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J. Mycopathol. Res. 60(3) : *363-375, 2022;*
ISSN 0971-3719

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J. Mycopathol. Res. **60(3)** : 363-375, 2022; (ISSN 0971-3719)

Integrated disease management of root rot and stem decay disease in *Amaranthus hybridus* caused by *Fusarium solani*

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Received : 30.03.2022 Accepted : 31.07.2022 Published : 26.09.2022

To control root rot and stem decay disease of *Amaranthus hybridus,* different strategies were undertaken – (1) application of potent antagonists. (2) application of botanicals, (3) application of fungicides and antibiotic, (4) soil solarization and integrated disease management. Antagonistic effect of *Trichoderma harzianum* and *Trichoderma viride* were assessed *in vitro* and in field condition to control the pathogen, *Fusarium solani*. In both the cases, *Trichoderma harzianum* exhibited better performance to control the disease. *Trichoderma harzianum* can inhibit 100% growth *in vitro* at a dose of 5ml/ 15ml PDA. Likewise, in field condition *Trichoderma harzianum* can reduce disease incidence 31.69 % and 41.28% in 2013 and 2014 respectively. Inhibitory effect of phytoextracts was studied *in vitro* and in field conditions. *In vitro* effect of fungicides and antibiotic on the growth of pathogen was carried out. Bavistin proved its superiority as it can inhibit 94.03% growth of the pathogen *in vitro*. Bavistin showed highest inhibition (50.75 % and 54.95 % in 2013 and 2014 respectively) whereas Baynate showed 27.01% and 33.37% disease reduction in 2013 and 2014 respectively. Integrated disease management (IDM) approach was undertaken to control root rot and stem decay disease. IDM was carried out by the different combinations of potential antagonist (*Trichoderma viride* and *Trichoderma harzianum*), phytoextracts (*Azadirachta indica* and *Allium sativum*) and minimum dose of fungicide (Bavistin) *in vitro*. But the combinations of *Trichoderma harzianum*+ Bavistin+ *Azadirachta indica* and *Trichoderma viride* +Bavistin+*Azadirachta indica* showed good performance at minimum concentration (1:1:1).

Key words: Antagonists, *Amaranthus hybridus,* botanicals, fungicides, *Fusarium solani*, integrated disease management, soil solarisation,

INTRODUCTION

Amaranthus hybridus is a leafy vegetable, belonging to family Amaranthaceae. It is reported that Amaranth production is reduced by various fungal diseases (PROTA, 2004). Rot disease cause severe damage in Amaranth production (Awarum and Ogbonna, 2013).

To avoid damage in productivity, farmers use chemical pesticides like Dithane M-15, Benlate etc. (PROTA, 2004). These chemical pesticides caused chemical hazards as well as poisoning of the leafy vegetable (FAO, 2000). However in the recent past root rot and stem decay of *Amaranthus*, a new emerging disease caused by *Fusarium solani* (Parveen *et al.* 2016), posing a serious threat to *Amaranthus* cultivation in agro-ecological conditions of West Bengal, which causing up to 95 per cent crop damage. Integration of different treatments, including seedling dip, with carbendazim, addition of vermicompost, drenching with fungicide (carbendazim+ mancozeb), and application of *Trichoderma viride* was found to be effective in managing the disease, in comparison to individual treatments (Kumar and Singh, 2016). Madhavi and Bhattiprolu (2011) reported complete reduction of the pathogen with the help of soil solarization alone or with low dosages of a fungicide, biocide and biological control agent. Integrated plant disease management is a process which involves the use of multiple agents to minimize the growth of pathogen. It is an approach that uses all available management strategies to control disease under an economic injury threshold (Khokar and Gupta, 2014). Integrated disease management strategy of *Fusarium oxysporum* causing wilt of *Coleus* was attempted by using

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antagonistic agents like different species of *Trichoderma*, phytoextracts and fungicides (Khatun, 2020). The aim of the study is to standardize a formulation for controlling the root rot and stem decay disease of *Amaranthus* in an ecofriendly way.

MATERIALS AND METHODS

Application of potent antagonist against F. solani in vitro

15 ml of PDA medium (pH 7.4) was plated in sterilized petridishes. Prior to solidification of PDA medium, soluble metabolites of *Trichoderma viride* and *Trichoderma harzianum* at different doses (1, 2, 4 and 5 ml/ 15 ml PDA medium) were mixed to Petridishes separately and homogenized thoroughly. The medium was allowed to cool to room temperature and inoculated with test pathogen and finally incubated at $28^0 \pm 2^0$ C for 7 days. The percentage inhibition of the growth of the pathogen was calculated against control set. Four replications were made for each treatment.

Field trials with the application of potent antagonists to control the infection

Potential antagonists i.e. *Trichoderma viride* and *Trichoderma harzianum* were subjected to field trial for two consecutive years (2013 and 2014) to assess the degree of controlling infection of the cultivar caused by *F. solani.* Colony interaction between antagonists and test pathogen (*F. solani*) was tested by "Dual culture technique". Preparations of different biocontrol agents were made from pure culture of antagonists. Individual antagonists (*Trichoderma viride* and *Trichoderma harzianum*) and test pathogen (*F. solani)* were grown separately in PDA medium. Further, PDA medium were taken (250 ml in each flask) and autoclaved. Then these media were inoculated with respective antagonists (*Trichoderma viride* and *Trichoderma harzianum*) and incubated at $28^0 \pm 2^0$C for 15 days. After incubation, these mycelial mats were harvested and the culture filtrates were collected. These culture filtrates were taken separately for each antagonist, which served as a source of biocontrol preparations. The culture filtrates were applied directly to the soil at an interval of 15 days. The mycelial mats as one of the biocontrol preparations were applied to the soil by mixing with organic wastes like wheat bran and saw dust in the form of mass inoculums, with a view to get the mass inoculums established more successfully and vigorously. Saw dust and wheat bran were soaked in water for 45 min. and excess water drained off. They were mixed thoroughly in 1:1 proportion in which small quantity of malt extract was added. The mixture was taken in autoclavable plastic bags made air tight and sterilized at 20 lbs pressure, 121^0C for 1 h and finally inoculated separately with the mycelial mats of the antagonists. The bags were then inoculated at $28^0 \pm 2^0$C for 15 days. After the incubation, the mass inoculums of the respective antagonists by removing the bags were directly applied to the soil. Control sets were also being maintained. Results were recorded.

Control of disease by the application of botanicals

Screening of botanicals on the basis of growth inhibition of the pathogen

Plant extracts were prepared following the method of Ansari (1995). Fresh and healthy young parts of different plants like leaves of Neem (*Azadirachta indica*), garlic bulb (*Allium sativum*), Bokul leaf (*Mimusops elengi*), Amlaki fruit (*Phyllanthus emblica*), Papaya (*Carica Papaya*) were cut into small pieces (1-2cm). Plant parts (50 g) were washed thoroughly with distilled water and crushed in a mortar and pestle separately. They were filtered through two layer of cheese cloth and then filtered through whatman-1 filter paper. The filtrate was centrifuged at 5000 rpm for 20 minutes. The supernatant was filtered with sterilized sintered glass filter (pore size 1-2 µm). The sterilized filtrates were stored at 4^0C for future use.

Bioassay of plant extracts

To evaluate the effect of different phytoextracts as biocide against invitro growth of the pathogen was studied following "food poisoning technique" (Mondal *et al.* 1995). PDA medium was sterilized and maintain pH 7.4. 15ml PDA different quantities of different phytoextracts (1.0, 2.0, 4.0, and 5.0 ml) was added to 15 ml PDA medium prior to its solidification. Then mix thoroughly to have a homogeneous mixture, cooled at room temperature and inoculated with pathogen, *Fusarium solani*. The plates were inoculated at $28^0 \pm 2^0$C for 7 days and radial growth of the pathogen was measured. Growth inhibition Percentage of

pathogen is was calculated against control set. Four replications were made for each treatment.

Field trials with potent plant extracts to reduce the infection

The plant extracts (neem, garlic) showed the most effective response in comparison to other botanicals by maximum inhibition of growth and development of *F.solani in vitro* . Hence, these 2 phytoextracts were also subjected to field trials in order to determine their potentiality in controlling the incidence of the pathogen. Field trial was performed in four replications following Randomize block design. Each plot is 2m X 2m in size. Plant extracts were directly added to the soil at 10 days intervals at rate of 500 ml/ plot. Water sprayed plot is considered as control set. Treatments were carried out in two successive years (2013 and 2014). Percentage of disease reduction was recorded against control set.

Disease control by the application of fungicides and antibiotic

Screening of different fungicides and antibiotic on growth inhibition of the pathogen

Effect of different fungicides viz. Baynate (Bay Organics Ltd., MGR), Bavistin (BASF India Ltd., Mumbai), Captan (Captaf, Rallis India Ltd., Mumbai), Mancozeb (Anu Production Ltd., Haryana) and an antibiotic griseofulvin were studied *in vitro* growth of the pathogen. Different doses (0.01, 0.05, 0.1 and 0.5%) were used for each fungicide and antibiotic. Fifteen ml of potato dextrose broth at pH 7.4 was sterilized, cooled at room temperature and mixed thoroughly with 1 ml of different concentrations of the fungicides and antibiotic separately to make the medium poisoned and allow to solidify in room temperature. Then these media were inoculated with 5 mm inoculum discs of the test-pathogen *F.solani* and incubated at $28^0 \pm 2^0$C for 7 days. Identical control sets were also maintained. After the inculcation, the mycelial mats were harvested, washed with distilled water and dried to a constant weight. The percentage inhibition of the growth of the pathogen was calculated against control set. Four replications were made for each treatment and their mean values are presented.

Field trials with effective fungicides for reduction of infection

Field trials were conducted with the efficient fungicides at the dose, in which they exhibited most effective performance under *in vitro* trials. Field Trials were performed for 2 successive years. Four replications were made for the purpose following randomized block design. Each plot was 2m X 2m in size. First spray was applied before 15 days of transplant and subsequent two sprays were done at 10 days intervals (500 ml/ plot). Water sprayed plot was compared as control set.

Integrated disease management and soil solarization

Integrated management approach towards in vitro growth of pathogen

Integrated management of the disease was performed by the application of potential antagonist (*Trichoderma viride* and *Trichoderma harzianum*), fungicide (Bavistin) and phytoextracts (Neem and Garlic) *in vitro*. Different combinations were made viz. *Trichoderma harzianum* + Bavistin, *Trichoderma viride*+ Bavistin , *Trichoderma harzianum* + Bavistin + *Azadirachta indica, Trichoderma viride*+ Bavistin+ *Azadirachta indica, Trichoderma harzianum*+ Bavistin+ *Allium sativum,* and *Trichoderma viride* + Bavistin+ *Allium sativum.* PDA medium was prepared and pH was maintained at 7.4. The medium was autoclaved and and 15 ml. were poured to each Petridish. Prior to solidification of medium, different compatible combinations were applied to the medium and mixed thoroughly to make it homogeneous. The plates were allowed for solidifications and inoculated with *F.solani.* These plates were inculcated in $28^0c \pm 2^0c$ for 7days.The radial growth of the pathogen was measured against the control set (*F. solani*). Four replicates were made for each treatment.

Integrated disease management and soil solarization approach for control of F. solani in field condition

In the present investigation, effect of solarisation on soil temperature was evaluated by following the method of Sharma and Sharma (2002). This experiment was conducted in Crop Research Farm (CRF), The University of Burdwan, for two

successive years with four replications following Randomized Block Design (2013 and 2014). Each plot was 2 m X 2m. At first, plots were ploughed and irrigated to field capacity. Then plots were covered with thin transparent polythene sheets (0.25µm thick). To avoid border effect, one meter buffer zone was taken. This buffer zone was also under soil solarisation. The edge of polythene sheet was buried in soil so that it makes air tight. This solarization process was performed in the month of May. During the field trial, soil temperature was recorded in both plots i.e. solarized and non-solarized at the time of round about 3 pm under 10 cm depth of soil. After solarisation, plants were sown in the first week of June. According to *in vitro* performance of different integrated management components, the most efficient combinations were *Trichoderma harzianum*+ Bavistin+Neem and *Trichoderma viride* +Bavistin+Neem which were selected for field trials. These combinations were applied to both soil solarized and non- solarized plots.

RESULTS AND DISCUSSION

Disease control by application of potent antagonist

In vitro effect of potential antagonists against Fusarium solani

In vitro effect of *Trichoderma harzianum* and *Trichoderma viride* were tested to control growth of *F. solani* made in culture media. Both *Trichoderma harzianum* and *Trichoderma viride* isolates exhibited obvious antagonism against *F. solani*, causal organism of rot disease. *In vitro* effect of antagonist has been recorded in Table 1. *Trichoderma harzianum* mediated inhibition was 100% at concentration of metabolite 5ml/ 15 where as *Trichoderma viride* shows 87.78 % at the same concentration. From statistical point of view, it clarifies that *Trichoderma harzianum* can reduce the growth of test pathogen effectively.

Field trials with the application of potent antagonists to control the infection

Control of *F. solani* for root rot and stem decay disease of Amaranth cultivar was conducted in the field of CRF for two successive years (2013 and 2014) throughout the developmental stages with the application of *Trichoderma harzianum* and *Trichoderma viride.* These two antagonists showed positive performance in field condition also. *Trichoderma harzianum* treatment showed highest potentiality for the *Trichoderma viride* showed 31.69% and 29.17% in 2013 and 2014 respectively (Table 2). From statistical point of view, it is concluded that *Trichoderma harzianum* and *Trichoderma viride* could reduce the infection significantly. Disease control treatments using *Trichoderma* spp. also used in rot disease in Wheat (Foroutan, 2013), Chick pea (Akrami *et al*. 2013), Tomato (Karima and Nadia, 2012).

Significant control of ginger rhizome rot by use of different species *T.harzianum, T. aureoviride*, of *Trichoderma viz. T. viride* and *T. virens* was also reported by Ram *et al* (2000). The effect of various treatments with *T. viride* and *T.harzianum* under field condition showed significant influence in controlling rhizome rot of ginger and in increasing growth parameters and yield. *T. viride* when applied as seed treatment recorded that highest per cent disease control (84.9 %) reported by Kevimeo (2005). *T. viride* produced non-volatile substances which inhibited the growth of *F. solani,* causing rhizome rot of ginger by 70 % and 10 % respectively.

Control of disease by application of botanicals Screening of botanicals on the basis of growth inhibition of the pathogen

In vitro effect of different phytoextracts at different doses on the growth of *F. solani* were made in culture medium and recorded in Table 3. It is evident that botanicals have inhibitory effects on growth and development of *F. solani.* Among the phytoextracts, neem (*Azadirachta indica*) preparation shows highest inhibition (100.00%) at a dose of 4 ml/15 ml and 5 ml/15 ml growth medium. The inhibitory effect of remaining plant extracts was noted as *Allium sativum*, *Brassica nigra* (91.17%), *Carica papaya* (87.77%), *Phyllanthus emblica* (68.88%) and *Mimusops elengi* (58.88%) at a dose of 5 ml/15 ml growth medium.

Field trials with potent plant extracts to reduce infection

Control of *F. solani* for root rot and stem decay disease of Amaranth cultivar was conducted in the field of CRF for two successive years (2013 and 2014). From the statistical point of view, it is proved

Table . 1: *Trichoderma harzianum* and *Trichoderma viride* showing antagonist potentiality on growth of *Fusarium solani*.Data shows mean ± standard error of 5 replicates, indicate significant differences between the control and treated ones ($P < 0.05$).

Antagonist	Quantity of metabolites (ml)	Radial growth of pathogen(cm)	Growth inhibition of pathogen (%)
Trichoderma viride	1	7.9	12.22±0.08
	2	6.2	31.11±0.05
	4	2.4	73.33±0.11
	5	1.1	87.78±0.07
Trichoderma harzianum	1	7.1	21.11±0.06
	2	5.7	36.67±0.08
	4	1.8	80.00±0.05
	5	0	100.00±0.00
Control	0	9	

Table. 2: Field trials with the application of potent antagonist to control the infection. Data shows mean ± standard error of 5 replicates.

Trial year	Total number of plants	Number of infected plants	Percentage of infection	Treatments	Total number of plants	Number of infected plants	Infection (%)	Reduction of infection
2013	46	34	73.91	*Trichoderma harzianum*	37	14	37.83	36.07 ± 0.03
				Trichoderma viride	45	19	42.22	31.69 ± 0.01
2014	40	29	72.50	*Trichoderma harzianum*	48	15	31.25	41.28 ± 0.05
				Trichoderma viride	60	26	43.33	29.17 ± 0.01

that neem and garlic extracts were most effective for reduction of the disease incidence. These plant extracts also exhibited reduction of disease incidence in field condition for both the years. In this experiment, neem plant extract (42.70% and 43.86% in 2013 and 2014 respectively) showed superior result than the Garlic extract (23.73% and 30.63% in 2013 and 2014 respectively) in both the year. Result of field trials with neem and garlic extracts for the reduction of infection is presented in (Table 4). Neem aqueous extract treatment controlled many plant pathogen via metabolic changes in plants by the production of antioxidant defensive enzymes, phenols biosynthesis enzymes, and phenol accumulation in the plant system (Paul and Sharma, 2002; Guleria and Kumar, 2006; Aboellil, 2007; Farag *et al*. 2011). Damping off Disease of *Causuarina equisetifolia* was significantly inhibited by Neem extract and it can reduce 57% of the disease (Omokhua and Kalagbor, 2015). Neem extracts shows remarkable antifungal activity due to presence of active components like Azadirachtin, Nimbidin, Nimboidin, Nimbin etc.

Present finding also shows parallelism with earlier reports by several scientists for antifungal activity of garlic extract (Philippe *et al.* 2012; Anjorin *et al*. 2008; Sharma and Sharma, 2013; Kumar *et al.* 2014). Antifungal activity of *Mimusops elengi* was established by researchers (Ali *et al*. 2008; Satish *et al*. 2007; Prasad *et al*. 2012; Bobbarala *et al*. 2009; Pawar, 2011). *Carica papaya* and *Brassica nigra* has also good inhibitory potentiality against test pathogen. *Carica papaya* can inhibit 87.77% and *Brassica nigra* can inhibit 91.17% *in vitro* at

Table. 3: Effect of phytoextracts on growth of *Fusarium solani*. Data shows mean ± standard error of 5 replicates, indicate significant differences between the control and treated ones (P < 0.05).

Phytoextract	Quantity of extract (ml)	Radial growth of pathogen (cm)	Growth inhibition of pathogen (%)
Azadirachta indica	1	5.4	40.16 ± 0.17
	2	2.8	68.88 ± 0.04
	4	0	100 ± 0.00
	5	0	100 ± 0.00
Allium sativum	1	6.4	28.87 ± 0.04
	2	5.1	43.34 ± 0.04
	4	1.2	86.55 ± 0.10
	5	0	100 ± 0.00
Phyllanthus emblica	1	7.8	13.36 ± 0.05
	2	6.1	32.63 ± 0.40
	4	4.5	40.10 ± 10.01
	5	2.8	68.88 ± 0.02
Mimusops elengi	1	8.1	10.06 ± 0.03
	2	7.5	16.65 ± 0.02
	4	5.8	35.37 ± 0.18
	5	3.7	58.88 ± 0.02
Carica papaya	1	7.1	21.07 ± 0.02
	2	5.6	37.76 ± 0.03
	4	2.7	70.03 ± 0.03
	5	1.1	87.77 ± 0.03
Brassica nigra	1.	6.9	23.32 ± 0.04
	2	4.7	47.79 ± 0.04
	4	1.9	78.87 ± 0.03
	5	0.8	91.17 ± 0.04
Control	0	9	

Table. 4: Field trials with the application of effective phytoextracts on the reduction of infection caused by *Fusarium solani*. . Data shows mean ± standard error of 5 replicates.

	Untreated				Treated			
Trial year	Total number of plants	Number of infected plants	Percentage of infection (%)	Treatments	Total number of plants	infected plants (no.)	infection (%)	Reduction of infection (%)
2013	45	32	71.11	Neem	53	15	28.30	42.70 ± 0.12
				Garlic	38	18	47.36	23.73 ± 0.07
2014	28	21	75.00	Neem	45	14	31.11	43.86 ± 0.09
				Garlic	61	27	44.26	30.63 ± 0.07

Table. 5: *In vitro* effect of fungicidal chemical on growth of *Fusarium solani.* Data shows mean ± standard error of 5 replicates, indicate significant differences between the control and treated ones ($P < 0.05$).

Phytoextract	Dose (%)	Dry weight of mycelia (mg)	Growth inhibition of pathogen (%)
Mancozeb	0.01	161.32	14.36 ± 0.03
	0.05	140.57	25.36 ± 0.03
	0.1	118.72	36.99 ± 0.07
	0.5	105.48	44.03 ± 0.08
Bavistin	0.01	82.75	56.04 ± 0.09
	0.05	52.36	72.07 ± 0.07
	0.1	38.93	79.29 ± 0.10
	0.5	11.24	94.03 ± 0.06
Baynate	0.01	106.68	43.37 ± 0.07
	0.05	75.84	59.73 ± 0.10
	0.1	49.43	75.34 ± 0.07
	0.5	28.25	85.04 ± 0.05
Captan	0.01	136.86	27.34 ± 0.04
	0.05	100.40	46.66 ± 0.05
	0.1	69.75	62.93 ± 0.11
	0.5	42.25	77.56 ± 0.10
Griseofulvin	0.01	115.53	38.65 ± 0.12
	0.05	98.22	47.75 ± 0.05
	0.1	62.75	66.70 ± 0.10
	0.5	45.50	75.86 ± 0.06
Control	0	188.36	

the concentration of 5ml plant extracts/ 15ml PDA. Antifungal activity of *Carica papaya* was reported by several workers (Chávez-Quintal *et al*. 2011; Elie *et al*. 2015) Inhibitory effect of Mastard seeds on pathogenous fungi was reported (Rongai *et al*. 2012; Mejía-Garibay *et al*. 2015). Amlaki (*Phyllathus emblica*) fruit extract can reduce 68.89%. Inhibitory effect of Amlaki fruit (*Phyllathus emblica*) were

Table 6: Field trials with the application of effective fungicides on the reduction of infection caused by *Fusarium solani.* Data shows mean ± standard error of 5 replicates.

	UNTREATED				TREATED			
Trial year	Total number of plants	Number of infected plants	infection (%)	Treatments	Total number of plants	Number of infected plants	infection (%)	Reduction of infection (%)
2013	30	22	73.33	Bavistin	45	10	22.22	50.75 ± 0.45
				Baynate	41	19	46.34	27.01 ± 0.08
2014	35	27	77.14	Bavistin	36	8	22.22	54.95 ± 0.06
				Baynate	32	14	43.75	33.37 ± 0.05

Table. 7: Integrated disease management approach for control of *Fusarium solani in vitro.* Data shows mean ± standard error of 5 replicates, indicate significant differences between the control and treated ones ($P < 0.05$).

Treatment	Ratio	Radial growth of pathogen (cm)	Growth inhibition of pathogen (%)
Trichoderma harzianum+ Bavistin	1:1	6.3	29.98 ±0.68
	4:1	2.1	75.55 ± 0.63
Trichoderma viride +	1:1	7.1	21.47 ± 0.93
Bavistin	4:1	2.9	67.88 ± 0.61
Trichoderma harzianum+ Bavistin+	1:1:1	1.4	84.54 ± 0.44
Azadirachta indica	3:1:3	0	100 ± 0.00
Trichoderma viride +	1:1:1	1.9	78.89 ± 0.64
Bavistin+*Azadirachta indica*	3:1:3	0	100 ± 0.00
Trichoderma harzianum+ Bavistin+	1:1:1	2.9	67.69 ± 0.68
Allium sativum	3:1:3	0	100 ± 0.00
Trichoderma viride +	1:1:1	3.7	59.22 ± 0.39
Bavistin+ *Allium sativum*	3:1:3	0	100 ± 0.00
Control		9.00	0

Table . 8: Effects of soil solarisation and IDM schedule on *Fusarium solani*, causal organism of root rot and stem decay disease. Data shows mean ± standard error of 5 replicates, indicate significant differences between the control and treated ones ($P < 0.05$).

Treatment	2013			2014	
	Total number of plants	Total number of infected plants	Reduction of infection (%)	Total number of infected plants	Reduction of infection (%)
Solarized plot	25	10	39.78 ± 0.57	9	36.53 ± 0.85
Solarized plot+ *Trichoderma harzianum*+ Bavistin+ *Azadirachta indica*	25	0	72.32 ± 0.61	0	68.62 ± 0.91
Solarized plot+ *Trichoderma viride*+ Bavistin+*Azadirachta indica*	25	0	100 ± 0.00	0	100 ± 0.00
Non-solarized plot+ *Trichoderma harzianum*+ Bavistin+ *Azadirachta indica*	25	2	100 ± 0.00	3	100 ± 0.00
Non-solarized plot+ *Trichoderma viride*+ Bavistin+*Azadirachta indica*	25	4	64.57 ± 0.70	3	60.12± 0.15
Control (Non-solarized)	25	20		18	

reported by several workers (Shrestha and Tiwari, 2009; Devi *et al*. 2013; Malliga *et al*. 2015). But *M. elengi* shows less efficacy against test pathogen (58.88%).

The present findings therefore, provide promising approach towards controlling the pathogen and disease incidence by using different phytoextracts which are eco-friendly and harmless to the environment.

Disease control by the application of fungicides and antibiotic

Screening different fungicides and antibiotic on the growth inhibition of the pathogen

In vitro effect of different fungicides and antibiotic were recorded (Table 5) which indicated that fungicides and antibiotics have inhibitory effect on growth and development of *F. solani.* Bavistin showed highest inhibition against *F. solani* (94.03%)

at 0.5% concentration. Baynate (85.04%) Captan (77.56%) and Griseofulvin (75.86) exhibited growth inhibition of pathogen at 0.5% concentration. Mancozeb (44.03%) showed least potentiality to reduce growth of the pathogen among the all fungicide. Griseofulvin shows growth inhibition 75.86% and this finding is supported by Biancalana *et al*. (2008). Response of Captan was supported by Rathod *et al*. (2010), Dar *et al*. (2013), Gholve *et al*. (2014).

Field trials with effective fungicides for reduction of infection

To control the incidence of rot disease, different fungicides and antibiotic were tested *in vitro*. Among them, Bavistin and Baynate proved their superiority against *F. solani* which cause root rot and stem decay disease in *Amaranthus* cultivar. In this regard, those two fungicides were subjected for field trial in two consecutive years (2013 and 2014).The data were recorded in terms of percentage of reduction of infection and are represented in Table 6. Bavistin showed highest inhibition (50.75 % and 54.95 % in 2013 and 2014 respectively) whereas Baynate showed 27.01% and 33.37%in 2013 and 2014 respectively. Hence, Bavistin proved its efficacy in growth inhibition of test pathogen, *F. solani* (Table 6) . Satisfactory performance of bavistin was reported by Cromey *et al*. (2002), Aggarwal *et al*. (2005), Hossain and Bashar (2011), Sarita *et al*. (2014). Carbendazim (Bavistin) effects on hyphal tip cells of *Fusarium* sp. and displace mitochondria, disappeared spizenokorpers and ultimately mitotic metaphase arrested. Next to Bavistin, Baynate exhibit its efficacy and it can inhibit 85.04% of the tested pathogen. This result is in conformity with Girish and Bhat (2008), Singh *et al*. (2000), Chaurasia *et al*. (2014). Bavistin and Baynate converted into Methyl benzimidazole Carbamate (MBC) and suppress the effect of pathogenic fungi (Mehrotra and Aggrawal, 2003).

Integrated management approach towards in vitro growth of pathogen

Integrated disease management strategy includes combination of biological, chemical, cultural and physical treatment towards reducing plant diseases more effectively and ecofriendly (Rafael *et al*. 2015). Integrated management of rot disease was performed by the application of combination of potential antagonist (*Trichoderma viride* and *Trichoderma harzianum*), phytoextracts (*Azadirachta indica* and *Allium sativum*) and minimum dose of fungicide (Bavistin) *in vitro*. Different combinations were made viz. *Trichoderma harzianum*+ Bavistin, *Trichoderma viride* +Bavistin, *Trichoderma harzianum* + Bavistin+ *Azadirachta indica, Trichoderma harzianum* +Bavistin+*Azadirachta indica, Trichoderma harzianum* + Bavistin+ *Allium sativum* and *Trichoderma harzianum*+Bavistin+ *Allium sativum.* In the present investigation, *Trichoderma harzianum* + Bavistin+ *Azadirachta indica, Trichoderma viride*+Bavistin+*Azadirachta indica, Trichoderma harzianum* + Bavistin+ *Allium sativum* and *Trichoderma viride* + Bavistin+ *Allium sativum* combinations resulted 100% inhibition at dose of 3:1:3 ratio. It is also indicated that 4 ml cell free culture filtrate of *Trichoderma harzianum* and 1 ml 0.1% Bavistin resulted in 75.55 % inhibition of pathogen. Similarly, 4 ml cell free culture filtrate of *Trichoderma viride* and 1 ml 0.1% Bavistin resulted 67.88 % inhibition of pathogen. The percentage inhibition of the growth of the pathogen was calculated against control set. Four replications were made for each treatment and their mean values are presented in (Table 7).

Chandel and Sharma (2014) managed rot disease with application of different plant extracts and biocontrol agents both under *in vitro* and field conditions. Combination of Neem gold and *T. harzianum* controlled rot disease 72.72% in *Dianthus caryophyllus* and can be a good alternative source to chemicals in managing stem rot of carnation in integrated form with minimum impact on the soil and environment. Efficacy of IDM practices was reported by several workers (Pandey *et al*. 2009; Mahesh *et al*. 2010).

From the statistical point of view, it is established that growth of pathogen was effectively inhibited by integrated disease management and combination of *Trichoderma harzianum*+ Bavistin+ *Azadirachta indica* is referred as most effective combination.

Effect of soil solarization and integrated disease management on root rot and stem decay disease towards field condition

Soil solarization with integrated disease management was trial following randomize block design in four replication in 2013 and 2014. Both

solarized and non-solarized plots were treated with combination of *Trichoderma harzianum*+ Bavistin+ *Azadirachta indica* and *Trichoderma viride* +Bavistin+*Azadirachta indica* separately. Control set contain non-solarized soil without treatment.

Solarized plot with *Trichoderma harzianum*+ Bavistin+ *Azadirachta indica* and *Trichoderma viride*+Bavistin+*Azadirachta indica* result 100% inhibition of disease in both the years 2013 and 2014. In non- solarized plot, *Trichoderma harzianum*+ Bavistin+ *Azadirachta indica* shows disease reduction 72.32% and 68.62% in 2013 and 2014 respectively (Table 8). Combinations of *Trichoderma viride* +Bavistin+*Azadirachta indica* exhibit infection reduction 64.57 % and 60.12% in 2013 and 2014 respectively. So, it is concluded that soil solarization with integrated disease management offer reduction of infection. Four replications were made for each treatment and their mean values are presented in (Table. 8).

It has been reported that increase in growth response to soil solarisation may be related to physiological changes like photosynthetic activity, accelerated tissue development and delayed senescence occurs in late developmental state of plant growth in solarized soil. Normal plant hormonal balances were altered and stimulated in heat treated soil (Candido *et al*. 2008). Soil solarization has proved effective control of several soil borne pathogens (Kurt and Emir, 2004; Cimen *et al*. 2010). These combinations also increase fruit yield and total soluble solid in Strawberry plant. Soil solarization was effectively utilized by several researchers for the management of plant diseases (Chakraborty *et al*. 2009; Levy *et al*. 2014; Hussain and Simon, 2015; Yilmez *et al*. 2011).

Integrated disease management and soil solarization approach for control of F. solani in field condition

Physical and biological changes in soil are beneficial to plant health and growth. Soil solarization is a non chemical technique by which covering soil with transparent sheet to trap solar radiation to heat the soil more . Biological control by soil pathogens means other than thermal inactivation done by soil solarisation.

Integrated disease management (IDM) is a disease control approach that uses all available management strategies to maintain disease pressures below an economic injury threshold. The routine application of fungicides for insurance purposes is not appropriate, as it does not focus the proper attention on the real problem and can lead to resistance and potential environmental issues. Added benefits of IDM are that disease control is greater than that achieved individual method. In the present investigation, soil solarization integrated with Neem extract, Bavistin and *Trichoderma harzianum* provides 100% reduction of rot disease in Amaranth species in tropical plains of North-Eastern part of India.

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