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In vitro efficacy of different fungicides against *Fusarium solani* isolate causing root rot of papaya (*Carica papaya* L)

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Abstract

Fusarium solani considered as an aggressive pathogen widely dispersed in soil and causes root rot disease in papaya plant which is responsible for the estimated loss up to 40-95 per cent. In the present study, six fungicides at different concentration viz; 50, 100 and 150 ppm were evaluated against five isolates of *Fusarium solani* (Fs-I to Fs-V) causing papaya root rot by applying poisoned food technique in the laboratory of the Department of Plant Pathology, DRPCA, Pusa, Samastipur in 2016. Observation of the experiment was taken at 240 hours for the assessment of their inhibitory effect. The data revealed that all the fungicide (contact and systemic) significantly reduces the mycelial growth of *Fusarium solani* isolate over control but Carbendazin 50% WP @150 ppm found effective with Fs-I (95.3%), Fs-II (94.9%), Fs-III (95.1%), Fs-IV (94.9%) and Fs-V (95.5%) mycelial growth inhibition over control followed by Propiconazole, Hexaconazole, Mancozeb, Copper Oxochloride. However, Azoxystrobin shows minimum inhibition per cent against all isolate Fs-I (75.0%), Fs-II (70.3%), Fs-III (74.5%), Fs-IV (71.5%) and Fs-V (74.4%) over control respectively.

Keywords: Fungicides, *Fusarium solani* isolate, root rot, papaya

Introduction

Papaya (*Carica papaya* L.) “Wonder fruit of the tropics “belonging to family Caricaceae. The genus *Carica* contains 21 species, out of which 5 species, *C. papaya*, *C. chilensis*, *C. goudotiana*, *C. monoica* and *C. pubescens* produce edible fruits. Papaya, a very wholesome fruit, is high in nutritive and medicinal value. Owing to increasing demand for its delicious fruit taste and extraction of its digestive constituent papain. Due to its high returns income, the area and production of papaya have increased next to banana during the last few decades. India is the largest producer of papaya in the world. The present area under this fruit crop is about 138 (000) ha and production is 5989 (000MT) with 42.3tonne/ha productivity. Maharashtra is the leading state of papaya production where it is grown mainly for papain extraction. Other states are Karnataka, Madhya Pradesh, Uttar Pradesh, Bihar, Gujrat, West Bengal, Tamilnadu, Andhra Pradesh, Kerala and Assam. In Bihar, its production was 42.72 (000 MT) from an area of 1.90 (000) ha (Horticulture statistic Division, DAC & FW, 2017-18) [5]. Diseases are a major limiting factor in the cultivation of papaya. Root rot disease of papaya caused by *Phytophthora nicotianae* was first reported in 1998 by Robert and Trujillo [12] and subsequently, Rodeiguez *et al.* (2001) [13] reported that the disease was incited by *P. aphanidermatum* from Tabasco, Mexico and the United States. But over the last four years; a new disease of papaya with a symptom of root rot has emerged as a serious threat to the crop in Bihar causing 90-95 per cent crop failure and resulted in gradual collapse of entire papaya plantation which inflicting a heavy loss to the growers. All the varieties are highly susceptible to this disease. The disease has been found to affect the crop round the year at all growth stages of the plants. However, development of disease becomes fast after rain occurring in any month. Singh and Kumar (2015) [14] first time established etiology of Papaya root rot in agro-ecological conditions of Bihar and reported that the root rot was caused by *Fusarium solani* (Mart.) Sacc. Due to its devastating nature, there is an urgent need to focus on remedies for its management. Hence present study investigates the *in-vitro* efficacy of different fungicides against *Fusarium solani* isolate causing root rot of papaya (*Carica papaya* L.).

Material and Methods

The standard laboratory techniques were used for the preparation of PDA media, cleaning and sterilization of glassware's, isolation of fungus, inoculation and maintenance of fungal culture, with modification whenever necessary.

Collection of disease specimen

During the experiment work, the roving survey was conducted in different districts of Bihar viz. Samastipur, Muzaffarpur, Vaishali, Begusarai, Saran, Supoul, Bhagalpur, Aurangabad, Rohtas and Nalanda to record the incidence of papaya root rot disease in major papaya growing areas. In each district minimum, five orchards with 40 plants /orchard were surveyed. Disease specimen of papaya showing characteristic symptoms of root rot were collected from the experimental plot of Dr. Rajendra Prasad Central Agricultural University, Pusa, and other surveyed districts of Bihar. The plant showed characteristic symptoms of root rot were brought to the laboratory of the Department of Plant Pathology, washed with running tap water to remove dust and dirt and then kept in the refrigerator for further study.

Isolation of *Fusarium Solani* isolates

For isolation, diseased specimen/disease plant part (root) of papaya showing characteristic symptoms of root rot were washed thoroughly in several changes of tap water for 5 minutes to remove dirt and were cut into small bits of 2-3mm dimension by tissue segmentation method (Bonmam *et al.*, 1987) [1]. These bits were surface sterilized by dipping in 0.1 per cent mercuric chloride solution for 30 seconds followed by washing in 2 changes of sterilized water, then placed aseptically on PDA slants with the help of inoculating needle under aseptic condition. These were incubated at 28±2 °C. After 4 days of incubation, the fungus was transferred to sterilize Petri-plates containing PDA medium and incubated in the same manner (Hemalatha *et al.*, 2018) [4]. After 6 days of incubation, a bit of hyphal growth from growing tips was transferred aseptically to fresh PDA slants. The fungus was purified by employing the single hyphal tip method (Singh, 1988) [15]. The fungus was identified following a mycological description (Ou, 1985) [11]. Further, the isolates were characterized/grouped in five different groups (Fs-I, Fs-II, Fs-III, Fs-IV, Fs-V) based on growth pattern and pathogenicity. However, the culture is maintained by periodically transfer on PDA slants for further studies.

Evaluation of fungicides against *Fusarium solani* in vitro

Six fungicides were evaluated *in vitro* against different isolates of *Fusarium solani* using poison food technique (Nene and Thapliyal, 1982) [10]. Their trade name, chemical name, formulation, chemical group and mode of action are given in (Table.1). A hundred ml sterilized Potato dextrose agar medium was fortified with 5, 10 and 15 mg of six fungicides separately to get 50 ppm, 100 ppm and 150 ppm concentrations. 20ml of media of each concentration was gently poured in sterilized Petri plate and allowed them to solidify. After solidification, 5 mm disc of seven days old culture of *Fusarium solani* isolates were cut with the help of cork borer and then placed in the centre of the Petri plates and incubated in B.O.D at 28 ± 2 °C. There will be thrice replication of each treatment along with control plate (without fungicide) in the medium for a more accurate result. After incubation, the radial growth of *Fusarium solani* isolates was measured with the help of ruler scale after 24 hrs and subsequent observation was recorded at 48 hrs of interval and

continued till full growth of the pathogen in control (90mm) i.e. 240 hrs. The per cent inhibition of fungus over control was calculated by using following formula as given by Vincent (1927) [16].

$$I = \left(\frac{C - T}{C} \right) \times 100$$

Where,

I = Percent growth inhibition = Colony diameter in control Petri plate;

T = Colony diameter in the treated Petri plate. The per cent inhibition data were analyzed statistically using completely randomized design (C.R.D)

Table 1: Fungicides details used against *Fusarium solani* isolates causal agent of root rot of papaya

Trade name	Chemical name	Formulation	Chemical group	Mode of action
Bavistin	Carbendazim	50% WP	Benzimidazole	Systemic
Tilt	Propiconazole	25% WP	Phenylamide	Systemic
Dithane M-45	Mancozeb	80% WP	Dithiocarbamate	Contact
Blitox-50	Copper oxychloride	50% WP	Copper compound	Contact
Contaf	Hexaconazole	75% WP	Copper compound	Contact & systemic
Amistar	Azoxystrobin	250 SC	Strobilurin	Systemic

Data analysis

Data were statistically analyzed using statistical analysis software (SAS) packages. Critical differences were calculated at 5% level of significance for comparison of treatment mean. The Microsoft Excel (2010) computer software package was used to prepare all the graphs.

Result and Discussion

The growth inhibition of *Fusarium solani* isolates (Fs-I, Fs-II, Fs-III, Fs-IV and Fs-V) the causing root rot in papaya has been tested at various concentrations of fungicides under *in-vitro* condition was recorded in table 2. The radial growth of *Fusarium solani* isolates was measured after 24 hrs of inoculation and subsequent observation was recorded at three days interval till full growth of the pathogen in control i.e. 240 hrs and per cent inhibition was calculated based on final observation. The perusal of result showed that (Table. 2) (Fig.1) among all the fungicides tested, Carbendazim 50% WP @150 ppm was found effective in inhibiting mycelial growth of *Fusarium solani* isolate by 95.3% in Fs-I, Fs-II (94.9%), Fs-III (95.1%), Fs-IV (94.95) and Fs-V (93.5%) followed by Propiconazole that inhibits mycelium growth of Fs-I (91.9%), Fs-II (91.3%), Fs-III (91.9%), Fs-IV (92.2%) and isolate Fs-V (92.3) while Hexaconazole, in the same manner, inhibits the mycelium growth by 91.7% in Fs-I and isolate Fs-II shows (91.6%), Fs-III (91.6%), Fs-IV (92.2%) and in isolate Fs-V 92.0 per cent over control respectively. Mancozeb, COC (Copper oxychloride) and Azoxystrobin were found less effective against all the isolates of *Fusarium solani* at 50, 100 and 150 ppm concentration over the control. The inhibitory effect of the test fungicides was increased with increasing fungicide concentrations. This finding is also consonance with the results of Kapratwar *et al.*, (2016) [6] studied the effect of Carbendazim against *Fusarium solani* causing rhizome rot of ginger at five concentrations such as 0.025, 0.05, 0.1, 0.15 and 0.2% respectively. Among all the concentration Carbendazim at 0.2% was considered as more

effective in disease control. The seven fungicides were screened against *Fusarium solani* causal agent of root rot of papaya and identified Carbendazim as a most effective fungicide, inhibited 93.5% mycelia growth followed by Thiophanate – methyl (89.7%) at 150 ppm was observed by Kumar and Singh (2015) [7]. Amongst all Copper oxychloride fungicide was found the most effective (85.05% inhibition) against *Fusarium solani* followed by Copper hydroxide (80.1%) and Hexaconazole (68.2%) at recommended dose respectively reported by Kumhar *et al.*, (2015) [8]. The effect of fungicide was evaluated against ten different isolates of *Fusarium solani* causing elephant foot yam and found that isolate fs-9 from Bidoor was more sensitive against Benomyl (25 ppm) on PDA medium while isolate fs-3 was highly resistant (100 ppm) by Dorugade *et al.*, (2015) [3]. Choudhari *et al.*, (2012) [2] conducted an *in-vitro* screening of five different fungicides viz; Carbendazim, Captan, Dithan-M45, Thiophanale methyle and Thiram against the fungus *Fusarium solani* and observed that among all Carbendazim completely inhibited the growth of pathogen at all three concentration ie.100,250 and 500 ppm respectively. Muneeb

et al., (2011) [9] conducted an experiment in which different isolates of fungus viz; *Fusarium oxysporum* f. sp. *ciceri*, *F. solani* and *Rhizoctonia solani* were isolated from the wilt infected chickpea (*Cicer arietinum*) plants. They also recorded that when fungicide Carbendazim @ 100, 200, 500 ppm concentrations applied it's caused the highest per cent inhibition of the pathogens under *in vitro* conditions. Fungicides when applied as a seed treatment, significantly reduced disease incidence up to some extent. However, it was also observed that seed treatment with Carbendazim found effective in increasing seed germination up to 71.24%, though it was at par with Carbendazim + Mancozeb (62.21%) and Mancozeb (61.46%). The yield increases spontaneously up to 10.10q/ha with the application of Carbendazim, followed by Carbendazim + Mancozeb (9.77 q/ha). The present results indicate that Carbendazim, followed by Propiconazole, Hexaconazole, Mancozeb, COC and Azoxystobin was quite effective in controlling papaya root rot pathogen. Such information will be helpful in the formulation of schedule for management of the disease.

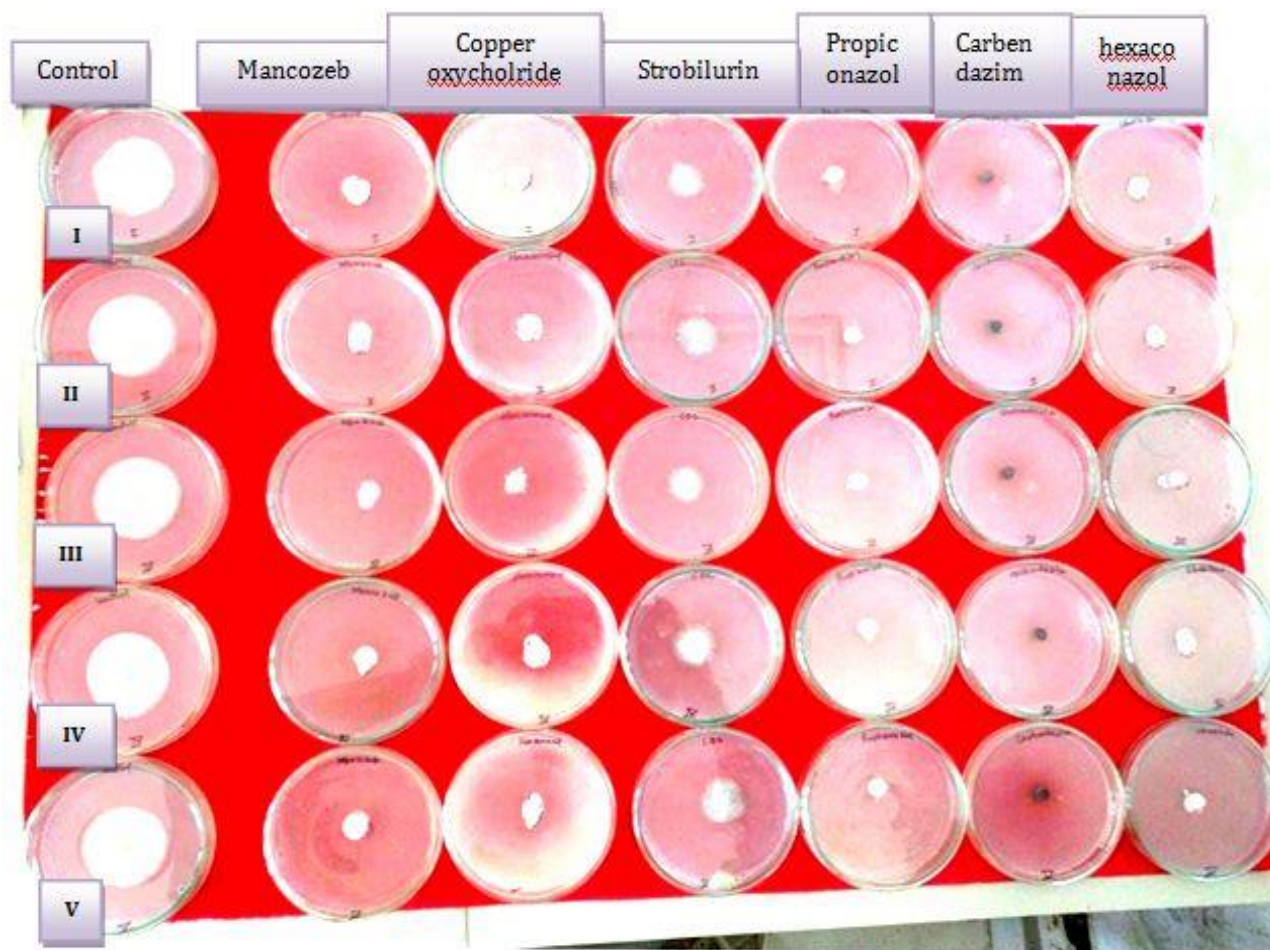


Fig 1: Effect of different fungicides at 150 ppm concentration on radial growth of *Fusarium solani* isolates on PDA

Conclusion

From the foregoing result it can be concluded that the tested fungicides significantly inhibited the root rot disease caused by *Fusarium solani*. Among all the six fungicides, Carbendazim was found quite effective in cent per cent inhibition of fungus mycelial growth followed by Propiconazole, Hexaconazole, Mancozeb, and COC over

control. Azoxystrobin showed least effective against the pathogen based on antifungal efficiency.

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Table 2: *In-vitro* efficacy of different fungicides against different isolates of *Fusarium solani*

Treatments	Concentration (ppm)	*Radial growth (mm) of <i>Fusarium solani</i> isolates									
		Fs-I		Fs-II		Fs-III		Fs-IV		Fs-V	
		240 hrs	Inhibition over control (%)	240 hrs	Inhibition over control (%)	240 hrs	Inhibition over control (%)	240 hrs	Inhibition over control (%)	240 hrs	Inhibition over control (%)
Carbendazim	50	9.8	88.6	11.3	87.1	9.8	88.7	9.9	88.6	9.6	88.7
	100	6.8	92.2	7.8	91.0	6.3	92.7	6.7	92.3	6.8	92.0
	150	4.0	95.3	4.8	94.9	4.2	95.1	4.4	94.9	3.8	95.5
Copper Oxy Chloride	50	30.0	65.1	31.7	63.8	29.9	65.4	31.8	63.5	28.5	66.7
	100	26.1	69.6	28.2	67.8	26.1	69.8	26.7	69.3	25.1	70.6
	150	20.6	76.0	23.4	73.2	21.2	75.4	22	74.7	21.0	75.4
Azoxystrobin	50	35.0	59.3	38	56.6	35.2	59.3	36.4	58.2	34.7	59.4
	100	28.8	66.5	33.4	61.8	29.1	66.3	32	63.3	28.5	66.7
	150	21.5	75.0	26	70.3	22.0	74.5	24.8	71.5	21.9	74.4
Mancozeb	50	20.9	75.6	21.8	75.5	19.8	77.1	21.8	75.0	19.7	76.9
	100	17.5	79.6	18.4	78.9	17.5	79.7	17.9	79.4	17.0	80.1
	150	13.5	84.3	14.5	83.3	13.7	84.1	13.8	84.1	13.6	84.1
Propiconazole	50	13.0	84.8	14.3	83.6	13.0	84.9	14.0	83.9	12.9	84.9
	100	8.8	90.2	10.1	88.4	8.9	89.7	8.8	89.9	9.0	89.4
	150	7.0	91.9	7.4	91.3	7.0	91.9	6.8	92.2	6.8	92.4
Hexaconazole	50	13.2	84.6	14.5	83.4	13.2	84.7	14.5	83.3	13.4	84.3
	100	8.9	89.7	10.3	88.2	8.9	89.7	9.2	89.4	8.9	89.6
	150	6.9	91.7	7.6	91.6	7.2	91.6	6.8	92.2	6.5	92.0
Control	-	86.0	-	87.6	-	86.5	-	87.2	-	85.6	-
CD at 5%		1.90	3.16	1.38	3.49	1.38	3.89	1.51	3.81	1.47	3.30
SE(m)±		0.66	1.09	0.48	1.21	0.48	1.35	0.52	1.31	0.51	1.14
CV (%)		4.68	2.34	3.86	2.64	4.18	2.88	4.37	2.84	4.52	2.44

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