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Coffee leaf blight disease caused by *Colletotrichum gloeosporioides* and bio-efficacy of fungicides to restrain the pathogen

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Coffee is an important plantation crop grown in India. Coffee plants is affected by fungal diseases like leaf rust, black rot, root diseases, stem canker and anthracnose in field conditions. Coffee Anthracnose caused by *Colletotrichum gloeosporioides* has three important symptoms *viz.,* twig die-back, leaf blight and stalk rot of berries. Leaf blight is observed during dry months and symptoms include small irregular necrotic spot brownish in colour initially and later develop yellowing along veins and midrib of the leaves which cause blightening of leaves leading to cessation. Black pin head like fruiting bodies of the pathogen were also observed on the necrotic spot. Leaf blight infected Arabica coffee (Sln.3) leaf samples were collected and subjected for isolation and purification of the pathogen *C. gloeosporioides* on PDA medium further Koch's postulates was proved. Seven fungicide molecules *viz.,* Carbendazim 50 WP, Hexaconazole 5 EC,Propiconazole 25 EC, Tebuconazole 430 SC, Mancozeb 75 WP,Pyraclostrobin 133g/l + Epoxiconazole 50 g/l w/v SE, Carbendazim 12% + Mancozeb 63% WP at three concentrations of (250, 500 and 1000 ppm) were evaluated against coffee leaf blight pathogen *C. gloeosporioides* under *in vitro* conditions by following poison food technique. Studies indicated that, the fungicides Carbendazim 50 WP,Mancozeb 75 WP and Carbendazim 12% + Mancozeb 63% WPwere effective and inhibited cent per cent mycelial growth of *C. gloeosporioides* in all the tested concentrations. Whereas, in Propiconazole 25 EC and Pyraclostrobin 133g/l + Epoxiconazole 50 g/l w/v SE complete inhibition was observed at 500 and 1000 ppm concentrations.

Key words: Coffee, leaf blight, *Colletotrichum gloeosporioides,* fungicides

INTRODUCTION

Coffee is one of the leading commodities in the international trade and comes next to the petroleum products in terms of volume and foreign exchange and is predominantly grown in the tropical and subtropical regions of the world. In India, major diseases affecting coffee plant is only by fungal pathogens *viz.*, leaf rust, black rot, anthracnose, root disease, stem canker and anthracnose in the field conditions.

Anthracnose disease in coffee is caused by *Colletotrichum gloeosporioides* Penz. has wide range on several cultivated crops, particularly tropical perennial crops (Bhat *et al.,* 2014). Coffee plantations are enriched with suitable inter-crops like black pepper, areca, orange, avocado, banana etc. The main inter crop black pepper are susceptible to *C. gloeosporioides*. Cross infection and molecular variability studies indicated strains infecting coffee and black pepper are genetically distinct (Santoshreddy *et al.* 2021). Coffee Anthracnose is reported from most of the coffee growing countries, its importance was recognised only by a few countries *viz.,* Brazil, Vietnam, India and African countries where it causes considerable crop loss every year. Both cultivated species of coffee arabica and robusta were susceptible for Anthracnose disease. Coffee Anthracnose causes three important symptoms *viz.,* twig die-back, the brown blight of leaves, and stalk rot of berries.

Coffee leaf blight disease is generally seen on leaves during hot and dry weather conditions of winter and summer months especially in the plantations exposed to direct sunlight. Injury caused by sun scalding or any other type of wounds on leaves predispose them for infection by *C. gloeosporioides*. Symptoms on infected leaves appear as small irregular necrotic brown color blighted

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lesions initially and later developed yellowing through leaf veins and midrib which caused the blight of leaves leading to cessation (Fig.1). Fruiting bodies of the pathogen were also noticed on the necrotic spots. Coffee plantations in western slope and exposed to sunlight, thin overhead canopy of shade, injury caused by sun scalding or any other type of wounds on leaves predisposes for leaf blight infection. Presently leaf blight disease is managed by maintaining good overhead shade to avoid sun scalding of leaves and by spraying 0.5% Bordeaux mixture. In the present study, Koch's postulates were proved to reconfirm the pathogen infection and efficacy of new fungicide molecules evaluated against *C. gloeosporioides*, the outcomes were conversed in the following paragraphs.

MATERIALS AND METHODS

Isolation, purification and Koch's postulates of Colletotrichum gloeosporioides

Coffee leaves which showed the symptoms of leaf blight disease on arabica coffee Sln.3 (S.795) were collected from Central Coffee Research Institute (CCRI) farm and subjected for isolation and purification of the pathogen *Colletotrichum gloeosporioides* on potato dextrose agar medium plates. The pure culture of the isolated pathogen was inoculated on the coffee seedlings. After the expression of symptoms, the fungus was re-isolated to prove the Koch's Postulates and to confirm the *Colletotrichum gloeosporioides* infection.

Bio-efficacy of fungicide molecules against Colletotrichum gloeosporioides

A total of seven fungicides *viz.,*Carbendazim 50 WP, Hexaconazole 5 EC, Propiconazole 25 EC, Tebuconazole 430 SC, Mancozeb 75 WP, Pyraclostrobin 133 g/L + Epoxiconazole 50 g/L w/v SE, Carbendazim 12% + Mancozeb 63% WP were evaluated against coffee leaf blight pathogen *C. gloeosporioides* in three concentrations i.e., 250, 500 and 1000 ppm under *in vitro* conditions by following poison food technique. Per cent inhibition of radial growth of mycelia was computed based on colony diameter on control plate adopting the following formula:

I = C - T / C × 100, where, I = Per cent inhibition, C=Mycelial growth inhibition in control and T =Mycelial growth inhibition in control in fungicide treatments

RESULTS AND DISCUSSION

Isolation, purification and Koch's postulates of Colletotrichum gloeosporioides

The pure culture of the isolated *Colletotrichum gloeosporioides* was maintained in the laboratory conditions. Coffee seedlings inoculated with *C. gloeosporioides* expressed symptoms after twelve days of inoculation, the symptoms of coffee leaf blight were observed on the inoculated leaves and these leaves were further subjected for re-isolation of the fungus to prove Koch's Postulates. When the pathogen was re-isolated same culture of the pathogen *C. gloeosporioides* was observed and confirmed the cause of coffee leaf blight.

Bio-efficacy of fungicide molecules against Colletotrichum gloeosporioides

Fungicides are the most common and practical method to manage anthracnose diseases. However, fungicide tolerance often arises quickly, if a single compound is relied upon too heavily. Coffee leaf blight disease caused by *Colletotrichum gloeosporioides* can be managed under normal weather conditions with a reasonable spray program. Results of the laboratory experiment on bio-efficacy of different fungicides to manage the coffee leaf blight pathogen *C. gloeosporioides* is discussed in the following paragraphs and details are mentioned in Table 1and Fig. 2.

Results indicated among systemic fungicides, carbendazim 50 WP inhibited cent per cent mycelial growth of *C. gloeosporioides* in all the tested concentrations. Whereas, in propiconazole 25 EC cent per cent mycelial growth inhibition was observed at 500 and 1000 ppm concentrations. Triazoles are sterol inhibiting fungicides affect cytochrome P-450 enzymes inhibitors of sterol C-14 demethylation by this they act against most of the ascomycota group fungal pathogens. Singh *et al.* (2012) revealed that propiconazole 25 EC @ 100 ppm inhibited cent per cent growth of *C. gloeosporioides* isolated from coffee under *in vitro* conditions. Carbendazim is effectively inhibited mycelial growth of *C. gloeosporioides* isolated from Mangrove (Pal, 2020) Contact fungicide mancozeb 75 WP also inhibited cent per cent mycelial growth

Table 1: *In vitro* evaluation of fungicides against coffee leaf blight pathogen *C. gloeosporioides*

	Treatments	Concentration (ppm)			
		250	500	1000	Mean
T_1	Carbendazim 50 WP	100.00 (90.04)*	100.00 (90.04)	100.00 (90.04)	100.00
T_2	Hexaconazole 5 EC	51.11 (45.66)	53.33 (46.93)	100.00 (90.04)	68.15
T_3	Propiconazole 25 EC	99.44 (85.76)	100.00 (90.04)	100.00 (90.04)	99.81
T_4	Tebuconazole 430 SC	79.52 (63.12)	98.89 (83.98)	99.44 (85.76)	92.62
T_5	Mancozeb 75 WP	100.00 (90.04)	100.00 (90.04)	100.00 (90.04)	100.00
T_6	Carbendazim 12% + Mancozeb 63% WP	100.00 (90.04)	100.00 (90.04)	100.00 (90.04)	100.00
T_7	Pyraclostrobin 133g/l + Epoxiconazole 50 g/l w/v SE	89.63 (71.24)	100.00 (90.04)	100.00 (90.04)	96.54
					CD @ 1%
Fungicide (F)					0.40
Concentration (C)					0.54
F x C					1.07

*Figures in parentheses are arc sine values.

in all the tested concentrations.

Use of combi product fungicides avoids the development of resistance of fungi to systemic fungicides because these systemic fungicides interfere with only one or sometimes two functions in physiology of fungus which it easily overcomes by either a single mutation or by selection of resistant individuals in a population. Wherein non-systemic protectant fungicides affect too many functions in fungus physiology and to develop resistance the fungus will have to make too many gene changes. Hence the combination of both systemic and non-systemic fungicides provides better management

Fig. 1 : Symptoms of coffee blight disease

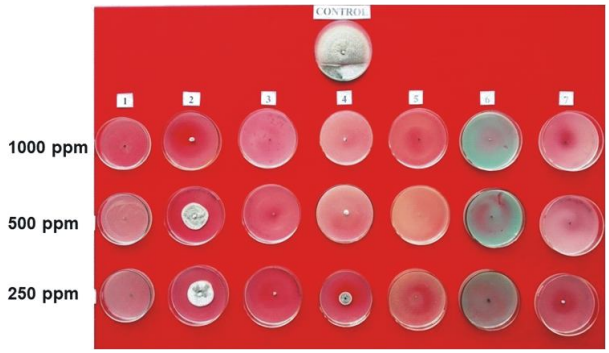

Fig. 2 : Bioefficacy of different concentrations of fungicides against *C. gloeosporioides*.

1- Carbendazinm 50 WP; 2- Hexaconazole 5 EC; 3-Propiconazole 25 EC; 4-Tebuconaxole 430 SC; 5-Mancozeb 75 WP; 6-Carbendazim 12% + Mancozeb 63% WP; 7- Pyraclostrobin 133g/l + Epoxico-nazole 50 g/l w/v SE

of plant fungal disease for long duration Among the two combi-product fungicides carbendazim 12% + mancozeb 63% WP inhibited cent per cent mycelial growth in all the tested concentrations whereas,pyraclostrobin 133 g/L + epoxiconazole 50 g/L w/v SE inhibited cent per cent mycelial growth at 500 ppm and 1000 ppm concentrations.Similarly, carbendazim 12% + mancozeb 63% WP and carbendazim 50 WP significantly managed the an

thracnose disease of black pepper caused by *C. gloeosporioides* (Kurian *et al.* 2008) Pyraclostrobin and mancozeb fungicides were found effective in management of die-back and fruit rot disease in chilli caused by *Colletotrichum* (Santoshreddy and Nargund 2015). Strobulirins act through inhibition of respiration by binding to the Qo center of the cytochrome b. These strobulirins are very broad and balanced spectrum of activity on the foliage and have very favourable toxicological profile rapidly dissipating from soil and surface water which are unlikely to cause hazard to non-target organisms and they have both protective and curative effect.

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