

**ROOT ROT OF CHILLI INCITED BY *Sclerotium rolfsii* Sacc. AND ITS MANAGEMENT- A
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Chilli (*Capsicum annum* L.) belongs to the family solanaceae is mainly cultivated for its vegetable green fruits and dry chilli as the spice. It is believed to be originated from South America. It is a rich source of Vitamin C, A and B. Chilli is valued for pungency which is imparted by an alkaloid, Capsaicin and the red pigments (Capsanthin, Capsorubin and Capxanthin). Chillies are widely used as spice, condiment, culinary, supplement, medicine, vegetable and are ornamental plants too. It is an important food flavouring ingredient for flavouring many vegetarian and non-vegetarian food products. In view of its multifarious uses, the demand for chillies has been on the increase world over. In India it is an important cash crop, which is grown for the domestic market and for export. In India, chillies are grown in almost all states of the country and the major growing states in terms of production are Andhra Pradesh, Karnataka, Orissa, Maharashtra, West Bengal, Rajasthan and Tamil Nadu. There are several varieties of chilli cultivated in India. The most popular among these are, Sannam, LC334, Byadagi, Wonderhot, Jwala etc.

The crop suffers from many diseases like damping off, anthracnose or fruit rot or dieback, wilt, leaf spots and powdery mildew. Among the fungal diseases, root rot of chilli caused by *Sclerotium rolfsii* has attained the economic importance. In recent years, this disease is causing the economic losses in chillies crop (Kalmesh and Gurjar 2001).

The disease

In India the root rot caused by this pathogen was first time reported by Shaw and Ajrekar (1915) who had isolated the pathogen from rotting potatoes which they identified as *Rhizoctonia destruens* Tassi. Later studies convinced Ajrekar that the fungus involved in rotting of potato was *Sclerotium rolfsii* but not *R. destruens* (Ramakrishna, 1930). During 1985, the color rot to chilli caused by *Sclerotium rolfsii* was observed in Maharashtra at Vidarbha region (Wangihar, 1985).

Sclerotium rolfsii Sacc. is a well known polyphagous, ubiquitous, omnivorous and most destructive soil borne fungus. This was first time reported by Rolfs (1892) as a cause of tomato blight in florida. Later, Saccardo (1911) named the fungus as *S. rolfsii* sp. But, in India, Shaw and Ajrekar (1915) isolated the fungus from rotted potatoes and identified as *Rhizoctonia destruens* Tassi. However, later studies showed that, the fungus involved was *S. rolfsii* (Ramakrishnan, 1930). Mundkar (1934) successfully isolated the perfect stage of *S. rolfsii*. Kulkarni *et al.* (1994) while studying the most susceptible growth stage of groundnut to *S. rolfsii* maximum infection, colonization, disease development and mortality was recorded in 15 days old plants and least, mortality in 105 days old plants.

Morphology of the Pathogen

Narsimha rao (2000), reported that, fungus *Sclerotium rolfsii* showed characteristics with white, radiating abundant mycelial growth on the affected portion of potato plant and tuber. Large number of sclerotial initial were noticed on the mycelial mat. Initially sclerotia exhibited white colour later turn to chocolate brown. Sclerotia were superficial on the affected part, spherical or ellipsoidal and measured 0.5 mm to 2.5 mm in diameter. Non-germianted sclerotia become soft and broke easily. Maceration of cortical cells was common. A bacterial like organism was consistently associated with the ruptured and macerated sclerotia.

Symptomatology

Kulkarni *et al.* (1995) reported that the pathogen damaged stem, root or tuber and infected the stem and produced dark brown lesions at collar portion causing wilt and ultimately plants get dried. Brownish sclerotia resembled like mustard seed, developed at later stages on the root and collar region of the infected plants. Afterwards tubers get infected and rotten in field. Kalmesh and Gurjar (2001) described the symptoms of root rot of chilli caused by *S. rolfsii*. Severe mortality of chilli plants was observed during the months of March-April in chilli growing areas. Mature plants of chilli from standing crop were collapsed and dried down suddenly. Close examination of the diseased plants showed deep cracks near collar region. Roots were shredded and unhealthy with full of white mycelial growth on the surface of freshly infected area. Somani and Chuhan (1996) observed a thin white mycelial mat on freshly harvested tuber surface and at times thick mat with mustard seed like sclerotia. *S. rolfsii* infected plants had white to mustard coloured sclerotia and thin white mycelial mat on almost all the infected plant parts and daughter tubers if the plants wilted after tuber formation. They also observed wilt at early stage before tuberization. In the heap, the rot was spread from diseased to healthy tubers.

Isolation and Proving Pathogenicity

Ramakrishnan *et al.* (1955) isolated *Sclerotium rolfsii* from the roots of the affected Chilli plants. Inoculations with this isolate produced hundred per cent infection on chilli plants while the control plants remained healthy. The isolate was also pathogenic to groundnut, ginger and zinnia (Garden plant). Sariah *et al.* (1995) reported that the viability of sclerotial bodies of *S. rolfsii* isolated from seedling blight and collar rot of chilli decreased rapidly with the time of submerging in flooded field soil. Surendranath (1999) also reported that, fully matured sclerotia of *S. rolfsii* the causal agents of white rot of onion were spherical to ellipsoidal and measured 0.5 mm to 1.75 mm in diameter. Artificial inoculation of the plants with the pathogen was done by different methods. Soil inoculation by the pathogen was studied by several workers Kajalkumar and Chitreswaran (2000), Gupta and Ashu Sharma (2004), Rajani *et al.* (2006) and Anitha Chowdary (1997). Seedling root dip in inoculum suspension was used to induce sclerotial wilt in bell pepper (Anita Chowdary, 1997).

Sclerotium rolfsii was isolated from different plants viz., diseased seeds and seedlings (Rama rao and Usharaja, 1980. Uma Singh and Thapliyal, 1998. Rajeevpant and Mukopadhyay, 2001), stem (Kajal kumar and Chitreswar Sen, 2000), collar region (Ansari and Agnihotri, 2000., Narasimha Rao *et al.*, 2004), root (Srikanta Das and Raj, 1995., Harinath Naidu, 2000 and Gaur *et al.*, 2005), tubers, crown, leaves and pods (Gupta and Ashu Sharma, 2004).

Maintenance of the Pathogen

Potato Dextrose Agar (PDA) was found to be the best supporting medium for *S. rolfsii* (Harinath Naidu, 2000, Amarsingh and Dhanbir Singh, 1994, Gupta and Ashu Sharma, 2004., Gaur *et al.*, 2005 and Raoof *et al.*, 2006). Narain and Mishra (1979) found that malt extract agar supported the large number and size of sclerotia of ragi isolate of *S. rolfsii*. *S. rolfsii* can also be maintained on potato sucrose agar medium (Ramarao and Usharaja, 1980).

Mass Multiplication of the Pathogen

The pathogen was multiplied on sorghum grains (Gupta and Kolte, 1982). Sorghum grains were pre-soaked in 2 per cent sucrose solution overnight, drained and boiled in fresh water for 30 minutes and drained again. This was transferred into 1000 ml flasks @ 400 g and autoclaved at 15 lb psi (121.6°C) for 20 minutes. The flasks were allowed to cool at room temperature and inoculated with five mm discs of 3 to 4 days old culture of *S. rolfsii* grown on PDA. Seven discs per flask were added and flasks were incubated for three weeks at $28 \pm 2^{\circ}\text{C}$.

In vitro Evaluation of Fungicides, Herbicides, botanicals, and bio-Agents Against *S. rolfsii*

Fungicides

Aycock (1959) reported that, soil application of captan at the rate of 87 lb per acre was found effective in controlling *S. rolfsii* without any phytotoxic effect. Effectiveness of plantavax and vitavax was recorded in inhibiting the vegetative growth of *S. rolfsii* while working with foot rot of various crop plants (Vyas and Joshi, 1977; Siddaramaiah *et al.*, 1979; Peshney and Moghe, 1980; Palakashppa, 1986; Kulkarni *et al.*, 1986). Siddaramaiah *et al.* (1979) reported that, calixin (Triademorph) completely inhibited the mycelial growth of *Sclerotium rolfsii* under *in vitro* condition, they also found the inhibition of sclerotial germination by Bayletan and Panoram at 100 ppm. Harlapura (1988), Vyas and Joshi (1977) reported the efficacy of thiram in inhibiting the growth of *S. rolfsii*, the casual agent of foot rot of wheat. Further, Harlapur (1988) noticed the complete inhibition of mycelial growth of *Sclerotium rolfsii* by agallol and Diathans M-45. Propiconazole was found highly effective in inhibiting the mycelial growth of *Sclerotium rolfsii* (Waterfield and Sister, 1990; Hagans *et al.*, 1992). Hagans *et al.* (1992) also reported the efficacy of tebuconazole, diniconazole and flutatanil against *S. rolfsii* a causal organism of stem rot of peanut. Patil *et al.* (1986) reported effectiveness of captan in reducing the mortality of *Piper betel* L. due to *S. rolfsii*. Pal and Choudhary (1983), Anilkumar and Pandurange Gowda (1984) reported the efficacy of vitavax as soil drenching fungicide against *S. rolfsii* in reducing the sunhemp seedling mortality. However, they reported that, bayleton and sicorol were also found highly effective for soil drenching.

Soil drenching with Dithane M-45, captan, difolatan and Boredeux mixture were found highly effective in reducing the mortality of Piper betel L. due to *S. rolfisii* (Patil *et al.*, 1986 and Virupaksha *et al.*, 1997). Harlapur and Srikant (1992) reported that, vitavax, bayleton and bayton were highly effective at all concentrations tested against, foot rot of wheat caused by *S. rolfisii* under glasshouse conditions and also that of vitavax was found to be the best in reducing the preemergence mortality of wheat seedlings. Advier and Anahosur (1995) observed that, all four ergosterol biosynthesis inhibiting trizole fungicides tested were found effective against *S. rolfisii* by reducing stem colonization to the extent of 64.2 per cent. Pranabdatta and Das (2002) conducted experiments for the management of collar rot of tomato by using chemicals and *T. harzianum*, *T. viride* and *T. koningii*. Among the bioagents, *T. harzianum* was found to be more inhibitory to *S. rolfisii* in dual culture technique. Among the chemicals, Dithane M-45 was found more effective. Madhavi (2011) reported the *in vitro* evaluation of nine fungicides by poison food technique showed that tebuconazole and combination of carbendazim+mancozeb were effective in inhibiting the mycelial growth (94.1%) followed by difenconazole (93.3%).

Herbicides

The effect of ten herbicides incorporated into PDA at 50, 100 and 500 $\mu\text{g ml}^{-1}$ reduced the mycelial growth and sclerotial formation of *S. rolfisii*, *M. phaseolina* and *F. oxysporum* isolated from the soybean cv JS-72-44 *in vitro* by Vyas *et al.* (1986a). 2, 4-D and fluchloralin drastically inhibited the growth of *S. rolfisii* and *R. bataticola* (Tripathi *et al.*, 1988). Lal and Nagarajan (1988) reported the evaluation of herbicides alachlor, basalin and trifluralin against *S. rolfisii* causing collar rot disease of tobacco at 125, 250, 500, 1000 and 2000 ppm concentrations. The per cent growth inhibition was 67.3 – 88.14 upto 500 ppm concentration. While cent per cent inhibition was noticed at higher concentrations (1000 and 2000 ppm). The effect of Dicuran MA on the colony growth of five soil borne fungal pathogens of chickpea (*Macrophomina phaseolina*, *Sclerotinia sclerotiorum*, *Sclerotium rolfisii*, *Rhizoctonia solani* and *Fusarium oxysporum*) was tested under *in vitro* conditions and the results showed that the treatment decreased the growth of all the pathogens tested, but to different extents. *S. rolfisii* was completely inhibited and *M. phaseolina* showed low sensitivity and the growth of other fungi was inhibited by 50- 62.4 per cent (Iqbal *et al.*, 1994). The effects of acetochlor, imazethapyr, metachlor, pendimethalin, trifluralin and mixture acetochlor and imazethapyr on the production and viability of *S. rolfisii* sclerotia were evaluated *in vitro* by Pastro and March (1999). Trifluralin and pendimethalin were the most efficient compounds because they notably reduced the production of viable sclerotia. Madhuri and Narayan Reddy (2013) evaluated eight herbicides tested for their efficacy on *S. rolfisii* by poisoned food technique and found that oxyflourfen, alachlor, quizalofop-p-ethyl and 2, 4-D sodium salt completely inhibited the growth of *S. rolfisii*.

Botanicals

Dutta and Deb (1986) studied the effect of organic and inorganic amendments on the soil and *Rhizosphere microflora* in relation to the biology and control of *Sclerotium rolfisii*. They reported that, leaf extract of *Eupatorium adenophorum* reduced the pathogen population in the rhizosphere. Sivakadacham (1988) reported that, leaf extracts of *Azadirachta indica* and *Cullen corylifolium* suppressed the mycelial growth of *Sclerotium rolfisii*. Singh *et al.* (1989) reported that, out of six plant oils tested against *S. rolfisii*, leaf oil of *Azadirachta indica* was found most effective followed by that from Eucalyptus globules and *Ocimum canum*. Singh and Dwivedi (1989) noticed that, leaf extracts of twelve plants were found toxic against *S. rolfisii*. Further, they also observed the morphological changes and significant reduction in mycelial growth of *S. rolfisii*. Singh and Dwivedi (1990) reported that, the viability of sclerotia was reduced when treated with neem oil. Dayaram and Tewari (1994) found that, the soil application of green leaves of *Azadirachta indica*, *Aegle marchelos*, *Anisomeles ovata*, *Azadirachta indica*, *Cymbopogon flexuosus*, rhizomes of *Curcuma amada* and resin of *Ferula foetida* at 2 to 5 per cent concentration reduced both pre and post emergence collar rot of chickpea caused by *Sclerotium rolfisii*. Five per cent *Ferula foetida* resin applied 48 hours before sowing of seeds in artificial inoculation of soil provided nearly 100 per cent protection. Parimelazhagan and Francis, 1999 reported that leaf extract of *Clerodendron viscosum* completely checked the radial growth of *Curvularia lanata*. Seshakiran (2002) reported that, *Eupatorium odoratum* L., *C.occidentalis* and *Azadirachta indica* were highly antifungal to mycelial growth of *S. rolfisii*. However, root extract of *Pathenium hysterophorus* L. exhibited maximum inhibition of mycelial growth of *S. rolfisii*.

Bioagents

Weinding (1932) was first to demonstrate the antagonistic effects of a soil fungus to *Sclerotium rolfisii*. He found over growth of hyphae of *Trichoderma viride* Pers on *S. rolfisii*. It secreted some substance which was found lethal to *Sclerotium rolfisii*. Later, Brain (1951) identified the antagonistic substance as gliotoxin. McMilan *et al.* (1949) suggested that, reduction in germinability of sclerotia was due to the action of antagonistic organisms Garrett (1956) defined the biological control of plant diseases as “any condition under which, survival and activity of a pathogen is reduced through the agency of any living organisms”.

Many workers reported that, *T. viride* was found to be antagonistic to *S. rolfsii* (Hino and Endo, 1940; Manjappa, 1979; Henis *et al.*, 1983; Khetmalas *et al.*, 1984; Pande, 1985; Palakshappa, 1986 and Patil, 1993). *Penicillium* sp. showed an antagonistic effect towards *Sclerotium rolfsii* which has been reported earlier by several workers (Agrawal *et al.*, 1977; Manjappa, 1979 and Virupaksha Prabhu *et al.*, 1997). *Trichoderma harzianum* Rafai was found antagonistic to *Sclerotium rolfsii* (Agrawal *et al.*, 1977; Arora and Dwivedi, 1979; Chet *et al.*, 1979; Elad *et al.*, 1980; Elad *et al.*, 1983 and Upadhyay and Mukhopadhyay, 1983). D' Ambra and Ferrata (1984) observed the reduction of mycelial growth, sclerotial formation, sclerotial germination and number of sclerotia of *Sclerotium rolfsii* when inoculated with different inoculum concentration of *Trichoderma harzianum*. Wokocho *et al.* (1986) noticed that, a native strain of *Pseudomonas fluorescens* Migula restricted the mycelial growth of *Sclerotium rolfsii* on peanut. Similarly, Ganesan and Gnanamanikyam (1987) found that, the native station of *P. fluorescens* restricted the growth of *S. rolfsii*. *Sclerotium* showed 10-20 per cent loss of germination after immersion in a bacterial cell suspension for one hour and about 50-60 per cent after one week. Sariah Meon (1991), studied hyphal interactions between *Trichoderma harzianum* and *Sclerotium rolfsii* (Pathogen capacity of chilli) where, *T. harzianum* produced thin and long hyphal branches towards *Sclerotium rolfsii*, which formed several short branches and coiled the hyphae of *S. rolfsii*. Muthamilan and Jeyarajan (1992) reported that, 67.4 per cent reduction of sclerotial production in *Sclerotium rolfsii* was observed in the presence of *Trichoderma viride*. Mature sclerotia from each dual culture plate were smaller than the control plate. Virupaksha *et al.* (1997) tested the antagonistic organisms against *Sclerotium rolfsii*. Among them, *Trichoderma harzianum* and *Trichoderma viride* were found to be effective in inhibiting the mycelial growth and reducing production of sclerotial bodies irrespective of inoculation periods. He also observed inhibition zone and reduction in size of sclerotial bodies in presence of antagonists. Iqbal *et al.* (1995) tested the micro-organisms for antagonism to *Sclerotium rolfsii*. All the organisms viz., *Trichoderma harzianum*, *Trichoderma koningii* Ouden, *Trichoderma viride*, *Gliocladium virens* Miller, *Aspergillus candidus* Link, *Paecilomyces lilacinus* (Thom) Samson and *Bacillus* spp. significantly inhibited the mycelial growth of *S. rolfsii*, *Trichoderma harzianum*, *Trichoderma koningii* and *Trichoderma viride* overlapped the pathogen and suppressed growth by 63.6 per cent, 54.9 per cent and 51.89 per cent respectively. The maximum inhibition zone was seen in *T. harzianum* and *T. viride*. This might be due to the production of antibiotics, which diffused air filled pores which are detrimental to the growth of *S. rolfsii* (Upadhyay and Mukhopadhyay, 1983; Karthikeyan, 1996 and Bhagwat, 1997). Narasimha Rao (2000) stated that, biological control is an eco-friendly and effective means of reducing the disease through potential antagonistic micro-organisms. Madhavi and Bhattiprolu (2011) evaluated five bio-agents against *S. rolfsii* revealed that, there is significant difference in per cent inhibition of mycelial growth of *S. rolfsii* by all the bioagents tested. Per cent mycelial inhibition of *Sclerotium rolfsii* was found highest (57.5%) with *Trichoderma harzianum* was significantly superior over the rest of bioagents tested. This was followed by *Trichoderma viride* I (55.8%) followed *Trichoderma viride* II (53.63 %), *T. hamatum* caused 44.46% inhibition and 40.7 % inhibition was recorded with *Pseudomonas fluorescens*.

In Vivo Studies

The superiority of mancozeb compound against *Sclerotium rolfsii* can be attributed due to inhibition of pentose phosphate pathway and chelation of required heavy metals followed by lethal catalysis in pathogen cell (Nene and Thapliyal, 1993). Integration of cultural, biological and chemical means of control has proved to be a very promising way of management soil-borne pathogens such as *S. rolfsii* (Upadhyay and Mukhopadhyay, 1986; Natarajan and Manibhushan, 1996 and Saralamma, 2000). Bhoraniya *et al.* (2002) reported the inhibition of sclerotial germination of *S. rolfsii* by different pesticides viz., metalachlor, fluchloralin, alachlor, pendimethalin, 2,4-D sodium salt, mancozeb, captan, copper oxychloride, tridemorph and carboxin through soil plate technique. The effectiveness of carboxin (98.99 per cent) followed by tridemorph (97.89 per cent) and fluchloralin (94.02 per cent). However, they were statistically at par (P=0.05), metalachlor was also found effective in inhibiting the sclerotial germination (62.78 per cent). *In vivo* soil drenching was done with nine fungicides and found that tebuconazole and combination of carbendazim+mancozeb proved effective in controlling the pathogen at 1000, 2000 and 3000 ppm (Madhavi and Bhattiprolu, 2011).

Integrated Management of Root rot of Chilli Caused by *S. rolfsii*

Maiti and Sen (1985) integrated the biocontrol agent *T. harzianum* multiplied on wheat bran with urea for soil application and found that viability of sclerotia of *S. rolfsii* significantly reduced and also increased survival of *T. harzianum* population. Patil *et al.* (1986) observed that, soil drenching of captan, copperoxychloride and difolaton were found effective in controlling wilt of betelvine caused by *S. rolfsii*. Alagarsamy and Sivaprakasam (1988) observed that, pilleting cowpea seeds with *T. viride* either alone or in combination with carbendazim inhibited the growth of *M. phaseolina* *in vitro*.

The treatment increased the germination and reduced the post emergence mortality under the pot culture conditions. Wokocha (1990) reported that tomato disease caused by *S. rolfsii* was controlled very effectively by using *Trichoderma viride* and some fungicides. Asghari and Mayee (1991), reported that, onion and garlic crop rotation with groundnut crop, application of *T. harzianum* and soil drenching with 0.2 per cent carbendazim reduced the stem rot of groundnut caused by 44-60 per cent and increased the pod yields by 17-47 per cent. Mukthapadhyay *et al.* (1992) reported biological seed treatment for control of soilborne pathogens by integration of biological (*Trichoderma* and *Gliocladium* species) and fungicidal (Carboxin (0.1%) seed treatments which has been found to improve the disease control potential to a great extent. Bicici *et al.*, (1994) reported that rotation of maize and wheat with groundnut spacing of 90 cm between rows, application of quintozone (PCNB) in combination with urea and calcium ammonium nitrate, soil solarisation with polythene sheet and application of biocontrol agent (*T. harzianum*) effectively reduced stem rot of groundnut. Hanumanthe Gouda (1999) reported that, seed treatment *T. harzianum* and soil application of *T. harzianum* recorded the low incidence of groundnut stem rot caused by *Sclerotium rolfsii*. Anahosur (2001) conducted an experiment on integrated disease management in farmers field during the rainy season of year 1999-2000. The results were encouraging and concluded that, IDM comprising the components *viz.*, crop rotation in rabi with rabi sorghum varieties for 2 years + application of FYM to the soil + Tuber treatment with *T. viride* or *T. harzianum* prior to planting @ 4 g kg has helped in reducing the sclerotium wilt of potato in the field. Vanitha and Suresh (2002) conducted a study to investigate efficacy of biological control agents and organic amendments in controlling collar rot of brinjal caused by *Sclerotium rolfsii*, where *Trichoderma viride* + FYM + dry adathoda leaf powder were found effective. Integration of different treatments including seedling dip with carbendazim+mancozeb, addition of vermicompost, drenching with fungicide and application of *Trichoderma harzianum* (7%) were found to be effective in management of disease in comparison with individual treatments (Madhavi and Bhattiprolu, 2011).

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