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Utilization of some botanicals and bioagents for the management of anthracnose of Bell Pepper caused by *Colletotrichum capsici* (Syd.) Butler and Bisby

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Abstract
Anthracnose of bell pepper caused by *Colletotrichum capsici* incites significant yield losses all over the world. In the present study, the efficacy of four botanicals *viz. Allium sativum*, *Curcuma longa*, *Aloe barbadensis*, *Acorus calamus* at 10% concentration (w/v) and talc formulations of three bio-agents *viz. Trichoderma harzianum*, *T. viride*, *T. asperellum* at 5g/l of water were tested by the foliar application method for the management of the disease. Among the test botanicals, garlic was found effective in reducing the disease with average percentage disease index (PDI) of 5.70 percent, whereas, it was 28.55 percent in the control. In the test bio-agents, *T. viride* was found effective in reducing the disease with average PDI of 7.65 percent. Similarly, Garlic and *Trichoderma viride* were found effective in reducing the disease incidence, disease severity and percentage of infected fruits while Turmeric was found the least effective in managing the disease with disease incidence, diseases severity and percent infected fruit per plant of 19.32, 11.15 and 20.77 percent, respectively. In addition, the treatment with garlic resulted in maximum number of fruits/plant *i.e.*,18.16, while, it was minimum in control *i.e.*, 4.58. Among the all-treatments, maximum average yield was recorded in Garlic (370.79 q/ha), which is 76 percent higher than the control. Therefore, the use of garlic effectively manages the disease in a sustainable manner and increases the crop yield.

Keywords: Bell pepper, Anthracnose, *Colletotrichum capsici*, botanicals, bioagents, PDI, disease incidence, disease severity, Yield

Introduction
Bell Pepper (*Capsicum annuum* L var. *grossum* Sendt) commonly known as sweet pepper, capsicum or Shimla mirch, belongs to the family Solanaceae. Globally, India ranks fourth in the production of capsicum. In India, it is cultivated in an area of 46,000 ha with the total production of 327 thousand tons and an average productivity of 71 q/ha (Anonymous, 2017). Bell pepper is commercially cultivated in Himachal Pradesh, Jammu & Kashmir, Uttarakhand, Arunachal Pradesh and Darjeeling district of West Bengal during April-July. In Uttarakhand, it is grown in about 2.63 (000, ha) with the production of 15.70 thousand MT, (Anonymous 2017). The production of capsicum is significantly low in rainy season due to the facts that most of the low lands are planted with rice and farmers do not prefer the cultivation of the crop because of high failure due to anthracnose. Anthracnose, caused by *Colletotrichum capsici*, is one of the economically important diseases of bell pepper causing yield losses upto 50 percent (Pakdeevarpurn *et al.* 2005) [11]. The term anthracnose is derived from the *Greek* word which literally means “anthrax” or “Coal” and was first coined by Fabra and Dunal in 1833. The pathogen infects all the plant parts including leaves, stems and fruits. The disease is generally characterized by the appearance of circular or angular, and depressed sunken lesions which later turn into black colored spots with acervuli in concentric rings at maturity (Fig. 1) (Oo and oh 2016) [10].

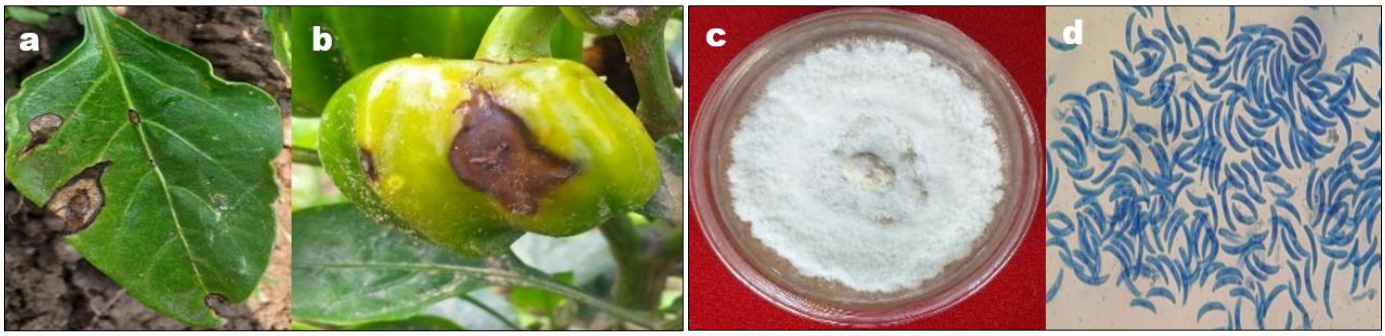

Fig 1: Illustrating the symptoms of Anthracnose and its causal agent. a-b, symptoms on leaf and fruit, respectively; **c**, colony of *Colletotrichum capsici*; d, conidia of *C. capsici* stained with cotton blue

Management of anthracnose of bell pepper has been relied on the use of chemical pesticides for many years. Despite controlling the disease and increasing the farmers' income, there are numerous negative impacts of these hazardous chemicals on the health of ecosystem. Thus, there is an urgent need to incorporate alternate management components including botanicals and bioagents that are effective in reducing the disease. Keeping in view of the above facts, the present study was carried out to check the efficacy of hot water seed treatment, various botanicals and bioagents against anthracnose of bell pepper caused by *Colletotrichum capsici.*

Materials and Methods

Experimental site

The field experiment was conducted at Vegetable Research and Demonstration Block, Plant Pathology laboratory, College of Horticulture, VCSG UUHF, Bharsar during *Kharif*, 2021. The location is about 58km away from Pauri city and situated at an altitude of 1900 meters above mean sea level. Geographical position of the experimental site lies between latitude 29^0 North and of 78^0 East Longitudes under Western Himalayan zone of Uttarakhand.

Evaluation of different treatments

The efficacy of various treatments including bio agents and botanicals were evaluated under field conditions against Anthracnose of bell pepper. The seedlings of bell pepper 'KTC-1' variety were planted in each of the several plots (2 m × 1.5 m) with row to row spacing of 60 cm and plant to plant spacing of 45 cm. Commercially available talc formulations of three bio agents *i.e., Trichoderma viride, T. asperellum* and *T. harzianum* @ 5g/ of water and aqueous extracts of four botanicals *viz., Allium sativum, Acorus calamus, Curcuma longa* and *Aloe barbadensis* (10% w/v) were used for foliar application. The untreated plants served as control. Spraying of the bioagents and botanicals was done after the first appearance of the disease. The experiment was conducted in a Randomized Complete Block Design (RCBD) with three replications for each treatment. Five plants from each treatment were randomly selected and tagged for observations on the disease. Disease incidence, percent disease index and percentage of infected fruits per plant from the tagged plants and fruit yield were recorded.

Disease incidence and severity in the natural field conditions were assessed following the standard 0-9 scale (Mayee and Datar, 1986), where, 0= no symptoms on leaf or fruit, 1= small irregular brown spots on less than 1% area of leaf or fruit, 3= dirty, brown and pin headed spots on 1-10% area of leaf or fruit, 5= dark brown to dirty black spots on 11-25% area of leaf or fruit, 7= dark brown, irregular to circular spots with blackish margin on 26-50% area of leaf or fruit and 9= dark brown, irregular to circular spots with blackish margin on 51% or above of leaf or fruit. Percent disease index or disease severity 75, 90 and 105 days after transplanting of the seedlings was calculated by using the formula proposed by Wheeler (1969). In addition to this, disease severity on fruits was also measured at harvest using the above mentioned scale.

The total number of infected fruits per plant at each picking from the selected plants was recorded. The percentage of infected fruits was estimated by adopting the grading formula given by Siddaramaiah *et al*., 1978. In order to estimate the yield gained by the application of bio agents and botanicals, average fruit yieldin treated and control plots was recorded and expressed in quintals per hectare. The percent yield gain was recorded using the following formula.

$$\text{Percent yield gain} = \frac{(\text{YTP}-\text{YCP})}{\text{YCP}} \times 100$$

Where,

YTP = Yield in treated plot, YDP = Yield in control plot.

The obtained data on the aforementioned parameters were statistically analysed using the online statistical software OPSTAT.

Results

Effect of bio test bio agents and botanicals on Percent disease index of anthracnose of bell pepper

The data on the effect of the test bio agents and botanicals on percent of disease index (PDI) of anthracnose of bell pepper 75, 90 and 105 days after transplanting is presented in Table-1. The recorded observations revealed that all the treatments were significantly superior over the control. The percent disease index increased over time irrespective of treatments. In addition, the mean performance of all the treatments ranged from 5.75 to 28.55 percent irrespective of the treatments and days after transplanting. At all the days after transplanting, the treatment with garlic showed maximum disease reduction with lowest mean PDI value of 5.70%. However among the bioagents, *Trichoderma viride* was found effective in reducing the disease that recorded mean PDI of 7.65%. This was followed by *T. harzianum* and vach extract, respectively.

Table 1: Effect of different treatments on percent disease index (PDI) after 75, 90, and 105 days after transplanting

Treatments	Per-cent disease index (PDI)			Avg. PDI	Percent disease reduction over control
	75days	90days	105days		
Control	26.98 ±0.29 (31.28)	28.47 ±0.26 (32.23)	30.22 ±0.30 (33.33)	28.55	-
Trichoderma viride	6.35*±0.07 (14.59)	7.78*±0.16 (16.19)	8.84*±0.09 (17.28)	7.65	73.55
Trichoderma asperellum	15.87*±0.06 (23.46)	16.65*±0.12 (24.07)	17.81*± 0.01 (24.95)	16.77	41.26
Trichoderma harzianum	11.11*±0.23 (19.46)	12.93*±0.04 (21.06)	13.83*±0.09 (21.82)	12.62	55.79
Garlic extract	4.76*±0.11 (12.59)	5.69*± 0.22 (13.79)	6.67*±0.05 (14.96)	5.70	80.04
Vach extract	12.68*±0.16 (20.85)	13.61*±0.12 (21.64)	14.75*±0.05 (22.57)	13.68	52.08
Aloe vera extract	18.52*±0.05 (25.48)	19.43*±0.12 (26.14)	20.11*±0.07 (26.18)	19.35	32.22
Turmeric extract	19.73*± 0.04 (26.36)	21.58*±0.20 (27.67)	23.79*±0.11 (29.18)	21.70	23.99
S.E. (d)	0.21	0.17	0.17	-	-
C.D. (p=0.05)	0.45	0.38	0.37	-	-

(), Value in parentheses are angular transformed; *, Values are significant at p=0.05

Effect of test bio agents and botanicals on other disease and yield parameters

The data of percent infected fruits/plant and disease (die back) incidence, disease severity at the time of harvest, average yield, number of fruit/plant and average fruit weight/plant are presented in Table-2. The observations on all the parameters showed significant differences between the treatments and the control. Significantly lower percentage of infected fruits per plant were observed in the treatment with garlic (10.51%) which was followed by *Trichoderma viride* (12.52%), *T. harzianum* (14.49%) and Vach extract (16.55%). While maximum percent of infected fruits per plant were recorded Turmeric (20.77%) and Aloe vera (19.77%), respectively. All the treatments under the study were found superior to the control.

Table 2: Effect of the treatments on disease and yield parameters at the time of harvest

Treatments	Percent infected fruit/plant	Disease Incidence	Disease Severity	No. of fruits /plants	Average fruit weight (g/plant)
Control	23.40±0.08 (28.92)	24.94 ±0.04 (29.95)	12.58 ±0.19 (20.76)	4.58±1.45 (11.87)	30.70 ±2.09 (33.59)
Trichoderma viride	12.52*±0.19 (20.71)	13.89*±0.28 (21.87)	7.46*±0.18 (15.84)	14.33*±1.88 (22.09)	49.89 *±3.63 (44.91)
Trichoderma asperellum	18.77*±0.09 (25.66)	17.64*±0.17 (24.82)	9.76*± 0.18 (18.19)	7.50*±1.69 (15.57)	41.92 *±3.53 (40.29)
Trichoderma harzianum	14.49*±0.10 (22.36)	15.49*±0.09 (23.92)	8.85*±0.05 (17.30)	11.16*±1.85 (19.29)	46.40 *±3.64 (42.89)
Garlic extract	10.51*±0.28 (18.90)	12.42*± 0.15 (20.62)	6.32*±0.10 (14.56)	18.16*±1.80 (25.13)	51.47 *±4.59 (46.99)
Vach extract	16.55*±0.15 (23.99)	16.46*±0.16 (23.92)	9.34*±0.09 (17.79)	8.66*±1.66 (16.85)	43.61*±3.76 (41.27)
Aloe vera extract	19.77*±0.14 (26.39)	18.59*±0.17 (25.53)	10.30*±0.19 (18.71)	6.33*±1.38 (14.31)	37.65 *±3.76 (37.76)
Turmeric extract	20.77*±0.10 (27.10)	19.32*±0.13 (26.06)	11.15*±0.07 (19.50)	5.41±1.32 (13.14)	35.68*±3.52 (36.98)
S.E. (d)	0.23	0.25	0.22	0.72	1.11
C.D. $_{(0.05)}$	0.51	0.54	0.47	1.52	2.32

(), Value in parentheses is angular transformed; *, Values are significant at p=0.05

In the similar manner, the data revealed on disease incidence ranged from 12.42 - 24.94%. A significantly lower percentage of incidence were observed in the treatment Garlic (12.42%) which was followed by *Trichoderma viride* (13.89%), *T. harzianum* (115.49%) and Vach extract (16.46%). While maximum percent of incidence was recorded in the control (24.94%), which was followed by Turmeric (19.32%), Aloe vera (18.59%), *T. asperellum* (17.64%). All the treatment under study was found superior to control for percent disease incidence. Similar trend was observed in case of disease severity in the treatment with garlic (6.32%) which was followed by *T. viride* (7.46%).

The observations recorded for number of fruits per plant ranged from 4.58-18.16 irrespective of the treatments. Significantly higher numbers of fruits per plant were observed in the treatment Garlic (18.16) which was found statistically at par with *Trichoderma viride* (14.33) and *T. harzianum* (11.16). While, minimum numbers of fruits per plant were recorded in the control (4.58), which was found statistically at par with Turmeric (5.41) and Aloe vera (6.33). Similarly, the maximum average fruit weight was recorded in the treatment Garlic (51.47 g), which was found statistically at par with *T. viride* (49.89 g), while, minimum fruit weight was observed in control (30.70 g).

The average total yield (q/ha) of bell pepper for the different test treatments is presented in Table 3. The maximum average yield was found in the treatment with Garlic extract (370.79 q/ha), which was followed by *Trichoderma viride* (342.84 q/ha), *Trichoderma harzianum* (319.88 q/ha) and Vach extract (304.08 q/ha). The minimum yield of 210.68 q/ha was found in the untreated control. However, the treatment with Turmeric (236.41 q/ha) was found least effective followed by Aloe vera (255.45 q/ha). In other words, the highest percent increase of yield over the control of 75.99 was observed in the treatment with Garlic, whereas, the lowest of 12.21 was found in the treatment with turmeric.

Table 3: Effect of bioagents and botanicals on average total yield (q/ha) of bell pepper

Treatments	1st Harvest	2nd Harvest	3rd Harvest	4th Harvest	Avg. Total yield	% increase over control
Control	42.04±0.93	58.34±2.53	56.08±1.87	54.22±1.29	210.68±2.49	-
Trichoderma viride	67.71±1.42	97.19±2.34	92.63±2.18	85.31±2.17	342.84±6.03	62.73
Trichoderma asperellum	54.30±0.88	81.75±1.01	77.89±1.92	74.35±0.95	288.29±4.64	36.84
Trichoderma harzianum	61.49±2.11	90.52±1.33	86.57±1.91	81.31±1.35	319.88±3.32	51.83
Garlic extract	74.31±2.14	106.93±4.55	97.11±2.34	93.11±1.47	370.79±3.17	75.99
Vach extract	56.71±0.84	85.93±1.45	83.44±1.68	78.39±1.09	304.08±2.10	44.33
Aloe vera extract	46.75±0.68	77.48±1.64	66.89±1.39	64.33±1.42	255.45±2.39	21.25
Turmeric extract	43.13±0.74	71.97±2.26	62.94±1.75	58.37±1.99	236.41±3.79	12.21
S.E.(d)	0.96	2.01	1.53	0.96	2.76	
C.D. (0.05)	2.05	4.30	3.28	2.07	5.91	

*, Values are significant at p=0.05

Discussion

In the present study, garlic extract was found most effective and showed minimum average percentage disease index (PDI) of 5.70% followed by *Trichoderma viride*7.65%, whereas, Tumeric was found least effective and showed maximum average percent disease index (PDI) of 21.70%. The antimicrobial activity of garlic is attributed to the biochemical compound known as allicin (Ankri and Mirelman, 1999) [3]. Various secondary metabolites from *Trichoderma* spp., act as antimicrobial or antifungal agents (Hermosa et al., 2014) [7]. Alves et al., (2015) found garlic and ginger extracts were efficient in controlling Anthracnose of bell pepper, where 6% garlic extract was the most effective. Similarly, Kamble *et al*., 2015 [8] found effective disease reduction with treatment of garlic (80.03%) followed by *Trichoderma viride* (73.20%). On the other hand, Shinde and Gawai (2014) [13] reported that turmeric and Aloe vera were least effective in reducing the disease over control. Recently, Vivekanand *et al*., 2018[14] observed that garlic extract followed by Neem extract were efficient in disease reduction, whereas among the test botanicals, *Trichoderma harzianum* followed by *Pseudomonas fluorescence* were reported to be superior. Bihari et al., (2018) [5] investigated the efficacy of different biocontrol agents and found *Trichoderma viride* was superior over others. However, Anand and Bhaskaran (2009) [2] recorded lower disease reduction with *Trichoderma viride* than *Pseudomanas flourescens*. This might be attributed to the difference in isolates which showed the different antagonistic ability. Dharajiya et al., (2017) [6] reported antifungal activity of methanolic and aqueous extracts of Aloe vera against some bacteria and fungi. Similarly in our investigation, the aqueous extract has reduced the disease incidence and severity. However, the reduction is not as efficient as that of extracts of garlic and Vach.

Aanthracnose results in a reduction in potential harvests and marketable fruit yield of pepper. In the present study, the percent increase of yield over the control was found in the range of 12.21 to 75.99 irrespective of the treatments. Moreover, garlic extract was also found in increasing the yield of the crop. Yield increase of bell peppers using these extracts was not reported so far. However, Ngullie *et al*., (2010) [9] have obtained 149.4% increase in yield over the control with *T. vriride* while working on anthracnose of chilli.

Conclusion

Our present study revealed that garlic extract was the best in reducing the percent infected fruits/plant, disease incidence and disease severity of anthracnose of bell pepper followed by *Trichoderma viride*. Similarly, the maximum average total yield was observed in in the treatment with garlic followed by *T. viride.* Therefore, it is advisable to include garlic in the integrated management of anthracnose of bell pepper, a severe pre and postharvest disease.

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Author contributions

SR has planned the experiment and helped in writing and editing the manuscript; NS has executed the experiment and written the manuscript.

Conflicts of interest

The authors have no competing interests to declare that are relevant to the content of this article.

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