 tons/ha) was obtained by Iprodione 0.2% sprayed plots, followed by Mancozeb 0.2% (19.60 tons/ha), while, the yield in control plots was 14.16 tons/ha.

Data on disease severity during 1992 also indicated maximum disease reduction in Iprodione 0.2% sprayed plots. Although Iprodione at 0.3 and 0.4% also gave better control of the disease, phytotoxic effect on leaves was recorded during 1992 also. Again, maximum reduction in fruit stalk infection and fruit infection was obtained in Mancozeb 0.2% sprayed plots during 1992 also. However, maximum yield of 23.99 tons/ha was obtained in Iprodione (0.2%) sprayed plots, followed by Mancozeb (0.2%) with the yield of 19.88 tons/ha, while the yield in control plots was 14.16 tons/ha.

Thus, the data on field experiments on control of early blight disease of tomato during 1991 and 1992 revealed the maximum control of Alternaria blight disease of tomato when the plots were sprayed with Iprodione (0.2%) followed by Mancozeb (0.2%). Several workers have reported the efficacy of Iprodione at various concentrations (Hemperson and Maude, 1982; Bedlan, 1987;

Hedges and Cole, 1988 and Aponyi et al., 1988) and Mancozeb (Johnson, 1969; Mathur and Shekhawat, 1986 and Sengupta and De, 1987) in the better control of diseases caused by Alternaria spp. on several crops.

The economic analysis of different fungicidal spray schedules indicated that additional gain per rupee invested was highest in case of 4 sprays of Mancozeb (0.2%) which was Rs.7.36 and Rs.14.90 during 1991 and 1992 respectively followed by the Iprodione 0.2% with net gain of Rs.6.68 and Rs.9.96 during the said years respectively.

Melanins are polymeric pigments produced by a wide variety of organisms. Fungal structures having pigment extremely resistant to harsh environmental conditions. It is significant that most of these resistant structures contain black pigments which have been described as melanins. Number of workers notably Lockwood (1960), Linderman and Toussoun (1966), Bloom Field and Alexander (1967), and Kuo and Alexander (1967) have commented upon the relationship between the presence of melanin or melanine like pigments in the cell walls of fungi and resistance to microbial lysis. However, reports on detailed analysis of isolation of fungal

melanins are very few. In the present investigation a preliminary studies on melanine indicated that the production of melanin was more with the increased age of the culture. Similar observation of increased production of melanine in an aged culture, when compared to an young culture of a fungus has been reported by several workers, while working with different groups of fungi (Chet et al., 1967; Bartnicki-Garcia and Reyes, 1964; Lingappa et al., 1963).

There are few reports in the literature that the fungal melanin will come in the way of fungicides (Schwinn et al., 1979). Thus in the present investigation the melanin produced by A.solani was tested to understand its effect on the fungicidal efficacy using F.oxysporum f.sp. cubens (being a hyaline fungus) as test organism and Mancozeb as a test fungicide. The investigation revealed that Mancozeb gave one hundred per cent inhibition of F.oxysporum f.sp. cubens at 250 ppm in the laboratory. When the fungicide at this concentration was mixed with melanin at various proportions (1-4 ml), the inhibition of the fungus by Mancozeb was drastically reduced as indicated by the growth of the fungus (30 to 58 mm growth at different proportions of melanin). The growth of the Fusarium fungus was also affected to some

extent when it was grown in melanin containing media (without the fungicide) as compared to the growth in the control (media without melanin and fungicide). Thus it can be concluded that the presence of melanin may affect the inhibitory effect of fungicides. Several workers such as Fryoyd et al. (1978) and Schwinn (1979) reported that several antipenetrant fungicides such as Tricyclozole, Pyroquilon, Chlorobenthiazone and rabicid compounds have no curative effects against melanin producing fungi. However, a detailed investigation has to be taken up with regard to the fungicides that can domelanize the melanin pigment in A.solani both in vitro and in vivo for developing an effective chemical against the disease. Before this the mode of interference of Melanin with fungicides and the range of fungicides they are affected by Melanin interference has to be investigated.

SUMMARY

VI. SUMMARY

An investigation on early blight of tomato caused by Alternaria solani was carried out with reference to survey of the disease, variability studies of the pathogen, survivability, host range, varietal reaction to pathogen, evaluation of chemicals against the disease both under laboratory and field conditions and studies on the production of melanin and its role on the efficacy of a fungicide.

A survey conducted in southern Karnataka during 1991 revealed that the per cent disease index ranged from 0 to 96.15. Maximum severity (96.15 PDI) was recorded on the variety Rupali at Bangalore. Rajani and some local varieties were free from disease at Mandya and Doddaballapur.

Five isolates of A.solani obtained from the tomato leaves showing typical early blight were collected from Jakkur (Bangalore), Gubbi (Tumkur), Budanur (Mandya), Kolar and Doddaballapur. The fungus on PDA produced dark brown mycelium and brown to olvaceous brown conidiophores with conidia having transverse and longitudinal septa with long beak. The description of the fungus agreed with the description given by

Commonwealth Mycological Institute, Kew, Surry, England (Ellis, 1971). Thus the pathogen causing early blight of tomato has been confirmed as A.solani. Successful pathogenicity of the fungus on tomato was obtained by inoculating the spore suspension.

All the five isolates produced maximum growth on PDA and sporulated well on Richard's media. Isolate AS_5 and AS_4 recorded more growth than the other three isolates on all the media tested.

Five isolates of the pathogen produced maximum growth at 25°C. Isolates AS_5, AS_3 and AS_4 produced significantly more growth than Isolates AS_1 and AS_2 at all temperatures tested.

All the five isolates recorded maximum growth and sporulation when exposed to alternate light and darkness. Isolate AS_2 and AS_3 produced maximum growth and sporulation than isolates AS_1, AS_4 and AS_5 at all light treatments tested.

Pathogenic variations among the five isolates of A.solani showed that isolate AS_3 and AS_2 produced more number of leaf spots and bigger lesions than the other isolates. Thus, these two isolates appeared to be more

aggressive in causing the disease than the isolate AS_1, AS_4 and As_5. Isolate AS_4 found to be weakly pathogenic on tomato causing only a few small lesions.

The fungus survived in infected tomato seeds upto 160 days, while the survivability in host debris was upto 180 days. Further, the survivability of fungus was maximum in sterilized soil (200 days) than unsterilized soil (160 days) at 30% moisture holding capacity.

A total of twenty five host plants of cereals, legumes, oilseeds, pulses, vegetables and a few weeds belonging to different families were tested to know the host range of A.solani. Only hosts belonging to family solanaceae viz. Solanum tuberosum, Capsicum annum and Lycopersicon esculentum produced mild to moderate symptoms when the fungus was inoculated to uninjured hosts and severe symptoms in case of injured hosts. The other non-solanaceous hosts developed mild to moderate symptoms only when the pathogen was inoculated to injured hosts.

Out of twenty one popular varieties of tomato screened under glass house conditions against A.solani by artificial inoculation, none of them showed immune reaction. While the varieties viz., Sonali, Vishali,

IAHS 881, IAHS 882, Rajani and all Arka varieties showed resistant reaction, the varieties Roma, Punjab Kesari, Rasmi and Naveen showed moderately resistant reaction. Pusa ruby gave a highly susceptible reaction.

Among the seven fungicides evaluated *in vitro* against *A.solani* Iprodione and Mancozeb gave total inhibition of the fungal growth at all concentrations.

The field evaluation of different chemicals viz., Iprodione (0.1, 0.2, 0.3 and 0.4%), mancozeb (0.2%), captafol (0.2%) and carbendazim (0.1%) during 1991 and 1992, indicated that Iprodione (0.2%) and mancozeb (0.2%) were most effective in reducing severity of the disease on the foliage, fruit stalk and on fruit and increasing the fruit yield during 1991 and 1992. However, maximum cost benefit ratio was obtained with Mancozeb (0.2%) and Iprodione (0.2%) sprays, when used in controlling early blight and Alternaria fruit rot of tomato.

Production of melanin in *A.solani* increased as age of the culture advanced. Melanin produced by *A.solani* reduced the efficacy of Mancozeb when tested against a hyaline test fungus *Fusarium oxysporum* f.sp. *cubens*.

REFERENCES

VII. REFERENCES

Abol-Wafa,M.T. and Kamara,A.M. 1975, Control of carly blight and leaf spot of tomato by various fungicides. Egyptain J.Phytopathol., 7:107-110.

Achuta,M. Pandu,S. and Satyanarayana, Y., 1988, Fungicidal control of Alternaria leaf spot of sesame. Indian J.Agri. Sci., 58:648-649.

*Andrus,C.F., Reynard,G.B. and Wade,B.L., 1945, Relative resistance of tomato varieties selections and crosses to defoliation by Alternaria solani, U.S.D.A. Circ., 652, pp.23.

Anon., 1991, The State of Food and Agriculture. Food and Agriculture Organisation of the United Nations, Rome, pp.171.

Aponyi,I., Kujani,O., Balong,B. and Varga,A., 1988, Ovary moulding disease of tomato-shaped pepper and control possibilities. Acta Horticulturae, 220:401-406.

Atherton,T.G. and Rudich,J., 1986, The tomato crop, Chapman and Hall Ltd., U.S.A., pp.512.

Balanchard,D., 1992, A colour atlas of tomato diseases. Wolfe Pub. Ltd., Brook house, London, pp.298.

Barris,B.C. and Mariotto,P.R., 1986, Action of fungicides on the development of Helminthosporium sativum Pamm. King and Bakke in vitro. Biologico., 52:65-72.

Barksdale,T.H., 1968, Resistance of tomato seedling to early bright. Phytopathology, 59:443-446.

Barnett,H.L. and Lilly,V.G., 1950, Nutritional environmental factors influencing asexual sporulation of Choanephora cucurbitaram in culture. Phytopathology, **40**:80-89.

*Bartnicki-Garcia,S. and Rayes,E., 1964, Chemistry of spore wall differentiation in Mucor rocini. Arch.Biochem.Biophys., **108**:125-133.

*Bashi,E., 1979, Spraying against Alternaria blight failed to increase yield of potatoes and tomatoes in trials in the Besor region. Hassadeh, **60**:261-265.

Basu,P.K., 1971, Existence of chlamydospores of Alternaria porri f.sp. solani as over wintering propagules in soil. Phytopathology, **61**:1347-1350.

Basu,P.K., 1974a, Measuring early blight, its progress and influence on fruit losses in nine tomato cultivars. Can.Pl.Dis.Sur., **54**:45-51.

Basu,P.K., 1974b, Reduction of primary infection of tomato early blight by fall fumigation of soil with verlex. Can.pl.Dis.Sur., **54**:24-25.

*Bedlan,G., 1987, Control of leaf spot in Chinese cabbage Bekampfung der Blattfceckon Krankheit on Chinakohl. Pfcanzenschutz, **3**:4-6.

Bell,A.A. and Wheeler,M.H., 1986, Biosynthesis and functions of fungal melanins. Ann.Rev. Phytopathol., **24**:411-51.

Bloom Field,B.J. and Alexander,M., 1967, Melanins and resistance of fungi to lysis. J.Bacteriol., **93**:1276-1280.

Bonde,R., 1929, Physiologic strains of Alternaria solani. Phytopath., **19**:533-548.

Bose,T.K. and Som,M.G., 1986, Vegetable Crops in India. Nayaprakash Publishing, Culcutta, pp.773.

Brock,R.D., 1950, A search for resistance to defoliation by A.solani in the genus Lycopersicon. J.Australian Inst.Agri.Sci., **16**:90-94.

*Carrasco,D.H., Valazquez Diaz,O., Garcialopez,J.L. and Gonzalez Ramos,L.A., 1977, Evaluation of experimental cultivars of tomato in Forme Cientifico teenico instituto de investigaciones Fundamentales on Agricultura Tropical, **39**:31-33.

Chamber,S.C. and Elentje,N.T., 1969, Relative effect of soil nitrogen and soil organism on survival of Ophiobolus graminis. Austr.J.biol.Sci., **22**: 275-278.

Charton,K.M., 1953, The sporulation of Alternaria solani in culture. Trans.Br.Mycol.Soc., **36**:349-365.

Chet,I., Henis,Y. and Mitchell,R., 1967, Chemical composition of hyphal and Sclerotial walls of Sclerotium rolfsii. Can.J.Microbiol., **13**:137-141.

Choulwar,A.B., Datar,V.V. and Shinde, 1990, Reaction of lines, varieties and hybrids to early blight in tomato. J.Maharastra Agri.Univ., **15**:94-95.

Chowdhury,S., 1944, An alternaria disease of sunflower. J.Indian bot.Soc., **23**:59-65.

Chu,S.B. and Alexander,M., 1976, Resistance and susceptibility of fungal spores to lysis. Trans.Brit.Mycol.Soc., **58**:489-497.

CoChrane,V.W., 1958, Physiology of fungi, John Wiley and Sons, Inc., London, pp.528.

Combe,M., 1960, Some effects of water and anerobic conditions on F.oxysporum f.sp. cubense in soil. Trans.Brit.Mycol.Soc., **43**:51-59.

Dahiya,T.S., 1989, Melanin biosynthesis in Alternaria brassicae. Indian J.Microbiol., **28**:42-49.

Datar,V.V. and Mayee,C.D., 1981, Assessment of loss in tomato yield due to early blight. Indian Phytopath., **34**:191-195.

Datar,V.V. and Mayee,C.D., 1985, Chemical management of early blight of tomato. J.Maharastra Agri.Univ., **10**:278-280.

De,B.K. and Chattopadya,S.B., 1984, Evaluation of fungicides against early blight disease of potato caused by Alternaria solani. Pesticides, 18:52-53.

Desai,S.A., 1979, Studies on Alternaria leaf and twig blight of cotton in Karnataka. M.Sc(Agri) Thesis, University of Agricultural Sciences, Bangalore, pp.93.

Dhiman,J.S., Bedi,P.S. and Bombawale,O.M., 1980, An easy method of preparing inoculum of A.solani for mass inoculation. Indian Phytopath., 33:359-362.

*Dorozhkin,N.A. and Ivanyuk,V.G., 1979, Epiphytotics of dry leaf spot of tomato and potato. Mikiologiya Fitopatologiya, 13:314-321.

*Drayton,F.L., 1937, The effect stage of Botrytis convoluta. Mycologia, 29:305-318.

Ellis,M.B., 1971, Dematiaceous hyphomycetes. Commonwealth Mycological Institute, Kew, Surry, England, pp.608.

Ellis,O.H. and Griffith,D.H., 1974, The location and analysis of melanin in the cell wall of some soil fungi. Can.J.Microbial., 20:1379-1386.

*Falck,R., 1907, Wachstumgesetze, Wachstufakoren und Temperaturwerte der holzer storenden. Mycelien, 1:153-154.

*Fencelli,M.I. and Kimati,H., 1990, Influence of culture media and fluorescent light on the sporulation of Alternaria dauci. Phytopathologica, 16:248-252.

Fryoyd,J.B., Paget,C.J., Gues,L.R., Dreikom,B.A. and Pafford,J.L., 1978, Tricyclazole:A new systemic fungicide for control of Pyricularia oryzae on rice. Phytopathology, 66:1135-39.

Gurwat,P.D. and Ghosh,S.K., 1979, Studies on the physiology of growth of Alternaria solani. Indian J.Mycol.Pl.Path., 9:138-139.

Groves,J.W. and Sholka,A.J., 1944, Notes on seed born fungi 11. Alternaria. Can.J.Res.Sect., 22:217-34.

Hanumanthaiah,B., 1976, Studies on leaf spot disease of redgram [Cajanus cajan (L.) Mill Sp.] caused by Alternaria tenuissima in Karnataka. Unpublished thesis. M.Sc(Agri), University of Agricultural Sciences, Bangalore.

Hedges,T.R. and Cole,D.L., 1988, Field resistance to Iprodione in Alternaria alternata on apples in Zimbawe. ISPPI Chemical Control Newsletter No.11:38-39.

Henning,R.G. and Alexander,W.J., 1959, Evidence of existence of physiologic races of A.solani. Pl.dis.Reptr., 47:643.

Holm,R., Pancho,V., Herberger,T.P. and Plucknett,D.L., 1981, A geographical atlas of world weeds, John Wiley and Sons, New York, pp.380.

Hemperson-Jones,F.M. and Maude,R.B., 1982, Control of the dark leaf spot (Alternaria brassicola) of Brassica oleraceae in seed production crops with foliar sprays of Iprodione. Ann.Appl.Biol., 100:99-104.

Inoue,S., Uematsa,T. and Kato,T., 1983, Mode of rice blast control by chlorbenthiazone. J.Pestic.Sci., 8:333-338.

Ismail,M., 1987, Evaluation of some potato varieties and clones for field resistance to early blight. Pakistan J.Agri.Res., 8:356-359.

Johnson,S.S., 1969, Fungicides for tomatoes. Queensland agric.J., 95:369-371.

Joshi,P.V., 1981, Studies on Alternaria gomphrenae causing leaf and stem blight of Gomphrena globosa Linn. Unpublished M.Sc(Agri) Thesis, University of Agricultural Sciences, Bangalore, pp.103.

Kaul,A.K. and Saxena,H.K., 1988, Physiologic specialisation in Alternaria solani causing early blight of tomato. Indian J.Mycol.Pl. Path., 18:128-132.

Kodmelwar,R.V., Wangikar,P.D. and Shukla,V.N., 1973, In vitro studies of some copper-based fungicides against Colletotrichum capsici and Alternaria solani. Punjab Krishi Vidyapeeth Research Journal, 1:212-213.

Kuo,M.T. and Alexander,M., 1967, Inhibition of the lysis of fungi by melanins. J.Bacteriol., 94:624-629.

Kvasnyuk,N., 1986, Effect of ecological factors on the over wintering of Alternaria solani. Phytopathology, 20:60-63.

Leonian,L.H., 1924, A study of factors promoting pycnidium formation in some Sphaeropsdales. Am.J.Bot., 11:19-50.

Lilly,V.G. and Barnett,H.L., 1951, Physiology of the fungi. McGraw Hill Book Company Inc., New York, pp.439.

Linderman,R.G. and Tousoun,T.A., 1966, Behaviour of albino chlamydospores of Thielaviopsis basicola. Phytopathology, 56:887 (Abstr.).

*Lingappa,Y., Sussman,A.S. and Bernstein,I.A., 1963, Effect of light and media upon growth and melanin formation in Aureobasidium pullulans. Am.Mycopathol.Mycol.Appl., 20:109-128.

Locke,S.B., 1949, Resistance to early blight and septoria leaf spot in the genus Lycopersicon. Phytopathology, 89:829-836.

Lockwood,J.L., 1960, Lysis of mycelium of plant pathogenic fungi by natural soil. Phytopathology, 50:787-789.

*Lodha,P.C. and Prasad,N., 1975, Efficacy of some fungicides and antibiotics against alternariosis of tomato. Phytopathologia maditerraneus, 14:21-22.

Ludwig,R.A., Richardson,L.A. and Unwin,O.H., 1962, A method for inducing sporulation of Alternaria solani in culture. Can.Pl.dis.Surv., 42:149-150.

Lukens,R.J. 1963, Photo-inhibition of sporulation in Alternaria solani. Am.J.Bot., 50:720-724.

*Mobbett,H.T.H. and Phelps,R.H. 1985, The behaviour of copper deposits from low and ultralow volume spray on tomato foliage and their control of early blight. Thai.J.agri.Sci., 18:81-100.

Madalageri,B.B., Dharmatti,P.P., Srikant Kulkarni and Madalgeri,M., 1988, Screening of tomato genotypes for combined resistance to Stemphylium solani, Alternaria solani and Septoria lycopersici. Plant Pathology Newsletter, 6:18.

*Maeso,D.C., 1986 Chemical control of early blight in tomatoes. Investigaciones Agronomicas, Centre de Investigaciones Agricolas. Alberto Boerger raguay, 7:31-45.

Mathur,K. and Shekhawat,K.S., 1986, Chemical control of early blight in kharif sown tomato. Indian J.Mycol.Pl.Path., 16:235-238.

McCarter,S.M., Jaworski,C.A. and Johnson,A.W., 1976, Soil fumigation effects on early blight of tomato transplants. Phytopathology, 66:1122-24.

Mahabaleswarappa,K.B., 1981, Studies on leaf spot of safflower (Carthamus tinctorius Linn.) caused by Alternaria carthami Choudhury. Unpublished M.Sc(Agri) thesis, University of Agricultural Sciences, Bangalore.

Mayee,C.D. and Datar,V.V., 1986, phytopathometry Technical bulletin-1. Marathwad Agricultural University, Parabhani, pp.[illegible].

Moore,W.D. and Thomas,H.R., 1942, Some cultural practices that influence the development of A.solani on tomato seedlings. Phytopathology, 33:117[illegible]-1184.

Mora,F.R., 1978, Prevention of leaf disease of tomato with fungicides. Agricultura toenicaen Mexico, 4:187-192.

Nagaraj,T.R. and Ponnappa,K.M., 1970, Blight of water hyacinth caused by Alternaria eichhorniae sp. nov. Trans.Br.Mycol.Soc., 55:123-130.

Neergaard,P., 1945, Danish species of Alternaria and Stemphylium Hamphry Millford, Oxford Univ.Press, London, pp.566.

Neto,F.B. and Oliveira,D.A., 1980, Effect of different fungicides on the growth of Alternaria solani "In vitro". Biologico., 46:101-106.

Padhi,M.N. and Rath,G.C., 1973, Sporulation of Alternaria solani in pure culture. Indian Phytopath., 26:495-501.

Patterson,C.L., 1991, Importance of clamodospores as primary inoculum for Alternaria solani, incitant of collar and early blight on tomato. Plant dis., 75:274-278.

Pigman,L., 1957, The carbohydrates, Academic Press Inc., N.Y., pp.512.

Portgieter,H.T. and Alexander,M., 1966, Susceptibility and resistance of several fungi to microbial lysis. J.Bacterial., 91:1526-1532.

Prasad,B. and Dutt,B.L., 1971, Laboratory assessment of the reaction of potato varieties to A.solani. Indian J.Microbiol., 11:91-96.

Quinn,J.G. and Johnson,D.R., 1979, Further evaluation of very low volume captafol spray to control tomato diseases in northern Nigeria. PANS, 25:257-264.

Rajagopal,R. and Vidyasekaran,P., 1983, Effect of fungicidal control of leaf spot disease of tomato on the quality of fruits. Indian Phytopath., 36:352-354.

Ramakrishnan,T.S., 1971, Diseases of rice. I.C.A.R., New Delhi, pp.24-40.

Ramakrishnan,L. and Kandaswamy,J.K., 1978, Efficacy of fungicides on the control of Alternaria leaf spot of tomato. Madras Agri.J., 65:122-123.

Ramakrishnan,L., Kamalanathan and Krishnamurthy,C.S., 1971, Studies on Alternaria leaf spot of tomato. Madras Agri.J., 58:275-280.

Rands,R.D., 1917a, Early blight of tomato and related plants. Wis.Agri.Exp.Stn.Bull., 42:1-48.

Rands,R.D., 1917b, The production of spores of Alternaria solani in pure culture. Phytopathology, 7:316-317.

Rangaswamy,G. and Vidyasekaran,P., 1963, Antibiotic production by Streptomyces spp. in corn rhizosphere. Phytopathology, 53:995-997.

Reda,F., Mousa,O.M., Sejiny,M.J. and Nawar,L.S., 1985, Response of tomato cultivars to infection with A.solani in relation to growth and chemical composition of their fruits. Egyptian J.Phytopathol., 17:83-93.

Reda,F., Mousa,O.M., Sejny,M.J. and Nowar,L.S., 1986, Effect of cystein on the resistance of tomato plants to early blight. Egyptian J.Phytopathol., 18:89-100.

Robert,D.A. and Boothroyd,C.W., 1972, Fundamentals of Plant Pathology, W.H.Freeman and Company, pp.248.

Rotem,J., 1966, Variability in Alternaria Porri f.sp. solani. Israel J.Bot., 15:48-57.

*Schwinn,F., Nakamara,M., Handschin,G., 1979, CGA 49104, a new systemic fungicide against rice blast. Phytopath.Soc.Abstr., 470.

Sengupta,P.C. and De,B.K., 1987, Studies on spray schedule of fungicides against early blight disease of potato caused by Alternaria solani. Indian Agri., 31:207-211.

Shahin,E.A. and Shepard,J.F., 1978, An efficient technique for inducing profuse sporulation of Alternaria spp. Phytopathology, 69:618-20.

Sharif,F.M., Al-Fodhal,F.A., Abdul-Al-Latif,N., Kasha,A.M. and Kadhum,K.J., 1988, Development of infection with leaf mold and early blight diseases on some varieties and hybrids of tomato. Journal of agriculture and water Resources Research, Plant Production, 7:85-97.

Singh,R.S., 1987, Diseases of Vegetable Crops. Oxford and IBH Pub.Co.Pvt.Ltd., New Delhi, Bombay, Culcutta, 419pp.

Sridhara,T.S. and Naik,L.B., 1983, Relative resistance of tomato cultivars to early blight. Madras Agri.J., 70:488-489.

Stevenson,W.R., 1977, Use of captafol and chlorothalonil on reduced application method schedules for tomato disease control in Indiana. Pl.Dis. Reptr., 61:803-805.

Stevenson,R.E. and Packer,S.P., 1988, Effect of radiation temperature and moisture on conidial germination of Alternaria solani. Phytopathology, 78:926-930.

Timmick,M.B., Lilly,V.G. and Barnett,H.L., 1951, The influence of light and other factors upon sporulation of Diaporthe phaeseoloram var. botatis from soybean. Phytopathology, 41:327-336.

Thind,T.S. and Jyooty,J.S., 1982, Persistence of fungitoxicity of Dithane M-45 on some tomato varieties. Indian Phytopath., 35:657-660.

Tuite,J., 1969, Plant pathological methods, Fungi and Bacteria, Burgess Publishing Company, U.S.A., pp.239.

*Vakalounakis,D.T., 1982, An efficient and simple technique for inducing profuse sporulation in Alternaria solani. Phytopathologia mediterranea, 21:89-90.

Vakalounakis,D.T., 1983, Evaluation of tomato cultivars for resistance to Alternaria blight. Annals of Applied Biology, 102:138-139.

Verma,V., 1970, Effect of temperature and hydrogen-ion concentration of three pathogenic fungi. Sydowia, 23:164-168.

Vidyasekaran,P., 1971, Saprophytic survival of Helminthosporium nodulosum and H.tetramera in soil. Indian Phytopath., 24:347-353.

Vidyasekaran,P. and Rangaswamy,G., 1969, Presence of an antibiotic in the root washing of maize plants. Indian Phytopath., 22:57-60.

Vincent,J.M., 1927, Distortion of fungal hyphae in the presence of certain inhibition. Nature, 159:850.

Walker,J.C., 1952, Diseases of vegetable crops. McGraw Hill Book Company Inc., New York, pp.529.

Wheeler,B.E.J., 1969, An introduction to plant disease. John Wiley and Sons Ltd., London, pp.301.

Yawalkar,K.S., 1969, Vegetable crops of India. Agri-Horticultural Publishing House, Nagpur-1, India, pp.300.

*Zhu,Z.Y., Huang,Y.M. and Li,Y.H., 1985, An efficient technique for inducing profuse sporulation of Alternaria solani in pure culture. Acta Mycologica Sinica, 4:180-184.

*Zink,P. and Fengal,D., 1989, Studies on the colouring matter of blue-stain fungi part-2-electron microscopic observations of the hyphae walls. Holzforschung, 43:371-75.

*Original not seen.

APPENDIX

APPENDIX I

Temperature, relative humidity sunshine hours and rainfall prevailed during study period from 4-8-1991 to 19-7-1992 at G K.V.K. Bangalore

Month	Temperature (°C) Max	Min	Mean	Relative Humidity (%)	Sunshine hours	Rain-fall (mm)
1991						
August	26.17	18.03	21.11	75.58	4.22	26.81
September	28.70	18.97	23.85	70.00	7.97	16.72
October	27.17	18.30	22.75	74.50	6.37	58.52
November	25.07	17 50	21.30	80.25	5 02	116.50
December	26 55	13.10	19.82	68.16	7.53	0.00
1992						
January	25 87	12.32	19.10	61.00	8 72	1.50
February	28.85	15.70	22.27	63.50	9.65	0.00
March	33.42	16.17	24.80	36 25	10.30	0.00
April	33.95	19.67	26.82	49.75	9.22	2.50
May	32.45	20.80	27.03	63.00	8.27	21.67
June	28.61	19.45	24.03	75.40	6 20	56.92
July	26 87	18.77	22 82	77.00	4.22	33.30