#### **Research Article**



# Compatibility of Trichoderma isolates with selected fungicides in vitro

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# ARITCLE INFO Article Chronicle : Received : 31.08.2011

**Received :** 31.08.2011 **Revised :** 18.10.2011 **Accepted :** 07.01.2012

Key words : Fungicides, Evaluation, Compatibility, Trichoderma

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# ABSTRACT

Eighteen selected fungicides were evaluated for their compatibility to *Trichoderma* based on *in vitro* senstitivity of *T. harzianum* and *T. virens*. Observations on radial growth indicated that, carbendazim, benomyl, carboxin, propiconazole, hexaconazole, tricyclozole, tridemorph, chlorothalonil were incompatible with *Trichoderma* spp. showing 100 per cent inhibition of radial growth at field concentration. While dinocap, copperoxy chloride, fosetyl-Al captan, thiram and metalaxyl were found to be least compatible showing more than 70 per cent inhibition of radial growth. Bordeaux mixture, azoxystrobin and mancozeb were moderately compatible with radial growth inhibition in the range of 20-45 per cent. Only wettable sulphur was found to be highly compatible with least inhibition of radial growth (2.2%) of test *Trichoderma* isolates.

*How to view point the article*: Ranganathswamy, M., Patibanda, A.K., Chandrashekhar, G.S., Sandeep, D., Mallesh, S.B. and Halesh Kumar, H.B. (2012). Compatibility of *Trichoderma* isolates with selected fungicides *in vitro*. *Internat. J. Plant Protec.*, **5**(1): 12-15.

# INTRODUCTION

Soil borne plant pathogenic fungi such as *Fusarium*, *Phytophthora*, *Pythium*, *Rhizoctonia*, *Sclerotium etc.* cause diseases in most of the economically important crop plants. Chemical means of managing the diseases caused by these pathogens are not practicable owing to high cost of chemicals and environmental pollution. Biological control offers a novel approach when applied either alone or in combination with other management practices without the demerits of chemical control (Papavizas, 1985 and Mukhopadhyay, 1987). *Trichoderma* is one of the most common soil inhabitants and extensively studied biocontrol agent in the management of soil borne plant pathogens (Elad *et al.*, 1980).

Species of *Trichoderma* are being used either as seed treatment or soil application. In both the cases, the antagonist has been continuously exposed to different fungicides applied to the field either in soil or as foliar sprays. Fungicides sprayed aerially reaches the soil (by means of air currents or are washed off the plant surface due to rain) and is likely to influence the efficacy of native or applied biocontrol agents like

*Trichoderma*. Hence, it is necessary to assess *Trichoderma* compatibility to fungicides in order to use in the Integrated Disease Management systems (Singh *et al.*, 1995). Variations in tolerance of *Trichoderma* isolates to several fungicides reported earlier (Pandey and Upadhyay, 1998; Vijayaraghavan and Koshy, 2004) were based on arbitrary concentrations that were less than the field concentrations. Hence, the present investigation was conducted to evaluate the compatibility of two isolates of *Trichoderma* spp. *viz.*, *T. harzianum* (isolated from cotton cropping system) and *T. virens* (isolated from citrus orchard) to selected fungicides at field concentration.

#### **MATERIALS AND METHODS**

In the present investigation, eight contact fungicides, *viz.*, copper oxychloride, Bordeaux mixture, wettable sulphur, mancozeb, thiram, captan, chlorothalonil and dinocap and ten selective systemic fungicides, *viz.*, carbendazim, benomyl, carboxin, metalaxyl, propiconazole, hexaconazole, tricyclazole, tridemorph, fosetyl-Al and azoxystrobin, were used to assess the *in vitro* sensitivity of *Trichoderma* isolates by using the poisoned food technique (Nene and Thapliyal, 1993). Radial growth of the test *Trichoderma* isolates were recorded after 48 h of incubation and per cent inhibition of growth over control (unamended medium) was calculated using the following formula:

 $I = \frac{C - T}{C} x \ 100$ 

I – per cent inhibition C – growth in unamended medium T–growth in fungicide amended medium

## **RESULTS AND DISCUSSION**

Both the isolates of *Trichoderma*, *viz.*, *T. harzianum* and *T. virens* grew equally well and attained a radial growth of 3.7 cm and 9.0 cm after 24 and 48 h of incubation at  $28\pm1^{\circ}$ C, respectively on control PDA plates (Table 1).

In fungicide amended medium, all the fungicides showed inhibitory effect on radial growth. Variation was observed in the compatibility of *Trichoderma* isolates towards fungicides.

#### Compatibility of *Trichoderma* with fungicides:

Variation between isolates of Trichoderma: Observations

made on the radial growth of Trichoderma indicated significant variation in the sensitivity of Trichoderma isolates to fungicides or toxicity of fungicides towards Trichoderma isolates. When observations were recorded on radial growth for two consecutive days except in Bordeaux mixture, copper oxychloride, wettable sulphur, mancozeb, captan, fosetyl-Al, azoxystrobin and dinocap amended plates, in all other plates the growth was completely inhibited within 24 h after inoculation. Further, all the fungicides showed significant reduction in the growth of Trichoderma isolates on 1st and 2nd day of observations except in wettable sulphur where in the growth was on par with unamended control. When mean inhibitory per cent in the radial growth of Trichoderma isolates was analyzed, cotton isolate T. harzianum was found less sensitive (75.8 % inhibition) compared to citrus isolate T. virens (79.5%) (Table 2). This difference in per cent inhibition of radial growth was due to more sensitivity of T. virens to seven out of eight contact fungicides, viz., Bordeaux mixture, copper oxychloride, wettable sulphur, thiram, captan and dinocap and also to one systemic fungicide Fosetyl Al. Compared to T. virens, sensitivity of T. harzianum was higher only with respect to metalaxyl. Reports on inhibitory effect of different fungicides were reported by Sharma and Mishra

Table 1: Effect of fungicides on Trichoderma radial growth (cm)								
Sr. No.	Fungicides	Conc. (%)	T.harzianum		T.virens			
	i ungiolaes		Day1	Day2	Day1	Day2		
1.	Copper oxy chloride	0.3	$1.0(1.4)^{\rm f}$	2.5 (1.9) <sup>de</sup>	0.6 (1.3) <sup>g</sup>	$2.0(1.7)^{d}$		
2.	Bordeaux mixture	1	3.4 (2.1) <sup>c</sup>	7.8 (3.0) <sup>b</sup>	2.5 (1.9) <sup>c</sup>	6.5 (2.7) <sup>b</sup>		
3.	Wettable sulphur	0.2	4.1 (2.3) <sup>a</sup>	9.0 (3.2) <sup>a</sup>	3.3 (2.1) <sup>b</sup>	8.7 (3.1) <sup>a</sup>		
4.	Mancozeb	0.25	2.5 (1.9) <sup>d</sup>	6.2 (2.7) <sup>c</sup>	1.9 (1.7) <sup>e</sup>	4.1 (2.3) <sup>c</sup>		
5.	Thiram	0.25	$0.0 (1.0)^{h}$	1.0 (1.4) <sup>g</sup>	$0.0 (1.0)^{h}$	$0.0(1.0)^{\rm f}$		
6.	Captan	0.2	$0.6 (1.3)^{g}$	$1.8(1.7)^{\rm f}$	$0.6 (1.3)^{g}$	$1.4(1.5)^{e}$		
7.	Dinocap	0.1	1.4 (1.5) <sup>e</sup>	3.1 (2.0) <sup>d</sup>	$0.9(1.4)^{\rm f}$	$2.0(1.7)^{d}$		
8.	Chlorothalonil	0.2	$0.0 (1.0)^{h}$	$0.0 (1.0)^{h}$	$0.0 (1.0)^{h}$	$0.0 (1.0)^{\rm f}$		
9.	Carbendazim	0.1	$0.0 (1.0)^{h}$	$0.0 (1.0)^{h}$	$0.0 (1.0)^{h}$	$0.0(1.0)^{\rm f}$		
10.	Benomyl	0.1	$0.0 (1.0)^{h}$	$0.0 (1.0)^{h}$	$0.0 (1.0)^{h}$	$0.0 (1.0)^{\rm f}$		
11.	Carboxin	0.2	$0.0 (1.0)^{h}$	$0.0 (1.0)^{h}$	$0.0 (1.0)^{h}$	$0.0 (1.0)^{\rm f}$		
12.	Metalaxyl	0.2	$0.0 (1.0)^{h}$	$0.0 (1.0)^{h}$	$0.7 (1.3)^{g}$	1.1 (1.4) <sup>e</sup>		
13.	Propiconazole	0.1	$0.0 (1.0)^{h}$	$0.0 (1.0)^{h}$	$0.0 (1.0)^{h}$	$0.0 (1.0)^{\rm f}$		
14.	Hexaconazole	0.2	$0.0 (1.0)^{h}$	$0.0 (1.0)^{h}$	$0.0 (1.0)^{h}$	$0.0(1.0)^{\rm f}$		
15.	Tricyclazole	0.06	$0.0 (1.0)^{h}$	0.0 (1.0) <sup>h</sup>	$0.0 (1.0)^{h}$	$0.0(1.0)^{\rm f}$		
16.	Tridemorph	0.1	$0.0 (1.0)^{h}$	$0.0 (1.0)^{h}$	$0.0(1.0)^{h}$	$0.0(1.0)^{\rm f}$		
17.	Fosetyl-Al	0.15	$1.1 (1.4)^{\rm f}$	2.4 (1.8) <sup>ef</sup>	$0.8 (1.3)^{g}$	$1.4 (1.5)^{e}$		
18.	Azoxystrobin	0.1	2.5 (1.9) <sup>d</sup>	5.6 (2.6) <sup>c</sup>	2.2 (1.8) <sup>d</sup>	6.1 (2.7) <sup>b</sup>		
19.	Check		3.7 (2.2) <sup>b</sup>	9.0 (3.2) <sup>a</sup>	3.7 (2.2) <sup>a</sup>	9.0 (3.2) <sup>a</sup>		
	CV (%)		1.6	2.1	1.7	2.3		
	C.D. (P=0.01)		0.06	0.10	0.06	0.10		

\*Each treatment replicated thrice

\*Figures in parentheses are square root transformed values

\*Figures with similar alphabets do not differ significantly

COMPATIBILITY OF	Trichoderma	WITH	FUNGICIDES

Table 2 : Effect of fungicides on Trichoderma radial growth - per cent inhibition									
Sr. No.	Fungicides	Conc. (%)	T.harzianum	T.virens	Mean				
1.	Copper oxy chloride	0.3	72.2 (58.2)	77.7 (61.8)	75 (60.0) <sup>e</sup>				
2.	Bordeaux mixture	1	13.6 (21.6)	27.7 (31.7)	20.6 (26.7) <sup>h</sup>				
3.	Wettable sulphur	0.2	0.0 (0.0)	4.4 (9.0)	$2.2 (4.5)^{i}$				
4.	Mancozeb	0.25	31.5 (34.1)	54.5 (47.6)	43.0 (40.8) <sup>f</sup>				
5.	Thiram	0.25	88.8 (70.4)	100.0 (90.0)	94.4 (80.2) <sup>b</sup>				
6.	Captan	0.2	80.4 (63.7)	84.8 (67.0)	82.6 (65.3) <sup>c</sup>				
7.	Dinocap	0.1	65.5 (54.1)	77.3 (61.6)	71.4 (57.8) <sup>e</sup>				
8.	Chlorothalonil	0.2	100.0 (90.0)	100.0 (90.0)	100.0 (90.0) <sup>a</sup>				
9.	Carbendazim	0.1	100.0 (90.0)	100.0 (90.0)	100.0 (90.0) <sup>a</sup>				
10.	Benomyl	0.1	100.0 (90.0)	100.0 (90.0)	100.0 (90.0) <sup>a</sup>				
11.	Carboxin	0.2	100.0 (90.0)	100.0 (90.0)	100.0 (90.0) <sup>a</sup>				
12.	Metalaxyl	0.2	100.0 (90.0)	88.1 (69.8)	94.0 (79.8) <sup>b</sup>				
13.	Propiconazole	0.1	100.0 (90.0)	100.0 (90.0)	100.0 (90.0) <sup>a</sup>				
14.	Hexaconazole	0.2	100.0 (90.0)	100.0 (90.0)	100.0 (90.0) <sup>a</sup>				
15.	Tricyclazole	0.06	100.0 (90.0)	100.0 (90.0)	100.0 (90.0) <sup>a</sup>				
16.	Tridemorph	0.1	100.0 (90.0)	100.0 (90.0)	100.0 (90.0) <sup>a</sup>				
17.	Fosetyl-Al	0.15	74.0 (59.3)	84.0 (66.4)	79.0 (62.8) <sup>d</sup>				
18.	Azoxystrobin	0.1	37.7 (37.9)	31.8 (34.3)	35.0 (36.1) <sup>g</sup>				
	Mean		75.8 (69.1) <sup>b</sup>	79.5 (72.3) <sup>a</sup>					
	CV (%)		2.3						
	C.D. (P=0.01)		Fungicides	Isolate	Fungicide X Isolate				
			2.3	0.7	3.3				

\*Each treatment replicated thrice.

\*Figures in parentheses are angular transformed values

\* Figures with similar alphabets do not differ significantly

(1995), Mondal *et al.* (1995), Karpagavalli (1997), Vijayaraghavan and Koshy (2004) and Pandey *et al.* (2006).

Relatively high sensitivity of *T. virens* may be due to the fact that the isolate was obtained from citrus orchard which was less exposed to the fungicidal application. In other words, *T. harzianum* was less sensitive as it was isolated from cotton where in the fungicide usage is more.

#### Variation in fungicide toxicity:

Among the systemic chemicals, azoxystrobin was found moderately compatible with least inhibitory effect on radial growth (35%) to *Trichoderma* isolates followed by 79 per cent with fosetyl Al and 94 per cent with metalaxyl showing least compatibility. All other systemic fungicides were completely incompatible showing 100 per cent inhibitory. It may be noted here that both the benzimidazole group fungicides (benomyl and carbendazim), all the three triazole fungicides tested were 100 per cent inhibitory. Among the contact group of fungicides, only chlorothalonil found completely incompatible showing 100 per cent inhibition on the radial growth of *Trichoderma* isolates. While Bordeaux mixture,azoxystrobin, mancozeb found moderately compatible with radial growth inhibition of 20.6 per cent, 35 per cent and 43 per cent repectively. Only wettable sulphur was found highly compatible with test *Trichoderma* isolates showing least inhibitory effect on the radial growth (2.2%),

Based on the results obtained, all the test fungicides were grouped into incompatible, least compatible, moderately compatible and highly compatible. Carbendazim, benomyl, carboxin, propiconazole, hexaconazole, tricyclozole, tridemorph, chlorothalonil were incompatible with *Trichoderma* spp. showing 100 per cent inhibition of radial growth at field concentration. While dinocap, copper oxychloride, fosetyl-Al, captan, thiram and metalaxyl were found to be least compatible showing more than 70 per cent inhibition of radial growth. Bordeaux mixture, azoxystrobin and mancozeb were moderately compatible with radial growth inhibition in the range of 20-45 per cent. Only wettable sulphur was found to be highly compatible with least inhibition of radial growth (2.2%) of test *Trichoderma* isolates.

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