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***In vitro* evaluation of fungicides against *Colletotrichum gloeosporioides* Penz. and sacc. Causing anthracnose in pointed gourd**

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Abstract

Use of fungicide is a common practice to control the diseases, so, the present investigation was carried out with isolation of the organism responsible for anthracnose to suggest suitable control measure in respect to fungicides in controlling anthracnose of pointed gourd caused by *Colletotrichum gloeosporioides* Penz and Sacc. Under *in vitro* studies thirteen fungicides at three different concentrations were screened by poisoned food technique for evaluating their efficacy. Among them Carbendazim + mancozeb (Sixer 75% WP) and propiconazole (Tilt 25% EC) at 500 and 1000 ppm, and hexaconazole (Contaf 5% EC) at 1000 ppm completely inhibited growth of the fungus and proved to be highly toxic to the growth of the *Colletotrichum gloeosporioides*.

Keywords: Fungicides, *Colletotrichum gloeosporioides*, Anthracnose, Pointed gourd, *in vitro*

Introduction

The Pointed gourd (*Trichosanthes dioica* Roxb.) commonly known as *Patal*, *Potol*, *Parwar*, *Parwal* or *Parval* is believed to be native of India and Assam is supposed to be the place of its origin. It is called "King of gourds" because of its higher nutrient content than other cucurbits (Mondal *et al.*, 2014) [2]. It belongs to family Cucurbitaceae. It is commonly climbing and perennial vegetable, most nutritive, easily digestible, diuretic, laxative and invigorates the heart and brain and useful in circulatory system.

Anthracnose disease caused by *Colletotrichum gloeosporioides* Penz. and Sacc. is one of the most important disease of pointed gourd in South Gujarat Region. The disease produce light yellowish, minute pin head sized, round to irregular spots appeared on the margins of the leaves, later they gradually increase in size and coalesce to each other and turn light brown in colour and were surrounded by yellow halo. In advanced stages, these spots spread to whole leaves and giving an appearance like necrotic lesions resulted in to withering and shedding of leaves. Finally, the affected leaves dried up and fall prematurely. There was very less scientific information available about anthracnose in pointed gourd and considering the seriousness of the disease, the present studies were undertaken.

Materials and Methods

The present study was carried out at Department of Plant Pathology at ASPEE College of Horticulture and Forestry, NAU, Navsari.

Isolation of pathogen

Infected leaves of pointed gourd were used to isolate the pathogen. The infected area was subjected to tissue isolation. The infected portion of the leaves was cut into small pieces in such a way that each piece consisted of infected as well as healthy tissues. The pieces were surface sterilized with 0.1 per cent mercuric chloride (HgCl₂) solution for 30 seconds followed by subsequent three washings with sterilized distilled water and then transferred aseptically under laminar air flow system on sterilized Petri plates containing 20 ml Potato Dextrose Agar (PDA) medium. The Petri plates were incubated at room temperature (28 ± 1 °C) and periodically observed for the growth. The fungal hyphae developing from the infected tissues were sub-cultured aseptically on PDA slants. The pure culture thus obtained was microscopically examined for identification and was further purified by using single spore isolation technique.

The culture obtained was maintained on PDA slants at low temperature for further investigations. Media used in this study were sterilized in an autoclave at 1.2 kg/cm² pressure for 20 minutes (Aneja, 2002).

***In vitro* evaluation of fungicides**

The fungicides were incorporated aseptically in the molten PDA in required quantities separately before pouring and shaken well for uniform dispersal of the fungicide. The medium was then poured in the Petri plates and on solidification of the medium, the plates were aseptically inoculated by placing 5 mm diameter culture disc in the centre by cutting the periphery of 8 days old pure culture of *Colletotrichum gloeosporioides* grown on PDA. The plates were incubated at room temperature (27 + 2 °C). The colony diameter of the fungus was recorded from three repetitions after 10 days. The plates without fungicides served as control. The per cent growth inhibition over control was worked out by using the formula given by Vincent (1927)^[6].

$$\text{PGI} = \frac{(\text{DC} - \text{DT})}{\text{DC}} \times 100$$

Where,

PGI = Per cent growth inhibition

DC = Average diameter of mycelial colony of control set (mm)

DT = Average diameter of mycelial colony of treated set (mm)

Results and Discussion

The results presented in Table 1 revealed that all the fungicides significantly reduced the growth of *C. gloeosporioides* as compared to control but all the fungicides and their concentrations significantly differed within themselves. Among all concentrations, the higher concentration of each fungicides produce maximum growth inhibition of the *C. gloeosporioides*. Out of all the fungicides tested, carbendazim + mancozeb (Sixer 75% WP) and propiconazole (Tilt 25% EC) at two concentrations viz., 500 and 1000 ppm, hexaconazole (Contaf 5% EC) at one concentration i.e., 1000 ppm completely inhibited growth of the *C. gloeosporioides* and proved highly fungitoxic at above mentioned concentrations.

Table 1: Evaluation of different fungicides against *C. gloeosporioides* *in vitro*.

Treatments	Technical name	Concentration (ppm)	Average colony diameter (mm)	Growth inhibition (%)
T1	Mancozeb	1500	5.6*(30.67)	65.79
		2000	4.9(23.67)	73.60
		2500	4.5(19.67)	78.06
T2	Copper hydroxide	1500	6.0(35.00)	60.96
		2000	4.6(20.33)	77.32
		2500	3.4(11.33)	87.36
T3	Propineb	1500	7.2(51.67)	42.37
		2000	6.5(42.00)	53.15
		2500	5.5(29.67)	66.91
T4	Chlorothalonil	1500	6.2(37.67)	57.99
		2000	5.6(31.00)	65.42
		2500	4.9(23.33)	73.97
T5	Carbendazim + Mancozeb	250	1.5(1.67)	98.14
		500	0.7(0.00)	100
		1000	0.7(0.00)	100
T6	Propiconazole	250	3.2(9.67)	89.21
		500	0.7(0.00)	100
		1000	0.7(0.00)	100
T7	Difenoconazole	250	3.1(9.33)	89.59
		500	2.9(7.67)	91.44
		1000	2.5(6.00)	93.30
T8	Hexaconazole	250	3.1(9.33)	89.59
		500	2.1(4.00)	95.53
		1000	0.7(0.00)	100
T9	Tebuconazole	250	6.0(35.33)	60.59
		500	5.5(29.67)	66.91
		1000	5.0(25.00)	72.11
T10	Benzimidazole	250	7.3(52.67)	41.26
		500	6.5(41.67)	53.53
		1000	6.0(35.33)	60.59
T11	Metalaxyl + Mancozeb	500	8.7(75.67)	15.61
		1000	8.0(64.00)	28.62
		1500	6.5(41.67)	53.53
T12	Cymoxanil +Mancozeb	250	6.5(41.67)	53.53
		500	5.8(32.67)	63.56
		1000	4.9(24.00)	73.23
T13	Pyraclostrobin 5% + Metiram 55% WG	250	7.8(59.67)	33.45
		500	7.4(53.67)	40.14
		1000	6.4(41.00)	54.27
T14	Control		9.5(89.67)	0
	S.Em. ±		0.11	
	C.D. at 5%		0.30	
	C.V.%		3.86	

These results are similar with the results obtained by earlier workers. Ramani *et al.* (2015) [5] observed that carbendazim + mancozeb completely inhibited the growth of *C. gloeosporioides* causing anthracnose in banana at all the concentrations *i.e.* 100, 250, 500 and 1000 ppm. Patel (2004) [4] observed that propiconazole, carbendazim and hexaconazole were proved strongly fungitoxic against dieback and fruit rot (*Colletotrichum gloeosporioides*) of chilli at all the concentrations *i.e.*, 250, 500 and 1000 ppm. Patel (2000) [3] observed cent per cent inhibition of growth of turmeric leaf spot pathogen *C. gloeosporioides in vitro* by carbendazim, propiconazole and hexaconazole at all the concentrations.

Conclusion

Thirteen fungicides were evaluated at three different concentrations by poisoned food technique for evaluating their efficacy against *C. gloeosporioides*. Among them, carbendazim + mancozeb (Sixer 75% WP), propiconazole (Tilt 25% EC) at 250, 500, 1000 ppm showed maximum inhibition of growth of the *C. gloeosporioides*. Next best was hexaconazole (Contaf 5% EC) at all three concentrations *viz.*, 250, 500, 1000 ppm inhibited cent per cent growth of the pathogen and proved strongly fungitoxic.

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