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IN VITRO EFFICACY OF FUNGICIDES AGAINST *COLLETOTRICHUM GLOEOSPORIOIDES* PENZ

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Abstract-The current investigation results showed that all six fungicides were examined compared to the untreated control and demonstrated antifungal efficacy against the test pathogen (*Colletotrichum gloeosporioides* Penz). Six fungicides were evaluated against *Colletotrichum gloeosporioides* in laboratory conditions and the results showed that all of them significantly inhibited the pathogen's growth. Mycelial growth of *Colletotrichum gloeosporioides* was totally (100% inhibition) inhibited by Propiconazole (0.1%), Copper Oxychloride (0.2%), and Difenoconazole (0.1%). These fungicides were followed by Thiophanate Methyl (0.1%) and Carbendazim (12%) + Mancozeb (0.2 %) showed 92.22 and 86.67 percent inhibition, respectively. Whereas Mancozeb (0.25%), with 31.11 percent inhibition, proved less effective than the rest of the fungicides.

INTRODUCTION

Sapota (*Manilkara achras* (Mill.) Forseberg) is referred to as "Chiku" (Phalke, 2009). It is a member of the Sapotaceae family (Ghirtlahre, 2015). It is a tropical American native and has expanded to other nations such as the Philippines, Malaysia, The United States of America, Sri Lanka, and India, where it was adopted very well. The first planting of this crop in India took place in the village of Gholwad in the Palghar District of Maharashtra state in 1898 (Chundawat, 1998). Today, this crop is grown in numerous parts of the nation, including Tamil Nadu in the south, Punjab region in the north, West Bengal in the east, and Rajasthan state in the west, with Tripura as well as Assam within north-eastern provinces, as well as the Andaman and Nicobar Islands. Its better ability to adapt to a variety of agro-climatic conditions and its capacity for continuous growth may be the causes of the plant's rapid proliferation across the nation.

India produces 10% of the world's fruit, and it leads nations worldwide in sapota production (APEDA Database, 2020). NHB Database reports that India produced 826.92 metric tonnes in 2020–21. Gujarat leads India in output, with 273.87 metric tonnes produced there in 2021–2022 (NHB, 2021). The Sapota region of Maharashtra spanned 70,000 hectares and produced 322 thousand tonnes with an average yield of 4.6 tonnes per hectare (Surwase *et al.*, 2015). Sapota is an evergreen fruit tree with a significant position among fruit crops. The fruit is primarily eaten in its fresh state and is very savory, sweet, and mildly astringent. It contains about 12 to 14 % sugar and is a good carbohydrate source. In addition to table usage, processed sapota fruit products include drinks, jams, sweet chutneys, pickles, and dehydrated slices. Sapota bark produces

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white resin latex (Warrington, 2021) that is used to make "chickle," a key ingredient in the production of chewing gums with a significant export market. The gum obtained from sapota latex can be utilized in dental surgery. The bark of the tree contains tannin, which has many industrial benefits. Economical amounts of liquid fat that can produce consumable oil and feed cake are present in the seed kernel. The fruit of Sapota has been used as a conventional indigenous medicine in many different cultures .

MATERIALS AND METHODS

Materials- Infected sapota leaves were collected from the College of Agriculture, Pune. For in vitro experiments, fungicides were obtained from the local market of Pune, India.

Isolation of *Colletotrichum gloeosporioides*

C. gloeosporioides was isolated from the infected portions of the sapota leaves. The Potato Dextrose Agar (PDA) medium was used to grow small pieces of the diseased leaves, which were then incubated at a temperature of 28 to 30 °C. Identified based on conidia as well as other morphological traits published according to

In vitro evaluation of fungicides against *Colletotrichum gloeosporioides*

In Table 1, the fungicides used in the current examination were included with their trade names, chemical ingredients, and sources of supply.

The following fungicides' effectiveness was evaluated using the potato dextrose agar (PDA) medium and the poisoned food technique. By using the method proposed by Vincent (1947), the efficiency of a fungicide was evaluated by calculating the percent suppression of mycelial growth over control.

Where,

I – Fungal growth inhibition in percent

C – Pathogen growth/colony diameter in control plate (cm)

T – Pathogen growth/colony diameter of the pathogen in treatment plate (cm)

Poisoned food technique

To obtain the needed concentration of fungicides, the necessary quantity of each fungicide was added to molten and cooled potato dextrose agar. After that, 20 ml of the poisoned medium was added to sterilized plates and allowed to harden. A sterile cork borer was used to cut out mycelial circular discs of 5 mm size from an actively developing culture of the fungus, and one circular disc was then kept in the middle of every agar plate. No fungicide was added to the medium of the control plate. Three replications of each treatment were performed. Radial fungal colony growth was assessed after an incubation period of eight days at a temperature of 27±2 °C.

RESULTS AND DISCUSSION

Assesment of fungicides against *Colletotrichum gloeosporioides In vitro* condition- Six different fungicides were investigated for effectiveness in this study, including Mancozeb (75% WP), Carbendazim (12%) + Mancozeb (63%) WP, Copper Oxychloride (50% WP),

Thiophanate Methyl (70% WP), Difenoconazole (25% EC), and Propiconazole (25% EC). The information gathered on the impact of various fungicides on mycelial growth and the percentage inhibition of *C. gloeosporioides* is shown in Figure 1, Table 2, and Plate 1.

Radial mycelial growth

From data, it was revealed that no mycelial growth was observed in the Petri plates where media was

Table 1. Treatment details of fungicides

Trial No.	Fungicides	Trade name	Active ingredients	Concentration (%)
T_1	Carbendazim+ Mancozeb	Saaf	12% + 63% WP	0.2
T_2	Mancozeb	M-45	75% WP	0.2
T_3	Copper Oxychloride	Tata Blitox	50% WP	0.2
T_4	Thiophanate Methyl	Roko	70% WP	0.1
T_5	Difenoconazole	Score	25% EC	0.1
T_6	Propiconazole	Tilt	25% EC	0.1

WP= Wettable powder, EC= Emulsifiable concentrate

Table 2. Assessment of fungicides against the *Colletotrichum gloeosporioides* Penz. *In vitro* condition

Trial No.	Fungicides	Concentration (%)	Mean colony diameter of pathogen (cm)	Percent inhibition over control
T_1	Carbendazim (12%) + Mancozeb (63%) WP	0.2	0.70	92.22
T_2	Mancozeb (75%WP)	0.25	6.20	31.11
T_3	Copper Oxychloride (50%WP)	0.2	0.00	100.00
T_4	Thiophanate Methyl (70%WP)	0.1	1.20	86.67
T_5	Difenconazole (25%EC)	0.1	0.00	100.00
T_6	Propiconazole(25%EC)	0.1	0.00	100.00
T_7	Absolute Control	—	9.00	0.00
	SE(m) ±		0.07	
	CD at 1%		0.28	

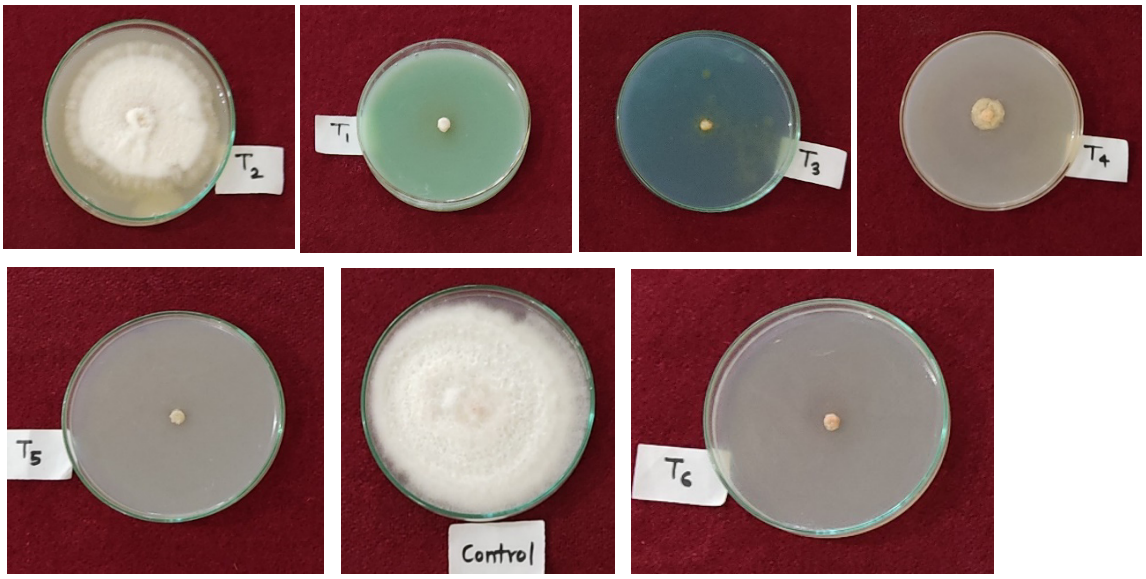

Plate 1. Impact of various fungicides with recommended concentration against *Colletotrichum gloeosporioides*

poisoned with copper oxychloride, Difenoconazole, and Propiconazole whereas, fungicides Carbendazim + Mancozeb and Thiophanate Methyl showed the least mycelial growth with 0.70 and 1.20 cm respectively. Maximum mycelial growth (6.20 cm) was recorded in media that was poisoned with Mancozeb.

Percent inhibition over control

The pathogen's growth in Petri plates was significantly suppressed by all of the tested fungicides, according to data comparing inhibition to control. Copper oxychloride, Propiconazole, and Difenoconazole, completely (100%) inhibited the growth of *C. gloeosporioides* over control. These fungicides were followed by Carbendazim + Mancozeb with 92.22 percent, Thiophanate Methyl with 86.67 percent, whereas Mancozeb with only 31.11 percent inhibition was proved less effective as compared to the rest of the fungicides.

All the fungicides were found effective against test fungi the observations of the current investigation conform with reports of Rajesh *et al*. (2010) and Kolase *et al.* (2014) who also stated that *Colletotrichum gloeosporioide's* mycelial growth has been demonstrated to be effectively inhibited by 0.1% carbendazim. Similar to this, Rathva *et al.* (2017) found that *Colletotrichum gloeosporioides* was extremely toxic to carbendazim + mancozeb and propiconazole at 1000 and 500 ppm, completely inhibiting fungus development. Somashekhara and Vani (2018) noticed that Propiconazole, Difenoconazole, and Tebuconazole were determined to be the most efficient in the inhibition of fungal growth in laboratory conditions. Ranjitha *et al*. (2019) reported that systemic fungicides

Table 3. Details of Plates

Trialno.	Name of treatment	Trial no.	Name of treatment
T_1	Carbendazim (12%) + Mancozeb (63%) WP	T_5	Difenconazole (25%EC)
T_2	Mancozeb (75%WP)	T_6	Propiconazole (25%EC)
T_3	Copper Oxychloride (50%WP)	T_7	Absolute Control
T_4	Thiophanate Methyl (70%WP)		-

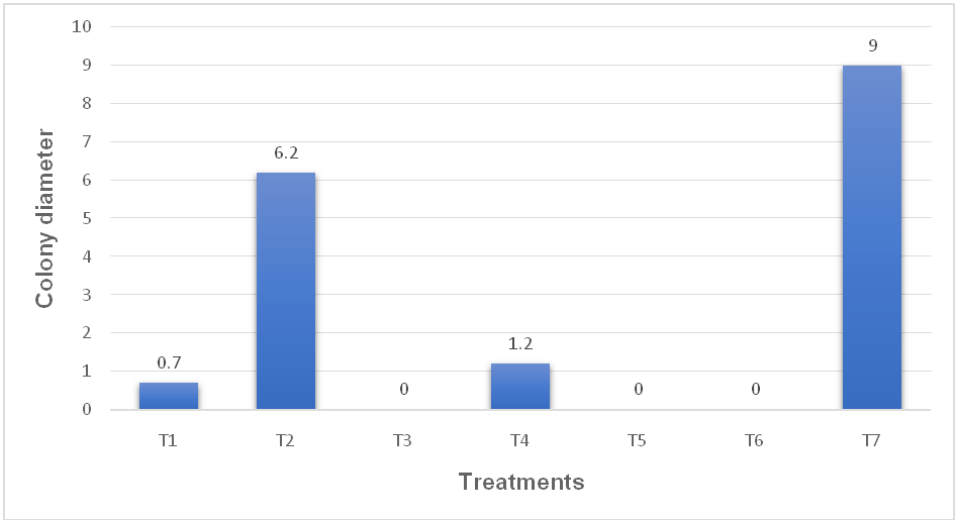

Fig. 1. *In vitro* impact of various fungicides on *Colletotrichum gloeosporioides* Penz.mycelial growth

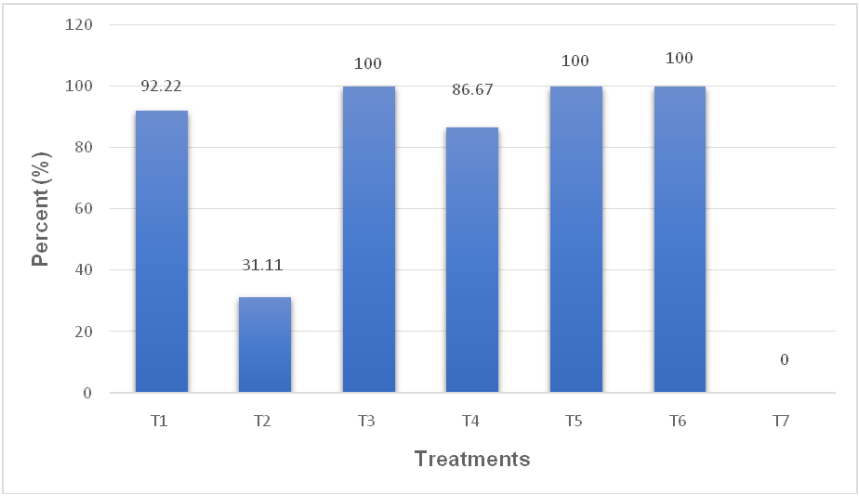

Fig. 2. Percent growth inhibition of *Colletotrichum gloeosporioides* different fungicides by different fungicides

Difenoconazole, Tebuconazole + Trifloxystrobin, and Propiconazole showed effective against *Colletotrichum gloeosporioides* by reducing its mycelial growth on media.

CONCLUSION

Fungicide Copper Oxychloride (0.2%) was found to be most effective, followed by Difenoconazole (0.1%) and Propiconazole (0.1%) which showed 100% inhibition over control.

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Declaration

The authors claim that there are no conflicting interests.

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