J. Hortl. Sci.
Vol. 6(2):141-144, 2011

Short communication

Evaluation of fungicides, soil amendment practices and bioagents against *Fusarium solani* - causal agent of wilt disease in chilli

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ABSTRACT

Chilli is affected by the wilt disease caused by *Fusarium solani,* under irrigated conditions. In absence of resistant cultivars, the disease needs to be controlled by management practices. *In vitro* evaluation of six fungicides by Poisoned Food Technique showed that a combination of carbendazim+mancozeb was effective in inhibiting mycelial growth (93.6%), followed by Carbendazim alone (92.4%). *In vivo* soil drench using the same fungicides proved effective in controlling the pathogen. Integration of different treatments, including seedling dip, with Carbendazim, addition of vermicompost, drenching with fungicide, and application of *Trichoderma viride* was found to be effective in managing the disease, in comparison to individual treatments.

Key-words: Chilli, *Fusarium solani*, fungicides, *Trichoderma viride*, soil amendment

Chilli (*Capsicum annuum* L.) an important condiment and vegetable crop is cultivated and raised for both green and dry fruits. Important chilli growing states of India are Andhra Pradesh, Maharashtra, Karnataka, Orissa and Tamil Nadu, accounting for more than 70% acreage. Andhra Pradesh ranks first in India both in area and production, with 2.04 lakh hectares producing 323 thousand tones (Anon., 2010). However, the crop suffers many diseases such as anthracnose, powdery mildew, wilts, viral diseases, etc. Of late, *Fusariam* wilt has become a serious problem in chilli-growing irrigated tracts of Andhra Pradesh particularly, in black cotton soils, leading to 25% yield loss (Madhkar and Naik, 2000). Symptoms include leaf chlorosis, vascular discoloration and wilting of plants. High temperature and high moisture are conducive for disease development. Reducing soil moisture is an important step for managing the disease. If economically viable, soil fumigants and solarization may be used to reduce pathogen population in the soil. Successful management of these wilt diseases is vital to ensure economic viability of chilli production. Chemical management is an important tool for control of diseases, including soil-borne diseases. In addition, identification of effective fungicides would enable consolidation of different components required to formulate integrated disease management

Isolation of the fungus

The wilt fungus was isolated from roots of chilli plants (cv. LCA) 334 grown at RARS, Lam, by the standard isolation method under aseptic conditions. Infected tissues of the root were cut into small pieces of 1-2 mm size and surface-sterilized with 0.1% mercuric chloride solution for 30 sec and washed repeatedly (thrice) in sterile distilled water and placed in petri plates containing sterilized PDA, and incubated at $28\pm{}^{o}C$. The culture thus obtained was purified by single-spore isolation and identified as *Fusarium solani* based on morphological description given by Barnett (1960).

Soil used

For all pot-culture experiments, black cotton soil sterilized with 5% formaldehyde solution was used, for raising chilli plants. Each plant was raised individually in 2kg capacity plastic bag. Three replications, each comprising 20 plants, were taken for each treatment.

In vitro evaluation of fungicides

Four systemic (Carbendazim 0.1%, Benomyl 0.1%, Tebuconazole 0.5%, Thiophanate methyl 0.1%) and one non-systemic (Pencycuron 0.5%) and one combination of systemic and contact (Carbendazim + Mancozeb 0.2%)

fungicides, were evaluated against *F. solani*. All fungicides were tested at their recommended dose of concentration.

Poisoned Food Technique

Recommended dose of fungicide was added to molten PDA just before pouring it into plates. Twenty milliliter of medium with the desired concentration of fungicide was poured into each sterilized petriplate. Suitable checks were laid for comparison. Five millimeter mycelial disc of *F. solani* was taken from the periphery of 10 day old culture and placed in the centre of the plate, and incubated at 28$\pm$ ^{o}C. Growth of the fungus was measured by measuring its diameter in two directions and the average value was recorded. Final-growth reading was recorded when growth of the fungus in the control plate was full. Per cent inhibition of growth was calculated using the formula given by Vincent (1947).

Evaluation of fungicide by soil-drenching

Sterilized soil was filled in polythene bags of size 7.5cm width and 20cm length. To evaluate six fungicides by soil-drenching, *F. solani* was mass-multiplied on sorghum grain and the culture was placed at the root zone in the chilli plant. All the test-fungicides were drenched at the rate of one libe fungicide solution/bag, and allowed to percolate to the corresponding depth. Their efficacy in inhibiting fungal growth was evaluated by recovering the fungus from the soil of rhizosphere.

Evaluation of *Trichoderma* species

Four isolates of *Trichoderma viride* collected from different chilli growing areas of Andhra Pradesh were categorized as I, II, III and IV, and evaluated under *in vitro* conditions for their antagonistic activity against the wilt pathogen, by dual-culture technique (Huang and Hoes, 1976). Mycelial discs, of 5mm diameter each of bioagents and the pathogen, were taken from the margins of actively growing cultures and transferred onto potato dextrose agar medium in Petri plates on the opposite sides. The Petri plates were subsequently incubated at 28$\pm$1^{o}C. Colony diameter of the test-fungus upto the zone of inhibition was recorded for each bioagent, and per cent growth inhibition of the test pathogen was calculated.

Mass multiplication of biocontrol agent *Trichoderma viride*

Effective isolate of *T. viride* identified in *in vitro* studies was mass-multiplied in potato broth and mixed in talc powder, and further used in pot-culture experiments.

Evaluation of the integrated disease management module for wilt disease

A pot-culture experiment was conducted in factorial randomized block design during 2008-09 and 2009-10 crop seasons at Regional Agricultural Research Station, Lam, Guntur.

Different treatments including seedling dip in fungicide, soil amendment with vermicompost, addition of biocontrol agent *T. viride*, drenching of effective fungicide and combination of all these treatments were evaluated to develop a reliable, eco-friendly integrated disease management approach.

Vermicompost was taken as soil amendment based on reports of significant superiority in reducing the *Fusarium* wilt disease incidence in fenugreek (Kamlesh Mathur *et al*, 2006) Best isolate of *T. viride* isolated was applied at the rate of 10g/pot at the time of inoculation of *F. solani*. Similarly the fungicides which proved effective in *in vitro* studies were used for seedling dip for 30 minutes before planting and also for drenching treatments.

Incidence of wilt was recorded after 10 days of inoculation.

Among different fungicides tested carbendazim + mancozeb was found to be highly effective in inhibiting the growth of *F. solani* (93.6%) followed by carbendazim (92.4%) and benomyl (91.34%). Tebuconazole (83.1%) and thiophanate methyl (80.1%) were also found effective in inhibiting the mycelial growth of *F. solani* whereas the pencycuron (4.1%) was least effective or ineffective for control of *F. solani* (Table1).

The results are in conformity with the findings of previous workers (Verma and Vyas, 1977; Haware *et al*, 1978; Hiremath *et al*, 1981; Nene *et al*, 1991 and Naik *et al*, 2007).

Among four isolates, *T. viride* I was the most effective (80.2 % inhibition of the pathogen), *T. viride* isolate II showed a growth inhibition of 67.5 % whereas the other two isolates of *T. viride* IV and *T. viride* III were less effective with 62.7 and 63.5% (Table 2) inhibition, respectively.

Wang *et al* (1995) found that *T. viride* had strong antagonistic activity towards *F. solani* under *in vitro* conditions. Different species of *Trichoderma* have been reported to be effective against *Fusarium sp* causing wilt in chilli, cumin and gladiolus (Suneel Anand and Harender Raj Gautam, 2006)

Pot culture experiment

In soil drenching, all the systemic fungicides were found effective at any concentration and at any depth of inoculum tried viz., 10 and 15 cm in the soil column. Among them carbendazim+mancozeb and carbendazim, recorded 100 percent inhibition of mycelial growth at 1000, 2000 and 3000 ppm at both depths followed by benomyl which recorded 90 percent inhibition with 2000 and 3000 ppm at 10cm depth (Table 3). Whereas tebuconazole and thiophanate methyl were effective at 3000 ppm when applied at 10 and 15 cm depths. Contact fungicide pencycuron was ineffective in all 3 concentrations at 10 and 15 cm depths. The results are in conformity with the findings of previous workers (Verma and Vyas,1977; Haware *et a*l, 1978; Hiremath *et al*,1981 and Nene *et al*, 1991).

Table 1. Inhibition of mycelial growth in *Fusarium solani* by different fungicides

S.No.	Treatment	Mean radial growth of fungus (cm)	Per cent growth inhibition
1	Carbendazim + Mancozeb (0.2%)	0.57	93.60
2	Benomyl (0.1%)	0.77	91.34
3	Carbendazi-m(0.1%)	0.67	92.47
4	Tebuconazole(0.15%)	1.50	83.08
5	Thiophanate methyl(0.1%)	1.70	80.82
6	Pencycuron(0.1%)	8.50	4.13
7	Control	8.87	
	SEM±	0.05	
	C.D. (*P=0.05*)	0.15%	

Table 2. Inhibition of mycelial growth in *Fusarium solani* by different *Trichoderma viride* isolates

S.No.	Fungal isolate	Radial growth (cm)	Per cent inhibition of growth
1	*Trichoderma viride I*	1.78	80.2
2	*Trichoderma viride II*	2.93	67.5
3	*Trichoderma viride III*	3.33	63.5
4	*Trichoderma viride IV*	3.55	62.7
5	Control (*F. solani* alone)	9.00	
	SEM ±	0.04	
	C.D. (*P=0.05*)	0.12	
	CV%	1.90	

Table 3. Effect of drenching with different fungicides on inhibition in mycelial growth of *F. solani* at various depths

Chemical	Soil depth (cm)	Percent inhibition of growthConcentration		
		1000	2000	3000
Carbendazim + Mancozeb	10	100	100	100
M-45 (0.2%)	15	100	100	100
Carbendazim (0.1%)	10	100	100	100
	15	100	100	100
Benomyl (0.1%)	10	90	100	100
	15	90	100	100
Tebuconazole (0.15%)	10	50	100	100
	15	00	50	100
Thiophenate-methyl (0.1%)	10	00	00	50
	15	00	00	50
Pencycuron (0.1%)	10	00	00	00
	15	00	00	00

Integrated disease management of chilli wilt.

Of all the treatments integration of seedling root dip with carbendazim, addition of vermicompost, drenching of fungicide carbendazim+mancozeb and soil application of *T. viride* was found to be effective with minimum (Table 4) mortality of plants (5.83 %). The present findings are in conformity with the earlier reports, integration of disease management practices are more effective in controlling *Fusariam* wilt in gladiolus (Suneel Anand and Harender Raj Gautam, 2006), collar and root rot in strawberry (Bhardwaj and Gautam, 2004) and soil borne diseases in vegetable nurseries (Steven *et al*, 2003)

Next best treatment was the drenching with carbendazim+mancozeb (16.1%) followed by seedling root dip in carbendazim (21.6%) for 30 minutes. Similar findings were reported by earlier workers. Seedling dip in carbendazim at 0.1 and 0.2% significantly reduced the

Table 4. Efficacy of different treatment combinations in management of wilt disease of chillies caused by *F. solani* in pot-culture during 2009 and 2010

S.No.	Treatment	Per cent of plants dead (2009)	Per cent of plants dead (2010)	Per cent of plants dead pooled (2009 &2010)
1	T1 : Seedling dip-Carbendazim @0.1%	24.60 (29.73)	17.60 (24.80)	21.60 (27.69)
2	T2 : Vermicompost-100g/kg soil	44.00 (41.55)	36.60 (37.23)	33.60 (35.43)
3	T3: T1+T2	30.60 (33.58)	24.30 (29.53)	26.11 (30.72)
4	T4: Drenching with fungicide	11.60 (19.91)	20.60 (26.99)	16.10 (23.75)
5	T5: Application of *Trichoderma viridi* @10g/pot	07.30 (15.68)	30.30 (33.40)	18.80 (25.70)
6	T6: (T3+T4+T5)	0.00 (0.00)	11.60 (19.91)	5.83 (13.94)
7	T7: Uninoculated Check	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
8	T8: Inoculated Check	97.60 (81.09)	99.60 (98.00)	97.16 (80.19)
	SEM ±	1.13	1.83	2.59
	C.D. (*P=0.05*)	3.41	5.56	7.86

Figures in parentheses are transformed values tomato wilt incidence caused by *Fusarium oxysporum f.sp lycopersicii* (Saini *et al*, 2005).

In general, soil amendments found effective against the pathogen by changing pH and also by enhancing the growth of biocontrol agents. Addition of vermicompost with other treatments like seedling root dip (26.1%) and integrated treatment (5.83%) is effective when compared with individual treatments. Kamalesh Mathur *et al* (2006) also found that the addition of vermicompost was significantly superior in reducing the *Fusarium* wilt of fenugreek.

Application of *T. viride* (18.8%) alone was not much effective in reducing the disease *in vitro* studies, the reasons may be the climatic and the soil conditions at the time of application. Similar observations were reported by De Cal *et al* (1995) and Saini *et al* (2005).

Although disease resistant varieties are preferred over management of soil borne diseases such as *Fusariam* wilt there are not many alternatives except to take up drenching with fungicide particularly when the wilt has already appeared in the field. Based on the investigation in the abscence of resistant cultivar, drenching of systemic fungicide such as carbendazim+mancozeb can be recommended as immediate control measure to protect against wilt. Finally adoption of integrated disease management was suggested for effective control of wilt disease.

REFERENCES

Anonymous. 2010. http://www.ikisan.com

Barnett, H.L. 1960. Illustrated genera of imperfect fungi. 2nd edition, Burgess Publishing Company, Minneapolis

Bhardwaj, U. and Gautam H. R. 2004. Mulching with transparent polythene and root dip in fungicides for the management of collar and root rot of strawberry. *Indian Phytopath*., **57:**48-52

De Cal, A., Pascual, S., Larena, I. and Melgare, J. 1995. Biological control of *Fusarium oxysporium f. sp. lycopersici. Pl. Pathol.,* **44**:909-917

Haware, M.P., Nene, Y.L. and Rajeswari, R. 1978. Eradication of *Fusarium oxysporum* f. sp. *ciceri* transmitted in chickpea seed. *Phytopathol.*, **68**:1364-1367

Haung, H.C. and Hoes, J.A. 1976. Penetration and infection of *Sclerotina sclerotiorum* by *Coniothyrium minitans*. *Can. J. Bot*., **54**:406-410

Hiremath, P.C., Sulladmath, V.V. and Ponnappa, K.M. 1981. Chemical control of betelvine decline. *Pesticides,* **15**:11-12

Kamlesh Mathur, Bansal, R.K. and Gurjar, R.B.S. 2006. Organic management of *Fusarium* wilt of fenugreek (*Trigonella foenumgraecum* L.) A seed spice. *J. Mycol. Pl. Pathol.,* **36:**94-95

Madhukar, H.M. and Naik, M.K. 2004. Evaluation of bioagents against fusarium wilt of chilli (*Capsicum annuun* var. annuum) Proc. 15th *Int'l. Plant Protection Congress on Plant Protection Towards 21st Century,* Beijing (China), pp. 540

Naik, M.K., Madhukar, H.M. and Devika Rani, G.S. 2007. Evaluation of fungicides against *Fusarium solani*, the causal agent of wilt of chilli. *Veg. Sci*., **34**:173-176

Nene, Y.L., Reddy, M.V., Haware, M.P., Ghanekar, A.M. and Amin, A.S. 1991. Field diagnosis of chickpea diseases and their control. ICRISAT, Information Bulletin, No. **28,** Hyderabad, India

Saini, A.K., Indu Jalali and Vijay Pal. 2005. Eco-friendly management of *Fusarium* wilt-root-knot nematode complex in tomato. *J. Mycol. Pl. Pathol.,* **35:**309-311

Stevens, C., Khan V.A., Kabana, R., Ploper L.D., Bakkan P.A., Collins, D.J., Brown, J.E., Wilson, M.A. and Igmedge, E.C.K. 2003. Integration of soil solarization and chemical, biological and cultural control for the management of soil-borne diseases of vegetables. *Pl. Soil,* **253**:493-496

Suneel Anand and Harender Raj Gautam. 2006. Use of soil solarization, biocontrol agents, fungicide corn dip and soil amendments for management of *Fusarium* wilt pathogen of gladiolus. *J. Mycol. Pl. Pathol.,* **36**:201-204

Verma, R.K. and Vyas, S.C. 1977. Effect of seed treatment with systemic fungicides in gram wilt control. *Pesticides,* **11**:20-21

Vincent J.M. 1947. Distortion of fungal hyphae in the presence of certain inhibitors. *Nature,* **150**:840

Wang K., Zhang, Z., Chen, J.N., Fan, Z., Niu, B.S and Wang, K.C. 1995. A study on the occurrence of root rot of pea (*Pisum sativum*) and the technique for its integrated control. *Nigerian J. Agril. Forestry Sci. Tech*., **5**:1-5

(MS Received 27 December 2010, Revised 23 September 2011)