

Efficacy of Bioagents and Fungicides Against *Colletotrichum dematium*—the Incitant of Fruit Rot of Chilli

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Abstract

Studies were made to find out the effective fungicide and bioagents against *Colletotrichum dematium* causing fruit rot of chilli (*Capsicum annum* L.). Different fungicides and bioagents were tested *in vitro* in which TMTD (0.3%) and TMTD + carbendazim (0.2% + 0.1%) inhibited 100% growth of *Colletotrichum dematium*. While *in vitro* evaluation of bioagents, *Trichoderma viride* inhibited maximum of 40.89% growth of *C. dematium*, whereas *Pseudomonas fluorescense* restricted 38.00% of mycelial growth. The treatment of TMTD showed maximum germination of 87.33% and seedling vigor index of 1268.90. While highest SVI was recorded in Alliete as 1304.70 and germination of 85.33%. Among bioagents highest germination of 89.00% and SVI of 1515.67 were found in treatment with *Trichoderma viride* as compared to inoculated and uninoculated controls.

Key words : *Colletotrichum dematium*, Bioagents, Fungicides fruit rot, *Capsicum annum* L., Growth parameters.

Chilli (*Capsicum annum* L.) is an indispensable condiment in India and is cultivated as one of the important cash crops. It is also having medicinal value because the pungent principle of chilli “Capsaicin” is not only a digestive stimulant and preventive for heart diseases but also a curative for many rheumatic troubles. The green and red chilli fruits are valuable on account of their richness in ascorbic acid, carotene, protein, carbohydrates, mineral matters (CSIR 1950). But most of the commercial varieties of chilli under cultivation suffers from many destructive diseases like dieback (*Colletotrichum dematium*), fruit rots (*Alternaria alternata*, *Colletotrichum* spp. *Macrophomina phaseolina*), powdery mildew (*Levuelulla taurica*) and churda murda complex (Grover 1974), which results into considerable loss in yield and quality of green and red chillies. Among them the fruit rot is most serious disease, causing heavy losses in yield and quality of produce, also the fruit rotting fungi have direct bearing on yield (Chaudhary 1957). These fungi also affect the seed germination, seedling vigor of chilli (Dhawale and Kodmelwa 1978, Perane and Joi 1988). Hence an attempt was made here to find out the suitable solution, which restricts the

pathogen and for that different fungicides and bioagents were tested *in vitro*. Their effect on germination and seedling vigor were also studied.

Methods

Isolation of Fungi from Seeds

The seeds were given preliminary treatment as follows. Seed disinfected with 0.1% of mercuric chloride ($HgCl_2$), which were used for the isolation of internal seed borne mycoflora. Seeds without surface disinfection were also used for the isolation of external seed borne mycoflora.

The seeds were plated at equidistance by means of sterile flamed forcep. For blotter paper method and agar plate method, 25 seeds were plated in each petriplate. However, while plating the seeds on blotter surface, care was taken that the blotter were sufficiently moist. The unsterilized and surface sterilized seeds were placed to obtain the seed mycoflora from external and internal surface of seeds.

Seeded plates were incubated in laboratory at room temperature 27 ± 1 C under 12 hours of alternating cycles near ordinary tube light and darkness.

Table 1. *In vitro* evaluation of different fungicides against *C. dematium* by poison food technique.

Fungicides	Conc. (%)	Mean colony diameter (mm)				Per cent growth inhibition
		2 nd day	4 th day	6 th day	8 th day	
TMTD	0.3	0.0	0.0	0.0	0.0	100
TMTD + Carbendazim	0.2+0.1	0.0	0.0	0.0	0.0	100
Thiophanate-M	0.2	0.0	0.0	0.0	1.0	98.34
Metalaxyl	0.4	3.66	7.0	18.33	24.33	59.67
Alliete	0.4	0.0	1.66	2.33	2.33	96.13
Chlorothalonil	0.3	4.0	5.00	8.0	12.0	80.10
Curzate	0.2	0.0	0.0	2.0	3.33	94.18
Control	–	8.33	21.66	45.66	60.33	–

The fungal flora after 5 and 6 days were observed from blotter paper and agar plate method, respectively. Observations of fungal colonies on and around the seeds were lifted by sterile inoculating needle and transferred to PDA slants.

Identification of isolates was done on the basis of morphology and different characteristics of fungi given in the standard guides and manuals.

Evaluation of Fungicides

By Poison Food Technique. The nutrient medium was poisoned with fungitoxicant following the concentration and then allowed test fungi to grow on medium and then mycelial inhibition was studied. For that purpose TMTD, TMTD + carbendazim, thiophanate-M, metalaxyl, alliete, chlorothalonil, curzate fungicidal treatments were used. In each flask 100 ml of PDA was taken and sterilized in autoclave, in such flask required amount of fungicides were added separately. About 20 ml of this poisoned PDA medium poured in each sterilized petriplates and allowed to solidify. Then the plates were inoculated with 5 mm disc of *Colletotrichum dematium* in center and incubated 27 ± 1 C. Normal plates of PDA inoculated with test organism also serve as a check.

Table 2. *In vitro* evaluation of different bioagents against *C. dematium* by dual culture method.

Bioagents	Mean colony diameter (mm)	Per cent growth inhibition
<i>Trichoderma viride</i>	34.5	42.81
<i>Trichoderma harzianum</i>	37.16	38.40
<i>Pseudomonas fluorescense</i>	35.66	40.89
Control	60.33	–

Per cent inhibition of mycelial growth was calculated by the formula

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

Where, I– Inhibition percentage, C–Growth in check plates, T–Growth in treatment plates.

Towel Paper Method. First, 100 seeds in each treatment were inoculated with *Colletotrichum dematium* by rolling on 10 days old culture and incubated in moist chamber for 24 hours for growth of fungus on the seeds and then these seeds were treated with fungicides. Afterwards these seeds were plated in towel paper. Fifty seeds on each paper with two replications were plated. The observations of germination, shoot length, root length were recorded.

Testing of Bioagents

By Dual Culture Technique. Autoclaved PDA medium was poured in petriplates and allowed to solidify. Then 5 mm disc of *Colletotrichum dematium* in center and antagonist disc on the four sides of petriplates were plated and allowed to grow. Antagonists used were *Trichoderma viride*, *Trichoderma harzianum* and *Pseudomonas fluorescense*. The suitable control of *C. dematium* was kept. Observations of colony diameter were recorded and better zone was calculated.

Towel Paper Method. As mented for fungicides treatment, here instead of fungicides bioagents were used.

Table 3. Effect of seed dressing fungicides on germination and SVI of chilli. SVI—Seedling vigor index. Figures in parentheses are square root transformed values.

Fungicides	Conc. (%)	Germination (%)	Root length (cm)	Shoot length (cm)	SVI
TMTD	0.3	87.33 (9.35)	9.50	5.03	1268.90
TMTD + Carbendazim	0.2 + 0.1	74.33 (8.62)	8.00	4.76	948.45
Thiophanate-M	0.2	81.66 (8.98)	8.06	4.50	1025.65
Metalaxyl	0.4	79.00 (8.89)	7.93	5.10	1024.37
Alliete	0.4	85.33 (9.24)	9.86	5.43	1304.70
Chlorothalonil	0.3	80.66 (8.98)	9.80	5.00	1113.11
Curzate	0.2	71.33 (8.41)	6.83	4.66	819.58
Inoculated control	—	70.00 (8.37)	3.46	2.00	382.2
Uninoculated control	—	79.00 (8.89)	8.03	5.26	1045.17
<i>F</i> test	—	Sig.	Sig.	Sig.	
SE ±	—	0.06	0.18	0.09	
CD at 1%	—	0.19	0.54	0.28	

Results and Discussion

Table 1 shows that all the treatments were significantly superior over control, inhibiting the growth of *C. dematium*. Among all the fungicidal treatments TMTD (0.3%) and TMTD + carbendazim (0.2% and 0.1%) arrested complete mycelial growth of fungus (100%), followed by thiophanate-M (0.2%) having 98.34%. However, lowest inhibition was recorded in the treatment with metalaxyl (0.4%) that was 59.67% as compared to control. Similar effectivity of TMTD, TMTD + carbendazim against *C. dematium* was reported by Joshi and Wangikar (1978) and Kakade and Khune (1989). Similar results were also reported by Hansoon and Khan (1979), Rao et al. (1990).

Table 2 shows that all the bioagents were found to be effective in inhibiting the radial growth of *Colletotrichum dematium* over control. Amongst the three bioagents *Trichoderma viride* was found to be most promising with 42.81% growth inhibition followed by *Pseudomonas fluorescence* 40.89%. Least was observed in *Trichoderma harzianum* (38.40%). These results are in conformity with Denis and Webster (1977) when they studied the antagonistic properties of species *Trichoderma* grown against different test fungi and found that when they were grown in dual culture suppressed the growth of *Colletotrichum* spp. Similar results were also reported by Naik et al. 1997, Khan and Gupta 1998.

Table 4. Effect of bioagents seed treatment on germination and SVI of chilli. SVI—Seedling vigor index. Figures in parentheses are square root transformed values.

Bioagents	Germination (%)	Root length (cm)	Shoot length (cm)	SVI
<i>Trichoderma viride</i>	89.00 (9.43)	11.00	6.03	1304.70
<i>Trichoderma harzianum</i>	85.66 (9.25)	10.00	5.13	1113.11
<i>Pseudomonas fluorescence</i>	82.00 (9.05)	9.00	5.00	819.58
Inoculated control	70.99 (8.37)	3.46	2.00	382.2
Uninoculated control	79.00 (8.89)	8.03	5.26	1045.17
<i>F</i> test	Sig.	Sig.	Sig.	
SE ±	0.08	0.16	0.09	
CD at 1%	0.24	0.48	0.28	

Seed artificially inoculated with *Colletotrichum dematium* were treated with different fungicides. Table 3 reveals that among seven fungicides highest germination of 87.33% was found in treatment with TMTD (0.3%) and lowest germination in curzate (71.33%) as compared to inoculated control (70.00%). The highest seedling vigor index was observed in alliete (0.4%) that was 1304.70 followed by TMTD 1268.90. Minimum SVI was observed in curzate (819.58), and in uninoculated control germination was 79.00% and seedling vigor index was 1045.17. These findings are also in conformity with the results obtained by Grover and Bansal (1970), Jharia et al. (1977), Kumar and Mahmood (1986), Datar and Malale (1990) and Smith et al. (1999).

Table 4 indicates that all treatments were significantly superior over inoculated and uninoculated control. Among these *Trichoderma viride* has given maximum germination (89%) followed by *Trichoderma harzianum* (85.66%). Similar results were also reported by Handoo and Aulakh (1979), Martins Corder (1997). *Pseudomonas fluorescence* showed germination up to 82.00%. Lowest germination (70.99%) was observed in inoculated control. The highest seedling vigor index was also observed in *T. viride* (1515.67) followed by *T. harzianum* (1296.04) and lowest in inoculated control (382.20). Similar trends of results were also reported by Jayalakshmi et al. (1998), Tehrani and Omati (1999).

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