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Compatibility of *Trichoderma viride* with different fungicide and organic cake

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Abstract
In course of investigation on compatibility of the effective fungicides and organic cake with *Trichoderma viride* by food poisoned technique carbendazim showed almost complete suppression (94.2%) of *Trichoderma viride* followed by carbendazim+ mancozeb (93.8%) and thiophanate methyl (91.5%) at 150 ppm concentration followed by 100 ppm and 50 ppm. Among the cakes, the highest inhibition (19.2%) was recorded in neem cake followed by mustard cake (13.9%) and caster cake (7.6%). Thus all the cakes showed compatibility with *Trichoderma viride.* Present finding suggest that compatible fungicides and organic cakes can be used with *Trichoderma viride* in an integrated disease management practices for the control of soil born pathogen.

Keywords: *Trichoderma viride*, fungicides, organic cake and compatibility

Introduction
Fungi of the genus *Trichoderma* have emerged as most powerful bio-protectants for management of soil borne plant disease. *Trichoderma* have long been known as bio-control agents of plant diseases, and have become a valuable part of agricultural disease control. Various studies have been conducted earlier on compatibility of bio-control agents with chemicals and botanicals. Papavizas (1982) [12] reported that *T. harzianum* can tolerate neomycin sulphate, bacitracin, Penicillin-G and Chlorones-100 µg/ml, sodium propionate 500µg /ml and PCNB 100 mg/ml. Mukhopadhyay *et al.* (1986) [11] reported the tolerance of *T. harzianum* to metalaxyl concentration of 100 ppm. Mukherjee (1987) [10] reported insensitivity of *T. harzianum* to apron, fytolan but sensitivity to thiram. Bhat and Srivastava (2003) [3] reported that bavistin and benlate completely inhibited *Fusarium* and *Trichoderma* even at 250 ppm. Similarly, captaf, calixin, RIL F004, tilt, and indofil M-45 completely inhibited *P. aphanidermatum* and *S. rolfsii* at the same concentration. Indofil M-45 was fungistatic against *T. viride*, while showing complete inhibition of *F. solani, F. oxysporum, P. Aphanidermatum* and *S. rolfsii* at 500 ppm.
To develop an effective disease management programme the compatibility of potential bio-agents with fungicides and organic cakes is essential. Combining antagonists with synthetic and non synthetic chemicals eliminates the chance of resistance development and reduces the fungicides application. In view of this, laboratory experiments were conducted to test the possibility of combining *Trichoderma viride* with fungicides and organic cakes. The long term goal is to develop an effective integrated disease management practices for management soil borne plant diseases as well as to prevent the resistance development in pathogens to chemicals. Integrating chemical resistance *Trichoderma* strains has an important in the framework of integrated disease management.

Material and Methods
Pure culture of *Trichoderma viride* was isolated at Experimental Farm, RAU, Pusa Bihar from Rhizospheric soil from healthy papaya plants was collected in poly-ethylene bags and brought to the research laboratory. Serial dilution technique was used as per Johnson and Curl (1972) [6] to isolate fungal antagonist from rhizospheric soil of healthy plants and dried in shade. Antagonistic mycoflora were isolated on rose bengal agar medium by using a dilutions of $10^3$and 10^4. One ml of soil suspension was poured into sterilized Petri plates containing the melted and cooled medium was poured and then rotated gently to get uniform distribution of soil into the medium. Then, the plates were incubated at 27±2ºC and observed frequently for the development of colonies.
Three fungicides *viz.,* carbendazim, thiophanate methyl and carbendazim + Mancozeb and three organic cakes *viz.,* Mustard Cake, Neem Cake and Castor Cake were evaluated against

Trichoderma viride in vitro to study the sensitivity of bio-agent to fungicide sand organic cakes. Fungicides were tested at 50 ppm, 100 ppm and 150 ppm concentration and each organic cake used at 10 per cent concentration. 20 ml medium having respective concentration was poured under each treatment in three replications in sterilized Petri plates. After solidification of the medium 5 mm disc of test bioagent was inoculated centrally in each plate and incubated at 28±2°C. PDA plate not amended with fungicides and inoculated with bio-agent served as check. Observation on colony diameter was recorded at 240 hrs of incubation.

The radial growth of *Trichoderma viride* was measured in all treatments after 240 hrs of inoculation and compared with control. The per cent inhibition was calculated by using following formula as given by Vincent (1947) [15].

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

Where,

I = Per cent growth inhibition

C=Colony diameter in control Petri plate;

T =Colony diameter in the treated Petri plate.

The per cent inhibition data were analysed statistically using completely randomized design (C.R.D).The calculated value of F was compared with the tabulated values at 5% level of significance for an appropriate degree of freedom.

Result and Discussion

In course of investigation on compatibility of the effective fungicides and organic cake with *Trichoderma viride* by food poisoned technique (Table 1) carbendazim showed almost complete suppression (94.2%) of *Trichoderma viride* followed by carbendazim+ mancozeb (93.8%) and thiophanate methyl (91.5%) at 150 ppm concentration followed by 100 ppm and 50 ppm. Among the cakes, the highest inhibition (19.2%) was recorded in neem cake followed by mustard cake (13.9%) and caster cake per (7.6%). Thus all the cakes showed compatibility with *Trichoderma viride.* Earlier various studies have been conducted on compatibility of bio-control agents with chemicals and botanicals. The pure culture (Fig 1) of *Trichoderma viride* and their compatibility test with various fungicides (Fig 2) and organic cakes (Fig 3) were demonstrated in respective figures given in parenthesis. Papavizas (1982) [12] reported that *T. harzianum* could tolerate neomycin sulphate, bacitracin, Penicillin-G and Chlorones-100 µg/ml, sodium propionate 500µg /ml and PCNB 100 mg/ml. Mukhopadhyay *et al.* (1986) [11] reported the tolerance of *T. harzianum* to Metalaxyl at concentration of 100 ppm. Mukherjee (1987) [10] reported insensitivity of *T. harzianum* to apron, fytolan but sensitivity to thiram.

Akbari and Parakhia (2001) [1] also evaluated 5 different fungicides *viz.*, thiram, mancozeb, tridemorph, metalaxyl and fosety-Al against *Trichoderma harzianum-II, T. viride-II and Gliocladium virens* and found them non-inhibitory at all concentration tested. Chlorothalonil was found inhibitory to *T. harzianum-II* and *T. viride-II* but not to *G. virens* at all four concentrations tested. The systemic fungicide Carbendazim had strong inhibitory effect on antagonists' at all four tested concentrations. Among these, *T. harzianum*-II was more sensitive to carbendazim than *T. viride-II* and *G. virens.* Non-systemic fungicide copper oxychloride was found inhibitory to *T. harzianum, T. viride-II* and *G. virens* but was less inhibitory in comparison to carbendazim. In a similar study, *Trichoderma harzianum* was found highly sensitive to mancozeb, tebbuconazole and thiram, less sensitive to benomyl, triadimenol, dichlofluanid and relatively insensitive to procymidone and captan (Mclean *et al.*, 2001) [9].

Bhat and Srivastava (2003) [3] reported that bavistin and benlate completely inhibited *Fusarium* and *Trichoderma* even at 250 ppm. Madhusudhan *et al.* (2003) [8] reported that mancozeb (75% WP) was safe to *Trichoderma viride* isolates and less effective against *Fusarium solani*. Carbendazim (50% WP) propiconazole (25% EC), tridemorph (80% EC) and hexaconazole (5% EC) were found to be effective against *Fusarium solani* and not safe to *Trichoderma viride* isolates. Upadhyay *et al.*, 2004 reported complete inhibition of growth and sporulation of *Trichoderma viride in vitro* by bavistin and kri-benomyl @25µg/ml while kavach @250µg/ml exhibited only 8.88% growth inhibition and gave good sporulation (8.4 x 106 spore/ml). Singh and Varma (2005) [13] reported that out of the fungicides tested; mancozeb at all concentrations was the most effective in reducing the mycelial growth of the *Fusarium solani* f. sp. *glycines* followed by carbendazim. Mancozeb was also compatible with *T. harzianum* and *T. viride* at 0.05 and 0.1% concentrations. Carbendazim was the least compatible. Gangopadhyay *et al.* (2009) [5] reported that carboxin was highly toxic to *T. harzianum* while chlorothalonil was highly toxic to *T. viride.* Dubey 2009 [4] studied the compatibility of *Trichoderma* species with fungicides, plant extracts, oil cakes and rhizobium. Karanj *(Pongamia pinnata)* cake as well as leaf extract while carboxin, captan and tetramethyl thiram disulphide were selected for further integration as they were compatible with *Trichoderma* as well as *Rhizobium.* Shivpuri and Mali, 2009 recorded maximum tolerance between apron 35 SD (90 mm) at 500 and 1000 ppm followed by dithane M-45, thiram and vitavax power. Bavistin completely inhibited the mycelia growth *Trichoderma* spp. Bagwan (2010) [2] reported that *Trichoderma* was most sensitive to captan, tebbuconazole, vitavax, propiconazole and chlorothalonil likewise Kumar (2012) [7] reported that *Trichoderma virens* was compatible with carboxin + thiram, Karanj cake, acid nitric and Eucalyptus oil.

In course of investigation on compatibility of the effective fungicides and organic cake with *Trichoderma viride* by food poisoned technique. Carbendazim showed almost complete suppression of *Trichoderma viride* followed by carbendazim+ mancozeb but thiophanate methyl at lower concentrations would be compatible for the integrated management of soil borne disease. All the oil cake- mustard cake, neem cake, castor cake were found compatible with *Trichoderma viride*, but mustard cake was also found to be the efficient inhibitor of the pathogen. Long term goal is to develop an integrated disease management strategy by combining *Trichoderma viride*, chemicals and organic cakes so as prevent pathogen from gaining resistance as well as in building up of *Trichoderma* population levels in the soil that will be effective on along term basis.

Table 1: Compatibility of *Trichoderma viride* with some fungicides and cakes

Treatments	Concentration (ppm)	*Radial growth of *Trichoderma viride* (mm)	Inhibition over control (%)
Carbendazim	50	5.0	94.2
	100	5.0	94.2
	150	5.0	94.2
Thiophanate methyl	50	15.7	81.8
	100	10.0	88.4
	150	7.3	91.5
Carbendazim+Mancozeb	50	12.7	85.3
	100	8.7	89.9
	150	5.3	93.8
Mustard cake	10%	74.3	13.9
Neem cake	10%	69.7	19.2
Castor cake	10%	79.7	7.6
Control		86.3	_
LSD (0.05)		2.6	4.2
S. Em. (±)		0.9	1.4
C.V. (%)		5.1	3.5

*Mean of three replications

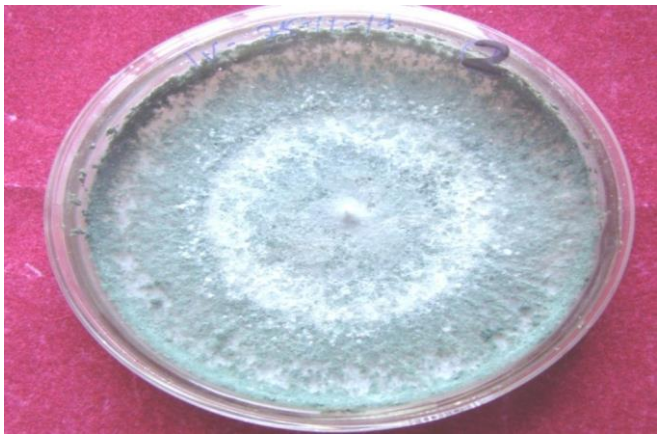

Fig 1: Pure culture of *Trichoderma viride*

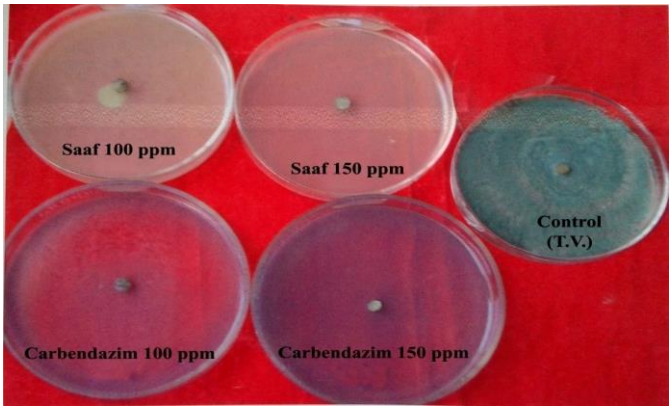

Fig 2: Compatibility of different fungicides with *Trichoderma viride*

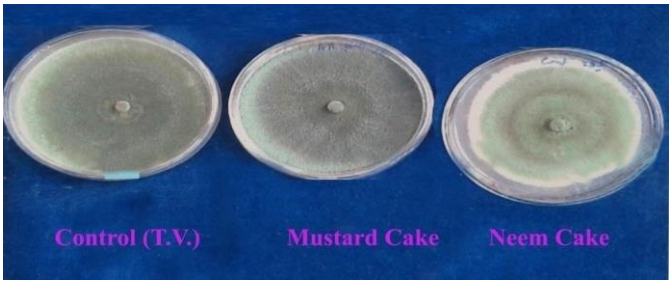

Fig 3: Compatibility of different organic cakes with *Trichoderma viride*

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