INTRODUCTION

*Capsicum annuum* L. (Chilli), an annual sub-shrub constitutes one of the most important spices cultivated all over the world except in colder parts. Chillies are cultivated mainly in tropical and sub-tropical countries like India, Japan, Mexico, Turkey, United states of America and African countries. India is the largest producer of chillies in the world, accounting for over 45% of the total area under cultivation. India is well known as the land of spices the world over. Chillies are grown practically all over India. In India, they occupied about ninety thousand hectares with a production of ninety thousand tones of chillies. Andrapradesh, Maharashtra, Karnataka, Orissa and Tamilnadu alone account for about 75% of the total ares as well as production. Almost all the varieties of low and medium pungency that are cultivated on field scale in India belong to *Capsicum annuum*. Export of chillies during 1999-2000 was 64776 metric tones valued at Rupees 250.66 crores. However, chilli is the second commodity in our export basket earning nearly 13% foreign exchange from spices (Balasubramaniam, 2001).

In 2004-05, India accounted for about 45 per cent of the global spice exports. It exported 335,488 tonnes of spices and spice products valued at Rs.2200 crore. Indian spices are exported to more than 150 countries and the major markets are the USA, UK, Japan, UAE, Sri Lanka and Germany. It was a 35 per cent increase in volume when compared to the export of 254,382 tonnes valued at Rs.1911 crore in 2003-04.
Cultivation

Capsicum pepper refers primarily to *Capsicum annuum* L. and *Capsicum frutescens* L., plants used in the manufacture of selected commercial products known for their pungency and color. *Capsicum annuum* L. is a herbaceous annual that reaches a height of one meter and has glabrous or pubescent lanceolate leaves, white flowers, and fruit that vary in length, color, and pungency depending upon the cultivar. Native to America, this plant is cultivated almost exclusively in Europe and the United States.

The reported life zone for capsicum peppers is 7 to 29 degrees centigrade with an annual precipitation of 0.3 to 4.6 meters and a soil pH of 4.3 to 8.7. *Capsicum* species are cold sensitive and generally grow best in well-drained, sandy or silt-loam soil. Plantings are established by seeding or transplanting. Flowering usually occurs three months after planting. Hot and dry weather is desirable for fruit ripening. Fruit is generally handpicked as it ripens, and then allowed to dry in the sun, although artificial drying is often employed in Europe and the United States. The fruit may be ground intact or after the removal of seeds, placenta parts, and stalks, increasing the fruit color and lowering the pungency.

Chemical Constituents

The level of pungency of the *Capsicum* species depends upon the concentration of capsaicinoids, primarily of capsaicin, in the fruit. Capsicum peppers are classified commercially by the concentration of capsaicinoids, since confusion about the biological identities of some varieties has made other methods unreliable. Paprika comes from plants with 10 to 30 parts per million
capsaicinoids, chilli peppers from plants with 30 to 600 parts per million, and red peppers from plants with 600 to 13,000 parts per million (1.5-152). The chemical composition of the *Capsicum* species includes a fixed oil, pungent principles, volatile oil, and carotenoid, mostly capsanthin, pigments (6.1-65, 2.8-45). An oleoresin is obtained by solvent extraction. *Capsicum frutescens* L. is much more pungent than *Capsicum annuum* L.

**Economic Uses**

*Capsicum* species are used fresh or dried, whole or ground, and alone or in combination with other flavoring agents. *Capsicum annuum* L. is used in sweet bell peppers, paprika, pimento, and other red pepper products. *Capsicum frutescens* L. is used in tabasco, tabasco sauce, and other red chili pepper. Fruits of *Capsicum annuum* L., paprika types, are widely used as coloring agents. The extracts of *Capsicum* species have been reported to have antioxidant properties. Paprika is derived from *Capsicum annuum* L. and is used primarily in the flavoring of garnishes, pickles, meats, barbecue sauces, ketchup, cheese, snack food, dips, chilli cake, salads, and sausages. Spanish paprika is called pimento and is generally used for coloring purposes. Chilies and chili pepper from cultivars of *Capsicum annuum* and *Capsicum frutescens* L. are employed as a flavoring in many foods, such as curry powder and sauces. Chili powder is a blend of spices that includes ground chilies. Red or hot peppers from *Capsicum annuum* L. and *Capsicum frutescens* L. are the most pungent peppers and are used extensively in Mexican and Italian foods. Cayenne pepper is the ground product derived from the smaller, most pungent *Capsicum* species.
As a medicinal plant, the *Capsicum* species has been used as a carminative, digestive irritant, stomachic, stimulant, rubefacient and tonic. The plants have also been used as folk remedies for dropsy, colic, diarrhoea, asthma, arthritis, muscle cramps, and toothache. *Capsicum frutescens* L. has been reported to have hypoglycemic properties. Prolonged contact with the skin may cause dermatitis and blisters, while excessive consumption can cause gastroenteritis and kidney damage. Paprika and cayenne pepper may be cytotoxic to mammalian cells *in vitro*. Consumption of red pepper may aggravate symptoms of duodenal ulcers. High levels of ground hot pepper have induced stomach ulcers and cirrhosis of the liver in laboratory animals. Body temperature, flow of saliva, and gastric juices may be stimulated by capsicum peppers.

**Antioxidants content**

The plant can be used to relieve uterine pain associated with childbirth (Sumner, 2000). The leaves are used to treat toothache (Wee and Hsuan, 1990). The fruits are used to stimulate the digestive and circulatory systems. It is also carminative, used locally for neuralgia and for rheumatism.

Chillies have an inseparable presence in the spice kaleidoscope. Of all the Indian spices available in the market, chillies are the cheaper ones. Dry chilly is extensively used as spice in curried dishes. It is also used as an ingredient in curry powder and in seasonings. Chilli is used in making hot sauces as pepper sauce and Tabsco sauce, widely used for colour extraction. This colour is highly popular among food and beverage processors for its use as a colourant, since this being a ‘natural plant colour’.
As a medicine it is used as a counter irritant in Lumbago, Neuralgia, and Rheumatic disorders. Capsicum has a tonic and carminative action. Taken inordinately it may cause gastro-enteritis. The enzyme isolated from chilly is used in the treatment of certain type of cancers. Oleoresin capsicum is used in pain balms and vaporubs. Dehydrated green chilly is a good source of vitamin ‘c’, capsaicin, vanillin, solanine and chilli oleoresin. They are consumed in green, red as well as in sun dried condition. Among the spices consumed per head in India, dried chillies contribute a major share.

According to the latest findings of the Adayar Cancer Research Institute, Chennai, India, the common green chillies have been found to be the most fertile source for the enzyme ‘L-asparaginase’ which has an anti-tumor element and is used in the treatment of ‘Acute Lymphocytic Leukaemia’ a type of cancer (Pruthi, 1998).

Zibokere (1994) proved that red pepper powder acts as an effective insecticide against beetle population of cow pea than the whole chilli fruit. The fruits may contain some volatile substances and may act as insect repellent.

**Symptoms of Anthracnose of Capsicum annuum**

Fifty one different pathogens have been reported to cause diseases on various parts of chilli (Mukerji and Jayanthi Bhasin, 1996; Saha and Singh, 1988). Out of them, thirty nine belong to the fungi of classes Mastigomycotina, Ascomycotina and Deuteromycotina. Among the three major fungal diseases, anthracnose affects the yield directly by infecting fruits and indirectly by infecting stems and leaves and causing flower drop.
Colletotrichum capsici is capable of causing disease on virtually all parts of the Chilli plant during any stage of plant growth. However, fruit lesions is the most economically important aspect of this disease. The disease in chilli is observed in three phases:

(i) Seedling or damping off; prevalent in nursery.
(ii) Leaf spot and die-back which are initiated at different stages of growth.
(iii) Ripe rot.

Die-back

Die-back symptoms become very much prominent when the crop is near flowering stage. Die-back phase is characterized by the necrosis of tender twigs from the tip downwards. The entire plant or branch may wither away. The twigs become straw colored in the advanced stage of the disease. Large numbers of black dots (acervuli of the fungus) are seen scattered all over the necrotic parts of the plants. Only the top or side branches may be killed, sometimes in case of severe attack the entire plant may be withered.

Ripe –rot

During the ripe rot phase, which is the most devastating one, small black circular spots appear on the skin of the fruit and spread in the direction of long axis of the fruit. The spots are sunken with black margins. The sunken spots are covered with pinkish mass of fungal spores. With the advance of the disease the spots spread forming a concentric markings with dark fructification (acervuli of the fungus). The fruit with many spots drop off prematurely resulting in heavy loss in the yield. In the advanced stage, the seeds are covered with a mat of fungal hyphae.
Such seeds turn rusty in color. The fungi are externally seed borne. The disease spreads rapidly with rainy season. The fungus survives in the field, in the plant debris and through seeds collected from diseased fruits.

*Colletotrichum*

*Colletotrichum* was described for the first time by Corda in 1831. In 1849, a closely related genus *Gloeosporium* was described by Desmazieres and Montagne; it was distinguished from *Colletotrichum* merely on the basis of the absence of setae in the acervulus. The presence or absence of setae to distinguish these two genera was rejected since the formation of these structure depends upon external conditions and can be controlled by varying the relative humidity alone (Frost, 1964). A revision of *Colletotrichum* was made by Von Arx (1970, 1975) who included species of *Gloeosporium* in his study and proposed a key for the identification of 23 Spp. of *Colletotrichum*. So all the species which were formerly included in the genus *Gloeosporium* have been included in the genus *Colletotrichum* (Sutton, 1973).

The genus *Colletotrichum* has a very wide range of behavioral patterns in nature ranging from saprophytes to specialized parasitic strains. Over 1000 form-species of *Colletotrichum* have been described on the basis of the diseases caused on various hosts. *C. capsici* is a widespread fungus and has been mostly studied as a pathogen on *Capsicum annum*. It is a general unspecialized parasite and has been recorded from more than 100 plants in a wide range of families of Angiosperms.
Some of the economically important plants infected by *C. capsici* are:

1. *Abelmoschus esculentus* (Linn.) Moesch,
2. *Adathoda vasica* Nees.
3. *Amaranthus caudatus* Linn.
4. *Arachis hypogaea* Linn.
5. *Basella alba* Linn.
6. *Beta vulgaris* Linn.
7. *Cajanus cajan* (Linn.) Millsp.
8. *Capsicum annum* Linn.
10. *C. reoseus* Linn.
11. *Cicer arietinum* Linn.
14. *Mangifera indica* Linn
15. *Piper betle*. Linn
16. *P. nigrum*. Linn
17. *Rosa indica* Linn.
18. *Saraca indica* Linn.
22. *S. xanthocarpum* Schrad & Wendl.
Die-back and ripe fruit rot of chillies caused by this fungus is a very serious disease in India. The ripe fruits, turning red, show elliptical lesion which are greenish black or dirty grey in colour showing numerous dot-like black structure, the acervuli of the fungus. The fungus spreads to the central cavity of the fruit and infects the seeds. In Die-back as the name indicates, the disease causes necrosis of tender twigs from the tip backwards. The entire branch or the entire top of the plant may wither away.

Many plant pathologists had recorded this disease throughout the world. In China, Lee Ling and Lin (1994) reported the occurrence of *C. capsici* on chilli plant. Thompson (1928) reported that *C. capsici* was able to attack egg plant and tomato in Malaysia. Chandler (1958) reported that *C. capsici* was an incident fungus on Pimiento pepper in George, U.S.A. *C. capsici* produces dense, whitish to dark grey aerial mycelium, reverse of the colony is dark brown. Conidial masses are pale buff to salmon-coloured but individual conidia are hyaline, falcate, fusiform; apices acute, 18-24 X 3-5µ; setae numerous, trichiform, brown to blackish brown, 2-4 septate, 50-100 X 2-6µ. No sclerotia are produced. Appressoria are abundantly produced by the germinating conidia. In India, the disease was first reported by Sydow (1913) on the chilli samples from Madras. It was later reported by Dastur (1921) from Bihar and he named the causal organism as *Vermicularia capsici* Syd., (Synonym of *C. capsici*) and described the symptomatology and cultural characteristics of the parasite.

Chowdhury (1957) reported that 12 to 32% destruction of fruits was due to *C. capsici* in Assam. Jindal *et al.* (1994) reported seed borne nature of *C. capsici* and its transmission in Bel-pepper. Datar (1995) conducted a survey to evaluate the
damage of chilli fruit in the market. He reported that two species of *Aspergillus*, two species of *Fusarium*, *Drechslera australiensis* and *C. capsici* are responsible for post harvest damage. Among them *C. capsici* caused serious damage.

Siddiquie *et al.* (1977) reported seed borne nature of *C. capsici* in Karnal district. Thiram and difolatan were found to inhibit seed borne disease of *C. capsici* and sprays of the same in the field reduced incidence of *C. capsici* and increased the yield of chilli in the field.

Kannan *et al.* (1998) reported that ripe rot caused by *C. capsici* was more destructive as it damaged the chilli fruit partially or completely in the field as well as during transit and storage. Die-back caused by *C. capsici* led to drying up of the twigs and flower drop, thereby reducing the yield. They estimated the yield loss due to *C. capsici* infection was 25%.

Anthracnose is a serious disease in chilli and in suitable weather, it may cause 12-35% damage to the crop. Although the disease is found throughout India, it is more severe in the southern states. *Colletotrichum capsici* belongs to the order Melanconiales, containing a single family Melanconiaeeae, It was reported for the first time in India by Butler and Bisby. This disease causes severe damage to fruits in the fields as well as in storage and takes heavy toll upto 84%. Badly infected fruits may lose their normal red colour and turn straw coloured or in some cases, pale white. The pathogen also causes necrosis of tender twigs and the entire branch. It is characterized by the production of acervuli, which may develop subepidermally. An acervulus is a saucer-shaped fructification in which the hymenium of conidiogenous cells develop on the floor of the cavity from a
pseudoparenchymatous stroma beneath an integument of host tissue which ruptures at maturity.

The acervuli produced by *Colletotrichum* may be subcuticular, epidermal or subepidermal and may be either separate or confluent. The conidiophores are hyaline to brown, septate, branched at the base, smooth, formed from the upper cells of the fructification. The conidia are hyaline, unicellular, falcate or lunate (sickle-shaped) or cylindrical, more or less guttulate, muticate or with the apex prolonged into simple cellular appendages, produced from phialides (conidiogenous cells) enteroblastically. The conidiogenous cells form a closely packed layer and often intermingled with setae. The setae are straight, unbranched and tapered towards the apex. These are brown, smooth, thick-walled and septate and originate from the pseudoparenchyma. On germination conidia become pale brown and septate and form appresoria. Sclerotia are sometimes produced in the culture which are dark brown to black, often confluent, occasionally setose. The perfect state of the fungus belongs to *Glomerella*.

As soon as dispersed conidia of *Colletotrichum* spp. rapidly adhere to aerial parts of plants, to initiate disease. The conidia are embedded in water-soluble mucilage composed of high molecular mass glycoproteins which is responsible for initial attachment of conidia to hydrophilic substrate (Hughes et al., 1999). An important feature in the pathogenesis of fruit rot is the chemical mechanism of penetration of pathogens into the host and colonization of host tissue. Many plant pathogenic fungi produce an array of extra cellular cell wall degrading enzymes that may be important in pathogenicity (Walton, 1994). It has been already reported that many species of *Colletotrichum* secrete pectinolytic and cellulolytic
enzyme systems which play a significant role in pathogenesis (Cervone et al., 1981; Chacko et al., 1978 and Manjeet Kaur and Deshpande, 1980). Goodman (1960) reported production of toxin, colletotin by C. fuscum. Narain and Das (1970) confirmed the production of colletotin by C. capsici which kills the tissues. The pathogen C. capsici was reported to inhibit photosynthesis, alter the physiology of the host plant and degrade capsaicin content of the fruits (Grewal and Grover, 1974).

Thind and Randhawa (1957) studied the nutritional requirement of C. capsici. They tried different combination of nitrogen sources and carbon sources suitable for growth and sporulation. They found that organic nitrogen compounds were better than inorganic ones for the sporulation. Misra and Mahmood (1960) studied the effect of carbon and nitrogen on the growth and sporulation of C. capsici. Lactose, xylose, sucrose and glucose were found to be best carbon sources and asparagine and peptone are the best nitrogen sources for growth and sporulation.

**Anthracnose (Colletotrichum spp.)**

**Causal Organism**

Anthracnose is caused by fungi belonging to the genus Colletotrichum, and they are responsible for diseases on numerous plant species worldwide. Identification of Colletotrichum species is usually based on more than one characteristic, such as physical appearance and pathogenicity on host(s). Many species of Colletotrichum infect more than one host and to confound identification, more than one Colletotrichum sp. may be present on one host. At least three
species of *Colletotrichum* (*C. gloeosporiodes, C. capsici, and C. coccodes*) are reported to cause this disease on pepper in Florida.

**Symptoms**

*Colletotrichum* is capable of causing disease on virtually all parts of the chilli plant during any stage of plant growth. However, fruit lesions are the most economically important aspect of this disease. Fruit symptoms initially begin as water-soaked lesions that become soft, slightly sunken, and become tan. The lesions can cover most of the fruit surface and multiple lesions occur (Figure 1). The surface of the lesion becomes covered with the wet, gelatinous spores from salmon-colored fungal fruiting bodies (acervuli) with numerous black spines (setae) (Figure 2). Concentric rings of the acervuli are common within the fruit spots. In some cases, the lesions are brown, not orange, and then black from the formation of setae and sclerotia (a dark, fungal survival structure) (Figure as follows).

![Mature Chilli fruit exhibiting multiple lesions of Anthracnose.](image-url)
**Disease Cycle and Epidemiology**

The fungus survives in and on seeds. Anthracnose can survive between seasons in plant debris or on weed hosts. Alternative hosts include weeds and other plants in Solanaceae (tomato, potato, eggplant) although infections of these hosts are extremely rare in Florida.

Fruits are infected when spores of the fungus or infested debris is rain splashed onto pepper plants. New spores are produced within the infected tissue and then are dispersed to other fruit. Workers may also move spores with equipment or during handling of infected plants.

Infection usually occurs during warm, wet weather. Temperatures around 80°F (27°C) are optimum temperatures for disease development, although infection occurs at both higher and lower temperatures. Severe losses occur during rainy weather because the spores are washed or splashed to other fruit resulting in more infections. The disease is more likely to develop on mature fruit that is present for a long period on the plant, although it can occur on both immature and mature fruit.

**Management Approaches**

Disease control methods include classical methods of synthetic pesticides and modern methods of biocontrol agents and elicitors. A wide spectrum of diseases is controlled by various modes of application of fungicides such as seed dressing, foliar spray and soil application. Many chemical fungicides such as Dithane M-45, Dithane Z-78, Blitox-50, Cumin L and Bavistin are being used to
control the various diseases caused by various pathogens including *Colletotrichum capsici* (Srivastava and Gupta, 1977).

**CONTROL**

**Cultural Controls:** Pathogen-free seeds should be used. Overhead irrigation whenever possible is to be avoided. Injury to fruit should be avoided. Crop rotation may be important in reducing primary inoculum.

Control of the disease is through integrated management techniques. Only seeds that are pathogen-free should be planted. Transplants should be kept clean by controlling weeds and solanaceous volunteers around the transplant houses. The field should have good drainage and be free from infected plant debris. If disease was previously present, crops should be rotated away from solanaceous plants for at least 2 years. Sanitation practices in the field include control of weeds and volunteer chilli plants.

**Plant extracts**

In view of the increasing toxic effects such as non-degradable and persistent residues caused by the continuous and constant use of synthetic chemical fungicides, researches were initiated on the biocontrol of various diseases. To sustain and to augment the crop yield, it is necessary to adopt strong crop protection strategies without altering the ecological balance of the cultivated land and environment.

Phytoextracts of several higher plants have been reported to control various fungal diseases (Pramila and Dubey, 2003; Srivastava and Bihari, 1997; Chitra,
2002 and Gomathi, 2001). Dubey (2003) reported that combination based on bio-agents *Gliocladium virens, Pongamia glabra* cake, chemical fungicide-carboxin and *Rhizobium* were effective in controlling *R. solani* causing web blight of urd (*Vigna mungo*) and mung bean (*V. radiata*). Synergistic inhibitory effect of phytoextract and antagonistic fungi on the growth of *R. solani* infecting *Glycine max* was brought out recently (Asha, 2006).

As an alternative to antibiotics, several plant extracts have been tested against *Erwinia amylovora* *in vitro* and *in vivo*. Mosch *et al.* (1989) reported an antibacterial activity for 24 out of 139 plant extracts tested in an agar diffusion test. The antibacterial activity against *Erwinia amylovora* could also be observed with leaf extracts from *Rhus typhina, Berberis vulgaris*, and *Mahonia aquifolium* in field experiments. Extracts from these plants, applied as protective spray showed a high disease control (Mende *et al.*, 1993). Moreover, a high activity against the disease was reported for plant extracts from *Reynoutria sachalinensis, Hedera helix, Viscum album* and *Alchemilla vulgaris*. These extracts induced resistance in the high susceptible host plant *Cotoneaster waterei*, causing a slower multiplication of the bacterium and a reduction in disease severity. The same results have been achieved with extracts from *Hedera helix* and *Viscum album* on detached leaves of *Cydonia ablonga* (Mosch *et al.*, 1993). In field experiments with the apple variety ‘James Grieve’, an extract from *Hedera helix* showed an efficacy similar to that of streptomycin (Mosch *et al.*, 1996). The active components of the plant extract from *Hedera helix* which induce resistance have not been elucidated. In another study, an inhibitory effect against the disease was also reported for Bactosan, an extract from *Pingania piñata*, by Psallidas and Tsiantos (2000). A plant extract from *Hedera helix*, an inducer of resistance, was
shown to cause physiological changes. Thus, enhanced levels of PR proteins (chitinase, β-1,3 glucanase) and enzymes of phenol metabolism, which can be regarded as a marker of resistance induction was observed (Mosch et al., 1996). These enzymes have been reported as markers for induced resistance by several authors (Hammerschmidt et al., 1982; Binder et al., 1989; Metraux et al., 1989; Ward et al., 1991).

**Alternatives to antibiotic compounds**

**Bacterial antagonists**

A biocontrol method based on antagonism against the disease has already been developed about 30 years ago (Beer and Rundle, 1987). Especially with strains of *Erwinia herbicola*, the so-called yellow bacteria, first positive results have been achieved. Later on, a significant reduction of fire blight was achieved by the application of *Bacillus subtilis*, *Pseudomonas fluorescens*, *Pantoea agglomerans* or *Rahnella aquatilis* strains (Beer and Norelli, 1986; Zeller and Wolf, 1996). In recent studies, application of *Pseudomonas fluorescens* caused 40-60% reduction of fire blight symptoms, and for *Pantoea agglomerans* a control of 50-80% was observed (Laux et al., 1999). Based on the strains, *Pseudomonas fluorescens* A506 and *Pantoea agglomerans* C9-1, the product Blight Ban 10 (Plant Health Systems) was released on the US-market. Another antagonistic strain of *Bacillus subtilis* is the active ingredient of the commercial formulation “Serenade” (Serenade Product Information). However, in most cases the field efficacy of these products was significantly lower than that of antibiotics, and not constant from year to year (Fried et al., 1998). Therefore, no registration of these products in Europe has been achieved until now.
Induced resistance

During the past 20 years, our improved understanding of the underlying processes leading to the preconditioning of plants against pathogens and parasites has been largely driven by the discovery of biological and chemical agents that are able to elicit the innate defenses of plants. Several biological and chemical elicitors of induced resistance are now commercially available for use in conventional agriculture. Vallad and Goodman (2004) reviewed the concept of induced plant resistance and the literature evaluating the effectiveness of these elicitors in controlling diseases of various crops under field conditions. They then examine the benefits and drawbacks of such technology and future considerations for the improved use of chemical and biological elicitors of induced resistance.

Induced resistance is a term that encompasses a number of plant responses in which the plant's resistance to a later challenge is increased by a previous challenge, i.e. the resistance to the pathogen, pest or pollutant is higher on second challenge than on first (Sticher et al. 1997).

Plants have several challenge-inducible resistance mechanisms, broadly divisible into local and systemic defences. Local defences include structural changes, such as the formation of papillae, tyloses and abscission zones; necrotic changes, commencing with the efflux of protons and potassium ions from the cell and culminating in the oxidative destruction of the cell contents by lipid hydroperoxides and reactive oxygen species; and toxic changes, including the accumulation of phytoalexins, the synthesis of phenolic compounds and their subsequent oxidation to quinones by polyphenol oxidase and peroxidase (Agrios. 1997). Systemic defences involve the accumulation of anti-microbial compounds
in parts of the plant distant from the site of infection. Four main classes of compound can accumulate: hydrolases, particularly the pathogenesis related proteins (PR) (Stintzi et al., 1993). Defensins proteinase inhibitors (Schaller and Ryan, 1996) and cell wall components, particularly hydroxyproline-rich glycoproteins (HRGP) (Agrios, 1997) and lignin and its precursors (Sticher et al. 1997).

Local defences usually involve the death of the cell reacting to the challenge, by dehydration and abscission or by the hypersensitive reaction. Hence it is only the systemic, uninfect ed parts that can express true induced resistance, and this necessarily implies the existence of systemic signals from the necrotic areas to the rest of the plant. Five types of systemic induced resistance are known:

- Local acquired resistance (LAR), expressed in the immediate vicinity of the hypersensitive fleck caused by attempted pathogen invasion (Agrios, 1997)
- Systemic acquired resistance (SAR), expressed in the plant as a whole in response to pathogen attack (Ryals et al., 1994).
- Systemic gene silencing (SGS), a putative explanation for the recovery phenomenon by which systemic parts of a plant can exhibit resistance to the viruses in infected parts.
- Induced systemic resistance (ISR), which is induced by plant growth promoting rhizobacteria (PGPR) and expressed systemically.
- Systemic wounding response (SWR), caused by the wounds inflicted by chewing insects and leading to the induction of proteinase inhibitors in systemic parts of the plant (Schaller and Ryan, 1996).
Induced resistance has often been likened to immunisation in mammals, although this is not strictly accurate. The plant 'immune response' is much more like the innate immune response of animals than the adaptive, in that the plant does not develop specific resistance to the challenging pathogen, but rather a broad spectrum resistance to several (Sticher et al., 1997) As there is no evidence that elicitors can be translocated systemically, the systemic portions of the plant only receive general information that the plant is under attack (not specific information about the pathogen species). Hence, the plant can only produce a broad-spectrum resistance, presumably against the pathogens that have been most important in selecting for induced resistance over evolutionary time.

The five induced resistance types are not isolated phenomena, which makes both their study and interpretation difficult because the pathways of signal transduction in at least the first four have common elements. Further, abiotic stresses such as drought and wind can modify the responses (by the production of ABA and ethylene) and ozone can actually induce SAR (Sharma et al. 1996) by the production of reactive oxygen species.

**Local acquired resistance (LAR)**

LAR occurs in the tissues immediately surrounding the hypersensitive flecks caused by attempted pathogen invasion. It may simply be SAR at a short distance, but evidence suggests that at least part of the mechanism is different. At the point of contact between pathogen and plant, elicitors from the pathogen are able to interact with plant receptors, such as the leucine-rich-repeat proteins (Jones, 1997) which are not found in animal cells. By a transduction pathway yet to be elucidated, but possibly involving Ca\(^{2+}\) and/or protein kinases, the production of
hydrogen peroxide is initiated, probably by an NADPH oxidase, in analogy to that found in mammalian phagocytes. The peroxide kills the cell and has been found to induce salicylic acid (SA) synthesis. SA has been implicated in hypersensitive cell death: it may be involved in a positive feedback loop with peroxide via an inhibition of catalase, and appears to be necessary for cell death in *lsd (lesions simulating disease)* mutants 1, 6 and 7. If SA is produced at sufficient concentrations, it may be able to induce LAR in surrounding live tissues, without the need for the true systemic signal implied in SAR. Hence, HR leads to LAR which itself initiates SAR.

**SAR (Systemic acquired resistance)**

SAR is the resistance of tissues distant from the site of attempted penetration by a pathogen to a broad spectrum of potential pathogens (not just to the one that initiated the response). It differs from the pathogen-specific competitive effect a weak strain of a pathogen may show against a stronger strain of the same pathogen. SAR has three main components: an accumulation of PR proteins (termed SAR proteins, since PR protein accumulate at the HR fleck too), lignification and cross-linking of the cell walls of tissues distant from the HR, and the phenomenon of conditioning.

Systemic acquired resistance (SAR) refers to a distinct signal transduction pathway that plays an important role in the ability of plants to defend themselves against pathogens. After the formation of a necrotic lesion, either as a part of the hypersensitive response (HR) or as a symptom of disease, the SAR pathway is activated. SAR activation results in the development of a broad-spectrum, systemic resistance (Hunt and Ryals, 1996; Neuenschwander *et al.*, 1995). Although SAR is
interesting as a paradigm for signal transduction, it may have practical value as well.

Plants have evolved efficient mechanisms to combat pathogen attack. One of the earliest responses to pathogen attack is the generation of oxidative burst that can trigger hypersensitive cell death. This is called the hypersensitive response (HR) and is considered to be a major element of plant disease resistance. The HR is thought to deprive the pathogens of a supply of food and confine them to initial infection site.

The hypersensitive response, or HR, is a form of cell death often associated with plant resistance to pathogen infection. Reactive oxygen intermediates and ion fluxes are proximal responses probably required for the HR. Apoptosis as defined in animal systems is, thus far, not a strict paradigm for the HR. The diversity observed in plant cell death morphologies suggests that there may be multiple pathways through which the HR can be triggered. Signals from pathogens appear to interfere with these pathways.

Etheric oils

Besides plant extracts, etheric oils have been tested against *E. amylovora in vivo* and *in vitro*. An antibacterial effect against *E. amylovora* with essential oils from origanum, thyme, savory, cinnamon and garlic. In their studies an influence of essential oil constituents on bacterial growth was determined, as for instance by the terpenoids geraniol and citronellol. In addition, an etheric oil from *Thymbra spicata* was reported as an induction agent of systemic acquired resistance (SAR).
**Synthetic compounds**

The growth regulator Prohexadione-Ca has also been tested as an alternative compound against fire blight. However, sufficient efficacy in field experiments could not be obtained. Recently, the compound harpin was released on the US-market as resistance inducer, but it has not yet been tested against fire blight in the field (Psallidas and Tsiantos, 2000).

**ELICITOR**

In recent times, induced plant defense response methods emerge as the apt substitutes for the chemical pesticides. The most viable method is to protect the plant against pathogen by inducing resistance without changing any characteristics of the host plant due to the application of chemical substances called elicitors. A molecule, which is able to activate the immune system in the infected plants against pathogen attack, is commonly referred to as an ‘ELICITOR’ (Kogel *et al.*, 1989); but it is now commonly used for compounds stimulating any type of plant defense (Ebel and Cosio, 1994). Induction of plant resistance mechanisms (NDR-Natural disease resistance) by application of elicitor has become a realistic component in the crop protection strategies (Rajan Katoch, 2005). Eventually, the induction of defense responses may lead to enhanced resistance. Elicitors of NDR may be chemical, physical and biological.

Physical inducers include low temperature storage, wounding (Ismail and Brown, 1979) CO₂ treatment, heat treatment, ionizing irradiation and UV-C irradiation. Chemical inducers comprise of SA, JA, and ethylene. A number of
microbial antagonistic organisms are capable of inducing defense reactions in host for the control of diseases.

The broader definition of elicitors includes both substances of pathogen origin (exogenous elicitors) and compounds released from plants by the action of the pathogen (endogenous elicitors) (Ebel and Cosio, 1994). Chemical elicitors do not have any common chemical structure, but belong to a wide range of different classes of compounds including oligosaccharides, peptides, proteins and lipids.

The elicitors have several distinct advantages over chemical fungicides. Control of plant diseases by using elicitor is through the induction and enhancement of the plant's own defense mechanisms. Fundamental studies have helped us to understand many of the biochemical interactions occurring between host plants and plant pathogens. Thus we are able to describe resistance reactions as involving not only some preformed components but more importantly, an induced response to infection which includes a 'cascade' of induced responses. These include synthesis of novel antimicrobial compounds-phytoalexins, enzymes and PGIP (polygalacturonase inhibiting protein) and physical barriers to penetration. This cascade of resistance factors is induced when a plant recognizes that a potential pathogen is present and compounds which are capable of triggering such responses are termed elicitors.

Salicylic acid (SA) is one of the well known natural chemical elicitors of disease resistance in plants (Sticher et al., 1997). In recent years salicylic acid has been used as an alternative for fungicides of chemical and biological origin. Salicylic acid is also a signal molecule in systemic acquired resistance (SAR) and an inducer of the alternative oxidase protein synthesis as in tobacco cell
suspensions and during thermogenesis in aroid spadices (Raskin, 1992). Spray of salicylic acid on some plants triggers natural defenses that keep pathogenic fungi away. Plants make salicylic acid, particularly after encountering a pathogen, and use it as a key regulator of SAR and expression of defense genes. Salicylic acid plays a key role in both disease resistance and systemic acquired resistance signaling (Ryals et al., 1996). **Salicylic Acid** (SA) has been implicated strongly in the signal transduction pathway of SAR. The endogenous levels of SA increase during SAR induction.

**EFFECT OF SALICYLIC ACID (ELICITOR)**

Plants have always had some means to defend themselves; it's just that some don't recognize their microbial attackers in time. Spraying salicylic acid or certain other compounds puts their defenses on high-alert against future attacks.

Plant scientists first encountered the phenomenon, called systemic acquired resistance (SAR), in the 1930s. Plants make salicylic acid, particularly after encountering a pathogen, and use it as a key regulator of SAR and expression of defense genes. But only recently have companies begun marketing salicylic acid and other similar compounds as a way to activate SAR in crops-tomato, spinach, lettuce, and tobacco among.

The effect of salicylic acid (SA) was investigated in basal defence and induced resistance to powdery mildew (*Oidium neolycopersici*) and grey mould (*Botrytis cinerea*) in tomato (*Lycopersicon esculentum*) and tobacco (*Nicotiana tabacum*). Acetyl salicylic acid (ASA) treatments of potato foliage of field- and glasshouse-grown potato plants were compared for the control of two foliar
diseases, early blight (*Alternaria solani*) and powdery mildew (*Erysiphe cichoracearum*). Knauss (1977) identified salicylic acid (SA) and its derivatives as potential activators for induction of resistance in plants. Increased resistance in potato tubers against *Erwinia carotovora* sp. was observed when tubers were dipped in acetyl salicylic acid (ASA).

SA has been shown to play an important role in expression of both local resistance controlled by major genes and systemic induced resistance developed after an initial pathogen attack. Application of exogenous SA at a concentration of 1 to 5mM has been known to induce pathogenesis-related (PR) gene expression and acquired resistance against a variety of pathogens (Meena *et al.*, 2000). Anandhi and Ramanujam (1997) studied the effect of SA on seed formation and seedling growth of two cultivars of *Vigna mungo*. Effects of SA on nodulation, nitrogenous compounds and related enzymes of *Vigna mungo* were brought out by Ramanujam and his co-workers (1998) reported that reduction in the *Fusarium* wilt disease in chickpea was more pronounced when chemical resistance inducers (SA, acetyl SA, DL-norvaline, indole-3-carbinol and lichenon) were applied with *Pseudomanas fluorescens*. Among all chemical inducers, SA showed the highest protection against the disease. At lower concentration of SA (2000 mg ml\(^{-1}\)) mycelial growth was completely arrested.

However, survey of literature reveals that there seems to be no report on the effect of salicylic acid elicitor in controlling *Colletotrichum capsici* causing anthracnose disease in *C.annuum*. Hence, the present study was undertaken to find out the effects of salicylic acid on the induction of hypersensitive responses in the host plant, *Capsicum annuum* against the infecting *Colletotrichum capsici*.
OBJECTIVES:

Scope of the present investigation

The present study aimed at finding out the effects of salicylic acid in controlling *C. capsici* causing anthracnose of *Capsicum annuum* by studying the following parameters.

Until now, not much information is available regarding physiological changes and hypersensitive response (HR) in chilli against *C. capsici* during induced resistance. In the present study, salicylic acid was compared with two agents they are the synthetic fungicide Bavistin, and a biotic agent a *Gliocladium virens*. Experiments were primarily conducted to study the physiology of induced resistance, besides the direct efficacy of these agents against the disease.

The objectives of the present study were to find out the efficacy of salicylic acid in controlling *C. capsici* causing anthracnose of *Capsicum annuum* by studying the following parameters *in-vitro* and *in-vivo*.

*In-vitro* studies:

I. To evaluate of the efficacy of salicylic acid (elicitor) in comparison to that of *Gliocladium virens* (microbial antagonist) and Bavistin (chemical fungicide) on the various parameters under *in-vitro* condition.

1 Conidial and radial mycelial growth of *C. capsici*.
2 Pectinolytic and cellulolytic activities of *C. capsici*.
3 Extraction of polygalcturonase (PG) from fungus *Colletotrichum capsici*. 
In-vivo Studies:

II. To Study the effects of salicylic acid in inducing hypersensitive (HR) responses by studying the following parameters in the leaves of host plant *C. annuum* under control ((healthy), healthy treated, infected and infected treated conditions.

1. Study of host-pathogen interaction by analyzing the biochemical changes viz. electrolytic leakage, chlorophyll, sucrose, starch, carbohydrate, DNA and RNA, protein, phenol, proline and endogenous salicylic acid.

2. Estimation of the antioxidant enzymes such as superoxide dismutase, catalase, peroxidase and polyphenol oxidase.

3. Estimation of phenyl alanine ammonia lyase enzyme (PAL).

4. Purification of PGIP (Polygalacturonase inhibiting protein) from the leaves of *C. annuum*.

5. Assessment of PG inhibition by PGIP.