



Microbial Physiology

Management of blight of bell pepper (*Capsicum annuum* var. *grossum*) caused by *Drechslera bicolor*



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ABSTRACT

Sweet or bell pepper is a member of the Solanaceae family and is regarded as one of the most popular and nutritious vegetable. Blight, in the form of leaf and fruit blight, has been observed to infect bell pepper crops cultivated at the horticulture farm in Rajasthan College of Agriculture, Udaipur, India. Based on disease severity, we attempted to curb this newly emerged problem using different fungicides, plant extracts, bio-control agents, and commercial botanicals against the fungus in laboratory and pot experiments. Bio-control agent *Trichoderma viride* and plant growth promoting Rhizobacteria (PGPR) isolate Neist-2 were found to be quite effective against bell pepper blight. All evaluated fungicides, botanicals, commercial botanicals, and bio-control agents *in vitro* were further studied as seed dressers and two foliar sprays at ten days interval in pot experiments. The combinations of Vitavax, PGPR isolate Neist-2, and Mehandi extract were found to be very effective against bell pepper blight followed by Vitavax, *T. viride*, and Mehandi extract used individually. All treatments in the pot experiments were found to significantly reduce seedling mortality and enhance plant biomass of bell pepper. Thus, these experimental findings suggest that a better integrated management of bell pepper blight could be achieved by conducting field trials in major bell pepper- and chilli-cultivated areas of the state. Besides fungicides, different botanicals and commercial botanicals also seem to be promising treatment options. Therefore, the outcome of the present study provides an alternate option of fungicide use in minimizing loss caused by *Drechslera bicolor*.

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Introduction

Sweet or bell pepper [*Capsicum annuum* var. *grossum* (L.) Sendt.], a member of the Solanaceae family, is regarded as one of the most popular and nutritious vegetable. It is native to

tropical South America, especially originating from Brazil, and is widely cultivated in central and south America, Peru, Bolivia, Costa Rica, Mexico, Europe, China, and India. In India, it is commercially cultivated in Tamil Nadu, Karnataka, Himachal Pradesh, and in some parts of Uttar Pradesh. In North India, it is commonly known as “Shimla Mirch” and is an

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important summer vegetable grown extensively in the mid hills of Himachal Pradesh, which is then supplied to regions in the plains. It is also grown in Rajasthan especially in Jaipur, Tonk, Sawaimadhopur, and Udaipur districts. It is not only rich in nutrients but also added as a natural colorant in food preparations. It has a wonderful combination of sweetness and tanginess with a crunchy texture. This may be eaten raw (sliced in salads) or cooked. In stews, bell peppers are also used for pickling in brine, baking, and in stuffing. Diced green or red bell peppers are sometimes mixed with sweet corn and other vegetables. The fruits are large with basal depression, inflated, red or yellow in color, with a thick flesh and a mild taste. Bell peppers are a rich source of vitamins, even more so than tomatoes, especially vitamins A, B₆, and C, calcium, and folic acid. The production of sweet pepper is very low in India as compared to the US, Holland, Italy, France, and other capsicum-growing countries over the world. This low production is mainly attributed to the infection of bell pepper crops with diseases caused by fungi, bacteria, viruses, and mycoplasmas, which drastically reduce potential yields. These pathogens also infect the fruits during transit and storage.¹ The major fungal diseases of capsicum crops are damping off (*Pythium aphanidermatum* and *Phytophthora* spp.), leaf spots (*Cercospora capsici* and *Alternaria solani*), anthracnose and ripe rot (*Colletotrichum capsici*), fruit rot and leaf blight (*Phytophthora* spp.), powdery mildew (*Erysiphe cichoracearum* and *Leveillula taurica*), early blight (*A. solani*), wilt (*Fusarium oxysporum*), frog eye rot (*Phaeoramularia capsicicola*), leaf spot (*Septoria lycopersici*), fruit spot (*Phoma destructiva*), stem rot (*Macrophomina phaseoli*), dry rot (*Sclerotium rolfsii*), and fruit rot (*Phomopsis* spp.). The post-harvest rots are caused by *Aspergillus terreus*, *A. candidus*, *A. niger*, *F. moniliforme*, *F. sporotrichoides*, *Paecilomyces variotii*, and *Penicillium corylophilum*.^{1–3} Sharma and Sohi reported a new disease in a chili cultivar NP-46A caused by *Drechslera* sp. during kharif season of 1977–78 causing leaf blight and fruit rot.⁴ They found symptoms on margins of leaf lamina, spots on stem, branches and fruits; and the symptoms on fruits were water soaked brown black areas. Seeds from the infected fruit have very poor germination capacity. In Varanasi (Uttar Pradesh) *D. bicolor* has been found on seeds of *C. annuum*.^{5,6} In Udaipur, Bell pepper blight, on the leaves and fruits (cv. Bombay red and Nun 3020 yellow), was first time noticed in August 2006 at Hi-tech Horticultural Polyhouse Farm, Rajasthan College of Agriculture (RCA).⁷ *D. bicolor*, the causative agent for this disease, infects the fruits of chilies, tomatoes, and brinjals.⁷ The initial symptoms of the disease include yellowing of young leaves near the tip. As the disease progresses, large straw or brown blight patches appear throughout the leaf thereby resulting in coalescence and drooping of leaf. The apical portion of the bell pepper fruit gets rotted with rapid discoloration, ultimately progressing to internal decay and complete deformation of fruits. The morphological features of *D. bicolor* are well defined by Ellis,⁸ which include conidiophores emerging singly or in small groups, straight or flexuous, sometimes swollen at the base, the upper part often repeatedly geniculate with large, dark sears, golden brown, up to 400 µm long, 5–10 µm thick, straight conidia or rarely curved slightly, cylindrical or rather broader in the middle and tapering toward the ends, rarely obclavate,

rounded at the apex, often truncated at the base, with 3–14 pseudosepta, 20–135 × 12–20 µm, mostly 40–80 × 14–18 µm with 5–9 pseudosepta, central cells of mature conidia often dark brown or smoky brown and sometimes quite opaque but each cell remains hyaline or very pale and is frequently cutoff by a very dark septum; hilum flat, dark, 3–5 µm wide. The taxonomy of “*Helminthosporium*” species is well studied by Alcorn.⁹ According to Misra et al., the *D. bicolor* grows well on PDA medium and produces profuse bottle green to whitish-gray colored aerial mycelium with a smooth and circular colony surface, which becomes brownish when aged.¹⁰ In another study, *D. bicolor* grown on PDA medium produced gray-black fluffy mycelium with maximum growth and sporulation observed at 25 ± 2 °C, and at hydrogen ion concentration of 6.5.⁷ In addition, the maximum mycelial growth of *D. bicolor* was recorded at 100% relative humidity followed by that at 80%, whereas an abundant sporulation was obtained only at 100% humidity level. The maximum spore germination was observed at 100% humidity followed by that at 80%.⁷

When the crop loss is high due to the pests and diseases, then intense efforts are needed avoid such a damage. The plant diseases are controlled to a great extent through scientific approaches such as the development of several potent pesticide/fungicide chemicals and ecofriendly management by biocontrol agents, phytoextracts, and botanicals. Moreover, continuous initiatives are being taken in an effort to obtain better and more specific agents against pathogens responsible for many uncured plant diseases. Of the seven fungicides tested, Vitavax was most effective against *D. bicolor* followed by thiram, mancozeb, and thiophanate methyl.⁷ The ecofriendly and cheap plant protection measures are essential for a sustainable crop production. As an alternative to fungicides, various plant extracts can be helpful in protecting crops and farm production from harmful diseases. In the same study, Didvaniya⁷ evaluated the antifungal activities of 15 different plant extracts against *D. bicolor* (blight of bell pepper). The maximum fungal growth inhibition was obtained with the extracts of neem leaves followed by that of lantana leaves and garlic cloves. Yadav and Gour¹¹ studied reduction in disease index of leaf stripe by barley using fungicides (carboxin, thiram, captan, mancozeb, carbendazim, and thiophanate methyl), botanicals (*Lantana camara* and *Azadirachta indica*), and bio-control agents (*Trichoderma harzianum* and *T. aureoviride*) as seed treatments and foliar sprays both in pot and field experiments. Maximum disease control was observed by carboxin (0.15%) in both pot and field experiments, followed by botanicals and bio-control agents. The use of carboxin (Vitavax) at concentration of 0.15% to treat seeds and for two foliar field applications (at day 35 and 56 after sowing) showed maximum efficacy in controlling disease control and increasing in the grain and fodder yield. Several studies have found better plant disease management using different approaches.^{12–16} In view of its recurrence in the emerging vegetable crops in Udaipur (Rajasthan), it was decided to conduct an experiment to manage the disease. For efficient management of recurring bell pepper blight, an *in vitro* study was conducted earlier.¹⁷ However, the efficacy of bio-control agents were not investigated; therefore, this study was undertaken to evaluate the *in vivo* effects of these

Table 1 – The details of plants and their parts used as botanicals.

Name of plant	Botanical name	Plant part used
Marigold	<i>Tagetes erecta</i>	Leaf
Kaner	<i>Dalium sp.</i>	Leaf
Lat jeera	<i>Achyranthes aspera</i>	Leaf
Aak	<i>Calotropis procera</i>	Leaf
Jarayan	<i>Lantana camara</i>	Leaf
Citronella	<i>Cymbopogon nardus</i>	Leaf
Keweda	<i>Pandanus odoratissimus</i>	Leaf
Lemon grass	<i>Cymbopogon flexuosus</i>	Leaf
Tulsi	<i>Ocimum sanctum</i>	Leaf
Laxmana	<i>Solanum indicum</i>	Leaf
Mehandi	<i>Lawsonia inermis</i>	Leaf
Mulatti	<i>Glycyrrhiza glabra</i>	Leaf
Garlic	<i>Allium sativum</i>	Clove
Onion	<i>Allium cepa</i>	Bulb
Turmeric	<i>Curcuma domestica</i>	Rhizome
Neem	<i>Azadirachta indica</i>	Leaf
Babool	<i>Acacia nilotica</i>	Leaf
Vilayati Babool	<i>Prosopis juliflora</i>	Leaf

bio-control agents and also of previously tested botanicals and fungicides.

Materials and methods

Different plant extracts, biocontrol agents, and fungicides (systemic and non-systemic) were assessed for their efficacy against *D. bicolor*, which is the causal agent of leaf blight disease of bell pepper both *in vitro*¹⁷ and *in vivo* (pot experiments) experiments.

Extraction of botanicals

The extracts of different plant parts (Table 1) and some market available botanicals (Table 2) were evaluated *in vitro* using poisoned food technique.¹⁸ Different parts of the plant were first washed with water, surface sterilized (2% sodium hypochlorite), followed by three washings with sterile distilled water, and then were kept in the sterilized covered beaker and allowed to air dry. The plant materials were weighed, crushed with 80% ethanol (1:1, w/v) in a Warring blender. The mixture was filtered through a double layered muslin cloth, the filtrate was evaporated, and the extract was diluted with sterile distilled water in a ratio of 1:1 (w/v). The concentration of this extract was considered as 100%; and it was used for further experiments.¹⁹ The following botanicals, which were found to be effective in an earlier *in vitro* study, were evaluated using *in vivo* pot experiments.¹⁷

In vitro testing of bio-control agents (dual culture)

Native isolates of the bio-control agents (Table 3) were isolated from rhizosphere soils. A loopful of the soil was thoroughly mixed with a drop of sterilized distilled water, placed in the center of an empty sterilized plate, and allowed to air dry for 5 min. This was over layered with 0.1% malt extract agar medium and allowed to solidify. The plates were incubated at 25 ± 1 °C and observed daily under stereo binocular microscope. The hyphae/colonies of the biocontrol agents were marked and picked into fresh malt extract agar slants. Thus, the cultures of *T. viride* and *T. harzianum* were obtained. The culture of *Actinomyces* spp. was obtained from a field at RCA using the serial dilution technique, and the culture of plant growth promoting rhizobacteria (PGPR) was obtained from the Department of Plant Pathology, RCA, Udaipur.

The comparative antagonistic potential as well as a possible mode of antagonism of two selected bio-control agents, one *Actinomyces* sp. and two PGPR isolates, against *D. bicolor* were studied *in vitro* (a total three bacterial species and two fungal species). The fungal antagonist and bacterial antagonist PGPR were tested on the pathogen *in vitro*. The antagonistic effect of *T. viride* and *T. harzianum* was evaluated *in vitro* by dual culture technique, and cultures of PGPR and *Actinomyces* sp. were evaluated *in vitro* by poisoned food technique (CFU concentrations were added in the medium as per the requirement) on potato dextrose agar medium. A 2-mm disk of the antagonists was removed from the edge of a 7-day-old culture and placed at one end of a 9-cm Petri dish over the PDA medium and at the opposite end, a 7-day-old disk of *D. bicolor* was also placed. The zone of inhibition between the two colonies was measured after five days. Each treatment was replicated four times. Percent inhibition of growth was calculated using the following formula²⁰:

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

where *I* is the % inhibition, *C* is the colony diameter in control fungi (mm), and *T* is the colony diameter in treated fungi (mm).

In vitro assaying of fungicides (poisoned food)

The effect of nine fungicides (systemic and non-systemic) (Table 4) against the pathogen at different concentrations was evaluated *in vitro* using poisoned food technique.¹⁷ For *in vitro* evaluation, the following fungicides were found quite effective; therefore, they were also evaluated *in vivo* using pot experiments.

Table 2 – The details of two commercial botanicals used.

Common name	Trade name	Chemical name	Manufacturing company
Zetron	Jatropha oil	Acaciarin 50 ppm	Vijaylaxmi Agro Chemicals, Hyderabad, India
Neem oil	Neem oil	Nimbidin 50 ppm	Vijaylaxmi Agro Chemicals, Hyderabad, India

Table 3 – The details of bio-control used are as under.

Bio-control agents	Detail	Suspension/CFU
Neist-1	PGPR isolate	6×10^6 , 5×10^7 , and 3×10^8
Actinomycetes	–	7×10^6 , 6×10^7 , and 5×10^8
Neist-2	PGPR isolate	5×10^6 , 4×10^7 , and 3×10^8
Neist-P	PGPR isolate	8×10^6 , 7×10^7 , and 6×10^8
<i>Trichoderma viride</i>	–	1: 3, 1:2 and 1:1
<i>T. harzianum</i>	–	1: 3, 1:2 and 1:1

Pot experiments (evaluation of promising plant extracts, biological agents, and fungicides)

On the basis of *in vitro* antifungal activity of plant extracts (percent concentrations), bio-control agents (CFU for PGPR and spore suspension of *Trichoderma*), botanicals (percent concentrations), and fungicides (ppm) were assessed as seed treatment and foliar sprays against *D. bicolor*. In case of *T. viride*, the spore concentration of the stock solution was maintained at 15×10^6 and the required suspension was prepared using sterilized water in a different ratio. The experiment was conducted in pots (30 cm \times 15 cm) in a completely randomized design and was replicated four times. The plants inoculated without any treatment were used as control for comparison. A small quantity of seeds was soaked in solutions of plant extracts, biological agents, and fungicides or in distilled sterilized water (as a control) for 1 h. The treated seeds were air dried and sown in pots. After germination of the seeds, thinning of seedlings was done to maintain five seedlings per pot. To obtain an infection index (disease development), a scale was devised to categorize plants into arbitrary classes based on a scale of 0–5 as follows:

Category	Percent leaf area infected
0	0 (no disease)
1	1–10 (small scattered leaf spots)
2	11–30 (a bit larger spots)
3	31–50 (large necrotic spots covering about 50% leaf area)
4	51–70 (about 70% leaf area blighted)
5	Above 70 (distortion and defoliation of diseased leaves)

Thirty days after inoculation, observations for leaf blight disease were recorded on individual plants using a rating scale. Percent disease index (PDI) was calculated using the

following formula²¹:

Percent disease index

$$= \frac{\text{Sum of all individual disease ratings}}{\text{Total numbers of plants assessed} \times \text{Maximum rating}} \times 100$$

While percent efficacy of each treatment over control (PEDC) was calculated using following formula:

Per cent efficacy of disease control

$$= \frac{\text{Infection index in control} - \text{Infection index in treatment}}{\text{Infection index in control}} \times 100$$

Statistical analysis

The coefficient of variation was calculated and analyzed for data collected from various experiments. The numerical values were transformed into square root values and percentages into arc sine values and subjected to statistical analysis. For laboratory and pot experiments, a completely randomized design was followed. The mean values were compared using ANOVA to determine differences for the efficacy among the treatments. *p*-Vale of ≤ 0.05 was considered as significant.

Results and discussion

The *in vitro* testing of bio-control agents (Table 5) revealed significant inhibition of mycelial growth of *D. bicolor* at all levels of suspension and CFU. The lowest growth of test pathogen was recorded in a PGPR isolate Neist-2 (13.94 mm) and also the highest % inhibition (86.54%) was recorded followed by PGPR isolate Neist-2 (14.93 mm and 85.01%) and *T. viride* (20.61 mm and 77.77%) *in vitro*. The different CFU levels of PGPR and *Actinomycetes* and spore suspension of *Trichoderma* were further tested, and Neist-2 and Neist-P were found to be very effective at 5×10^6 and 8×10^6 CFU, respectively (96.05% and 94.48% inhibition, respectively). *Actinomycetes* were effective at 7×10^6 CFU (51.11% inhibition). *T. viride* and *T. harzianum* were both found effective at 1:1 suspension level (87.97% and 82.63% inhibition, respectively). Similarly, bio-control agents *T. viride* and *T. harzianum* have been evaluated *in vitro* by

Table 4 – Details of fungicides.

Common name	Trade name	Chemical name	Manufacturing company
Carboxin	Vitavax 75% WP	5,6-Dihydro-2-methyl, 1,4 oxathiin-3-carboxanilide	Pesticides India Ltd., Udaipur, India
Carbendazim + Mancozeb	SAAF 12% + 63% WP	Sodium salt of aryl and naphthyl sulfonate	United Phosphorus Ltd., Mumbai, India
Iprodione + Carbendazim	Quintal 25% + 25% WP	3-(3,5-Dichlorophenyl)-N-(1-methylethyl)-2,4-dioxo-1-imidazolidine carboxamide + 2-(methoxycarbonylamino)-benzimidazole	Aventis Crop science Ltd., Mumbai, India

Table 5 – In vitro efficacy of different bio-control agents against mycelial growth of *D. bicolor*.

Bio-control agents	Colony diameters (mm)	% Inhibition between suspension/CFU	Suspension/CFU	Colony diameters (mm)	Inhibition (%)
Neist-1	67	25.31 (30.20)	6 × 10 ⁶	67.67	24.24 (29.50)
			5 × 10 ⁷	63.67	29.25 (32.74)
			3 × 10 ⁸	69.67	22.58 (28.37)
Actinomycetes	57.22	36.05 (36.90)	7 × 10 ⁶	44.00	51.11 (45.64)
			6 × 10 ⁷	59.00	34.44 (35.94)
			5 × 10 ⁸	68.67	23.69 (29.13)
Neist-2	13.94	86.54 (68.48)	5 × 10 ⁶	3.67	96.05 (78.54)
			4 × 10 ⁷	9.00	90.02 (71.58)
			3 × 10 ⁸	29.17	67.59 (55.30)
Neist-P	14.93	85.01 (67.22)	8 × 10 ⁶	5.00	94.48 (76.41)
			7 × 10 ⁷	10.50	88.04 (70.03)
			6 × 10 ⁸	29.30	67.45 (55.21)
<i>Trichoderma viride</i>	20.61	77.77 (61.87)	1:3	30.50	66.12 (54.40)
			1:2	20.50	77.22 (61.49)
			1:1	10.83	87.97 (69.71)
<i>T. harzianum</i>	20.54	77.35 (61.58)	1:3	26.00	71.12 (57.49)
			1:2	20.00	77.78 (61.88)
			1:1	15.63	82.63 (65.37)
Control (<i>D. bicolor</i>)	90.0	0.0	–	90.0	0.0
	SEm±	CD (p = 0.05)		SEm±	CD (p = 0.05)
Bio-control	0.719	2.057		0.588	1.688
Suspension/CFU	1.437	4.115		1.177	3.376

Average of four replications; numbers in parentheses are arcsine / angular transformed values; SEm± is standard error of mean. The spore suspension of *Trichoderma* was prepared as stock solution: sterilized water.

Pandey and Hussain²² for their antagonistic profiling against *Drechslera tetramera* isolated from *Capsicum frutescens*. The efficacies of both species of *Trichoderma* against the in vitro pathogen growth were comparable. *Trichoderma* was found to be a potent bio-agent in controlling the growth of *D. tetramera*. Similarly, Pandey et al.,²³ reported the potential of bio-agents viz. *T. harzianum* (Delhi), *T. harzianum* (Kanpur), *T. viride* (Delhi), *T. viride* (Kanpur), *Gliocladium virens* (Kanpur), and *Trichoderma hamatum* (Kanpur) against *Drechslera oryzae* in inhibiting the growth of *D. oryzae*. Maximum reduction (98.8%) was recorded in *T. harzianum* (Delhi) isolate followed by *T. harzianum* (Kanpur) (94.6%). Treatment of rice seeds with a spore suspension of *T. harzianum* (Delhi) was found to be significantly superior in enhancing maximum shoot and root lengths in 30-day-old seedlings. The foliar spray with crude extract of bio-agents effectively reduced the number of lesion from 13.49 to 3.15. The severity varied significantly from 14.1% to 58.1% in different treatments. Pre-treatment with crude extract of *Chaetomium globosum* and pre-inoculation with a mildly virulent strain of *Drechslera sorokiniana* induced resistance in wheat seedlings against spot blotch (*D. sorokiniana*) with reduction in disease severity by 94.9% and 49.9%, respectively. Both the inducing agents were applied as foliar spray: *Chaetomium* crude extract as 0.2% solution in water and *D. sorokiniana* mildly virulent strain at 104 conidia/mL. The phenolic and soluble protein contents in wheat leaves were increased by both the treatments. Percent disease index (PDI) showed highly negative correlation with phenol content ($r=0.96$) and soluble protein content ($r=-0.99$).²⁴

Pot experiments

Three highly effective fungicides from in vitro study,¹⁷ viz., Vitavax (Carboxin), Saaf (Carbendazim + Mancozeb), and Quintal (Iprodione + Carbendazim) were evaluated by seed treatment and two foliar sprays at ten days intervals in pot experiments against bell pepper blight (Table 6). In addition, 20 promising botanicals, viz., *sadabahar*, marigold, *kaner*, *lat jeera*, *jarayan*, *aak*, lemon grass, *keweda*, citronella, *tulsi*, *laxamana*, *Mehandi*, onion, garlic, turmeric, neem, *vilyati babool*, *babool*, neem oil, and *zatropin*¹⁷ were also used as seed dressers and two foliar sprays for evaluation in pot experiments against blight (Table 6).

The results of the pot experiments (Table 6) suggested a remarkable reduction in disease severity after application of these fungicides and botanicals to seed or via foliar sprays. The percent pre- and post-emergence mortality, percent yellowing of seedlings, and percent decayed seeds (Table 6) were significantly lowest in combined treatment of Vitavax (200 ppm), PGPR isolate Neist-2 (5 × 10⁶ spore suspension), and *Mehandi* extract (40%), followed by Vitavax (250 ppm), *T. viride* (1:3 spore suspension), and *Mehandi* extract (40%) used individually in comparison to the control where the percent pre-emergence mortality, percent post-emergence mortality, percent yellowing of seedlings, and percent decayed seeds were significantly highest and the percent emergence and percent healthy seedlings were significantly lowest. Table 6 depicts the percent emergence, which was found to be highest (85.18%) in combined seed treatment with Vitavax (250 ppm), PGPR isolate Neist-2 (5 × 10⁶ spore suspension), and *Mehandi* extract (40%)

Table 6 – Efficacy of different treatments on seedling and plant parameters.

Treatments	Concentration (%/ppm/spore suspension/CFU)	% Emergence	% Pre emergence mortality	% Post emergence mortality	% Yellowing of seedling	% decayed seed	% healthy seedling	Root length (mm)	Plant height (cm)	Dry biomass (mg)
Vitavax	250 ppm	73.86 (59.25)	26.14 (30.75)	20.11 (26.64)	21.11 (27.77)	26.14 (30.75)	57.84 (49.51)	19.75	12.15	35.25
Saaf	500 ppm	51.25 (45.72)	48.75 (44.28)	21.83 (27.85)	20.97 (27.25)	48.75 (44.28)	56.74 (48.88)	16.75	10.25	29.25
Iprodione	500 ppm	60.07 (50.81)	39.93 (39.19)	25.00 (30.00)	20.64 (27.02)	39.93 (39.19)	54.04 (47.32)	20.00	09.75	25.25
Mehandi	40%	53.75 (47.15)	46.25 (42.85)	20.80 (27.13)	32.61 (34.82)	46.25 (42.85)	46.31 (42.88)	18.25	09.75	30.50
Marigold	40%	43.73 (41.40)	56.27 (48.60)	34.55 (36.00)	31.37 (34.06)	56.27 (48.60)	33.83 (35.56)	16.25	09.25	23.50
Kaner	40%	38.71 (38.48)	61.29 (51.32)	41.90 (40.34)	22.57 (28.36)	61.29 (51.52)	34.92 (36.22)	16.00	11.25	26.00
Lat jeera	40%	51.25 (45.72)	48.75 (44.28)	26.40 (30.92)	26.83 (31.20)	48.75 (44.28)	46.30 (42.88)	20.25	11.75	30.20
Jarayan	40%	38.71 (38.48)	61.29 (51.52)	45.46 (42.39)	22.27 (28.16)	61.29 (51.52)	31.94 (34.41)	19.25	11.75	29.25
Aak	40%	38.74 (38.49)	61.26 (51.51)	38.61 (38.41)	22.41 (28.25)	61.26 (51.51)	38.26 (38.21)	20.50	08.25	27.25
Lemon grass	40%	56.27 (48.60)	43.73 (41.40)	24.42 (29.61)	29.03 (32.60)	43.73 (41.40)	45.82 (42.60)	18.00	05.75	29.50
Keweda	40%	41.22 (39.94)	58.78 (50.06)	42.30 (40.57)	23.59 (29.05)	58.78 (50.00)	33.48 (35.36)	19.50	10.25	25.75
Citronella	40%	36.20 (36.99)	63.80 (53.01)	37.77 (37.92)	24.05 (29.37)	63.80 (53.01)	37.77 (37.92)	16.25	04.25	27.50
Tulsi	40%	53.75 (47.15)	46.25 (42.85)	27.86 (31.86)	25.38 (30.25)	46.25 (42.85)	46.35 (42.91)	16.25	10.50	28.50
Laxamana	40%	43.73 (41.40)	56.27 (48.60)	36.85 (37.37)	28.51 (32.27)	56.27 (48.60)	34.04 (35.70)	12.25	08.00	25.00
Mulatti	40%	41.22 (39.94)	58.78 (50.06)	36.47 (37.15)	21.29 (27.48)	58.78 (50.06)	41.35 (40.02)	17.75	08.25	23.25
Onion	40%	51.25 (45.72)	48.75 (44.28)	29.06 (32.62)	26.71 (31.21)	48.75 (44.28)	43.84 (41.46)	19.25	05.25	22.00
Garlic	40%	51.25 (45.72)	48.75 (44.28)	29.44 (32.86)	26.83 (31.20)	48.75 (44.28)	43.49 (41.26)	17.25	07.25	29.50
Turmeric	40%	41.24 (39.96)	58.76 (50.04)	33.22 (35.20)	32.72 (34.89)	58.76 (50.04)	33.48 (35.35)	16.25	10.25	32.50

Table 6 – (Continued)

Treatments	Concentration (%/ppm/spore suspension/CFU)	% Emergence	% Pre emergence mortality	% Post emergence mortality	% Yellowing of seedling	% decayed seed	% healthy seedling	Root length (mm)	Plant height (cm)	Dry biomass (mg)
Neem	40%	41.22 (39.94)	58.78 (50.06)	27.18 (31.43)	23.36 (28.90)	58.78 (50.06)	49.01 (44.43)	17.75	08.75	30.50
Vilayati babool	40%	53.77 (47.16)	46.23 (42.84)	21.00 (27.28)	18.05 (25.14)	46.23 (42.84)	60.34 (50.97)	16.00	06.25	33.50
Babool	40%	48.75 (44.28)	51.25 (45.72)	30.54 (33.55)	25.36 (30.24)	51.25 (45.72)	43.78 (41.43)	15.25	08.25	29.25
Neem Oil	40%	43.74 (41.41)	56.26 (48.59)	28.34 (32.16)	28.58 (32.32)	56.26 (48.59)	42.69 (40.80)	11.25	05.25	30.50
Zatropin	40%	51.25 (45.72)	48.75 (44.28)	31.24 (33.98)	34.38 (35.90)	48.75 (44.28)	33.99 (35.66)	14.25	10.25	29.25
Neist-2	5 × 10 ⁶ CFU	46.25 (42.85)	53.75 (47.15)	26.81 (31.18)	27.00 (31.30)	53.75 (47.15)	45.83 (42.61)	15.25	09.25	22.25
Actinomycetes	7 × 10 ⁶ CFU	51.25 (45.72)	48.75 (44.28)	24.45 (29.64)	26.83 (31.20)	48.75 (25.53)	48.27 (44.01)	13.25	11.25	24.75
<i>Trichoderma viride</i>	1:3 spore suspension	46.25 (42.85)	53.75 (47.15)	26.81 (31.18)	29.81 (33.09)	53.75 (47.15)	42.99 (40.97)	12.25	09.75	27.25
Vitavax + Neist-2	250 ppm + 5 × 10 ⁶ CFU	81.42 (64.47)	18.58 (25.53)	15.32 (23.04)	16.85 (24.24)	18.58 (25.53)	67.71 (55.37)	19.75	12.25	35.25
Vitavax + Mehandi	250 ppm + 40%	71.28 (57.59)	28.72 (32.41)	17.45 (24.69)	17.45 (24.69)	28.72 (32.41)	64.96 (53.70)	19.75	11.75	34.25
Mehandi + Neist-2	40% + 5 × 10 ⁶ CFU	73.86 (59.25)	26.14 (30.75)	15.11 (22.87)	15.11 (22.87)	26.14 (30.75)	69.75 (56.63)	19.75	11.75	35.25
Vitavax + Neist- 2 + Mehandi	250 ppm + 5 × 10 ⁶ CFU + 40%	85.18 (67.36)	14.82 (22.64)	08.65 (17.11)	17.47 (24.71)	14.82 (22.64)	73.70 (59.15)	19.50	12.25	34.75
Untreated control	–	36.20 (36.99)	63.80 (53.01)	31.00 (33.83)	27.92 (31.89)	63.80 (53.01)	40.85 (39.73)	8.25	05.25	20.50
Inoculated control	–	21.09 (27.34)	78.91 (62.66)	49.21 (44.55)	34.44 (35.93)	78.91 (62.66)	12.59 (20.78)	5.75	04.25	17.00
SEm±		1.32	1.32	2.11	2.21	1.32	2.52	0.629	0.46	0.55
CD (p=0.05)		3.70	3.70	5.91	6.20	3.70	7.08	1.765	1.28	1.56

Average of four replications; numbers in parentheses are arcsine / angular transformed values; SEm± is standard error of mean. The spore suspension of *Trichoderma* was prepared as stock solution: sterilized water.

Table 7 – Efficacy of different treatments on percent disease incidence (PDI) of *D. bicolor* and percent efficacy of disease control (PEDC) of treatments against *D. bicolor*.

Treatments	Concentration (%/ppm/spore suspension/CFU)	PDI 2007	PDI 2008	Mean PDI	PEDC 2007	PEDC 2008	Mean PEDC
Vitavax	250 ppm	32.3 (34.6)	34.1 (35.7)	33.2 (35.2)	66.1 (54.4)	64.4 (53.4)	65.2 (53.9)
Saaf	500 ppm	39.3 (38.8)	41.7 (40.2)	40.5 (39.5)	58.7 (50.0)	56.5 (48.8)	57.6 (49.4)
Iprodione	500 ppm	43.7 (41.4)	46.2 (42.8)	45.0 (42.1)	54.1 (47.3)	51.9 (46.1)	53.0 (46.7)
Mehandi	40%	47.0 (43.3)	49.2 (44.5)	48.1 (43.9)	50.6 (45.3)	48.7 (44.2)	49.6 (44.8)
Marigold	40%	55.3 (48.0)	56.7 (48.8)	56.0 (48.4)	41.9 (40.3)	40.9 (39.8)	41.4 (40.1)
Kaner	40%	55.6 (48.2)	56.8 (48.9)	56.2 (48.6)	41.6 (40.1)	40.8 (39.7)	41.2 (39.9)
Lat jeera	40%	58.1 (49.7)	60.4 (51.0)	59.3 (50.3)	39.0 (38.6)	37.0 (37.5)	38.0 (38.1)
Jarayan	40%	67.3 (55.1)	69.5 (56.5)	68.4 (55.8)	29.4 (32.8)	27.5 (31.7)	28.4 (32.2)
Aak	40%	68.9 (56.1)	71.2 (57.6)	70.0 (56.8)	27.7 (31.8)	25.8 (30.5)	26.7 (31.1)
Lemon grass	40%	69.1 (56.2)	70.3 (56.9)	69.7 (56.6)	27.4 (31.6)	26.7 (31.1)	27.1 (31.4)
Keweda	40%	73.1 (58.8)	77.2 (61.5)	75.2 (60.1)	23.2 (28.8)	19.5 (26.2)	21.3 (27.5)
Citronella	40%	70.2 (56.9)	73.5 (59.0)	71.8 (58.0)	26.3 (30.9)	23.4 (28.9)	24.8 (29.9)
Tulsi	40%	73.6 (59.1)	74.0 (59.3)	73.8 (59.2)	22.7 (28.5)	22.9 (28.6)	22.8 (28.5)
Laxamana	40%	71.8 (57.9)	74.7 (59.8)	73.2 (58.8)	24.6 (29.8)	22.1 (28.0)	23.4 (28.9)
Mulatti	40%	71.0 (57.4)	74.1 (59.4)	72.5 (58.4)	25.4 (30.3)	22.8 (28.5)	24.1 (29.4)
Onion	40%	72.8 (58.6)	77.2 (61.4)	75.0 (60.0)	23.6 (29.0)	19.6 (26.3)	21.5 (27.7)
Garlic	40%	75.0 (60.0)	75.0 (60.0)	75.0 (60.0)	21.2 (27.4)	21.9 (27.9)	21.6 (27.7)
Turmeric	40%	73.2 (58.8)	73.2 (58.8)	73.2 (58.8)	23.1 (28.8)	23.7 (29.1)	23.4 (29.0)
Neem	40%	74.4 (59.6)	76.6 (61.1)	75.5 (60.3)	21.9 (27.9)	20.1 (26.6)	21.0 (27.3)
Vilayati babool	40%	74.2 (59.5)	78.9 (62.7)	76.6 (61.1)	22.1 (28.0)	17.8 (24.9)	19.9 (26.5)
Babool	40%	74.3 (59.5)	76.5 (61.0)	75.4 (60.3)	22.0 (28.0)	20.3 (26.8)	21.1 (27.4)
Neem Oil	40%	66.8 (54.8)	69.7 (56.6)	68.3 (55.7)	29.9 (33.1)	27.4 (31.6)	28.6 (32.3)
Zatropin	40%	67.6 (55.3)	69.2 (56.3)	68.4 (55.8)	29.0 (32.6)	27.9 (31.9)	28.5 (32.2)
Neist-2	5 × 10 ⁶ CFU	69.9 (56.7)	71.2 (57.5)	70.5 (57.1)	26.7 (31.1)	25.8 (30.5)	26.2 (30.8)
Actinomycetes	7 × 10 ⁶ CFU	73.3 (58.9)	74.5 (59.6)	73.9 (59.2)	23.1 (28.7)	22.3 (28.2)	22.7 (28.5)
<i>Trichoderma viride</i>	1:3 spore suspension	44.2 (41.7)	44.4 (41.8)	44.3 (41.7)	53.6 (47.1)	53.7 (47.1)	53.6 (47.1)
Vitavax + Neist-2	250 ppm + 5 × 10 ⁶ CFU	45.2 (42.2)	44.8 (42.0)	45.0 (42.1)	52.5 (46.5)	53.2 (46.8)	52.9 (46.7)
Vitavax + Mehandi	250 ppm + 40%	49.3 (44.6)	49.5 (44.7)	49.4 (44.7)	48.2 (44.0)	48.4 (44.1)	48.3 (44.0)
Mehandi + Neist-2	40% + 5 × 10 ⁶ CFU	73.8 (59.2)	78.5 (62.4)	76.2 (60.8)	22.6 (28.4)	18.2 (25.2)	20.3 (26.8)
Vitavax + Neist-2 + Mehandi	250 ppm + 5 × 10 ⁶ CFU + 40%	25.4 (30.3)	26.9 (31.2)	26.2 (30.8)	73.3 (58.9)	72.0 (58.0)	72.6 (58.5)

Table 7 – (Continued)

Treatments	Concentration (%/ppm/spore suspension/CFU)	PDI 2007	PDI 2008	Mean PDI	PEDC 2007	PEDC 2008	Mean PEDC
Untreated control	–	93.1 (74.8)	94.4 (76.3)	93.8 (75.5)	2.2 (8.5)	1.6 (7.2)	1.9 (7.9)
Inoculated control	–	95.3 (77.4)	96.0 (78.5)	95.6 (77.9)	0	0	0
SEm±		0.217	0.477	0.262	0.268	0.540	0.301
CD (p = 0.05)		0.610	1.340	0.732	0.753	1.516	0.841

Average of four replications; numbers in parentheses are arcsine / angular transformed values; SEm± is standard error of mean. The spore suspension of *Trichoderma* was prepared as stock solution: sterilized water.

followed by Vitavax alone seed treatment (73.86%); the percent pre-emergence mortality was found to be lowest (14.82%) in this combination followed by Vitavax alone (26.14%). Seed treatment with the above-mentioned combination also had lowest percent post-emergence mortality (08.65%), lowest percent yellowing of seedling (17.47%), lowest percent decayed seed (14.82%), and highest percent healthy seedling (73.70%) followed by Vitavax alone (20.11%, 21.11%, 26.14%, and 57.84%, respectively). Plant characteristics such as root length, plant height, and dry biomass were significantly highest in combined treatment of Vitavax (200 ppm) and *Mehandi* extract (40%), followed by Vitavax (250 ppm) and *Mehandi* extract (40%) individually (19.50 mm, 12.25 cm, and 34.75 mg, respectively) followed by Vitavax (250 ppm), *T. viride* (1:3 spore suspension), and *Mehandi* extract (40%) individually in comparison to the control where the root length, plant height, and dry biomass were significantly lowest.

In the experiment, the percent disease index in both years 2007 and 2008 and the mean of both years were found to be lowest in the combined application of Vitavax (250 ppm) with PGPR isolate Neist-2 (5×10^6 spore suspension) and *Mehandi* extract (40%) (25.4%, 26.9%, and 26.2%, respectively) followed by Vitavax (250 ppm), *T. viride* (1:3 spore suspension), and *Mehandi* extract (40%) individually in comparison to the control where the PDI was significantly highest (Table 7). The PEDC calculated accordingly showed that the combined application of Vitavax (250 ppm) with PGPR isolate Neist-2 (5×10^6 spore suspension) and *Mehandi* extract (40%) (73.3%, 72.0%, and 72.6%, respectively) followed by Vitavax (250 ppm), *T. viride* (1:3 spore suspension), and *Mehandi* extract (40%) individually in comparison to the control where the PDI was significantly highest (Table 7). The above-mentioned treatments served as best seed dressers and two foliar sprays at ten days interval to better control the disease. Vitavax (250 ppm), *T. viride* (1:3 spore suspension), and *Mehandi* extract (40%) when used individually also showed effectiveness against the disease.

Similarly, in case of chili diseases, faytolan (*C. capsici*)²⁵; mancozeb, thiophanate methyl, captafol, ziram, and carbendazim (*C. capsici*)²⁶; blitox-50, ridemil MZ and bavistin 50 WP (*Alternaria* sp.)²⁷; carbendazim²⁸; mancozeb (*A. alsenata*)²⁹ as best fungicide(s). Yadav³⁰ evaluated fungicides against *D. graminea* and found Vitavax to be the best followed by thiram and captan. Maximum percent efficacy of disease control was observed in Vitavax treatments in both pot and field

experiments. Jadeja et al.,²⁹ also found Vitavax as the most effective fungicide followed by thiram, mancozeb, and thiophanate methyl against *D. bicolor*. The results obtained by these workers are very close to the findings of the present investigation and several studies have found methods to better manage plant diseases using different management approaches.^{12–16,31–36}

Conclusion

This was a preliminary study on the management of bell pepper blight in which different fungicides, botanicals, and commercial botanicals were evaluated against the pathogen *in vitro* and in pot experiments. The present results can pave the path toward the better integrated management of bell pepper blight by conducting field trials in the major bell pepper- and chili-cultivated areas of the state. Besides fungicides, different botanicals and commercial botanicals showed promising results in our investigation. A wide availability of these botanicals as an alternate option of fungicides will certainly help the farmers involved in cultivating bell peppers. Bio-control agents are also important entities as they are ecofriendly, and they can be studied against *D. bicolor*. Hence, the integration of fungicides, botanicals, bio-control agents, and resistant bell pepper cultivars would help positively manage this new threat in bell pepper cultivation. Thus, we conclude that the fungicides Vitavax, botanicals, marigold, *lat jeera*, lemon grass, *Mehandi*, onion, neem, and neem oil and the bio-control agents *T. viride* and PGPR isolate Neist-2 were quite effective in controlling bell pepper blight. The combinations of Vitavax, PGPR isolate Neist-2, and *Mehandi* extract were found excellent against the bell pepper blight, and also worked satisfactorily when applied. These bio-control agents, botanical, and fungicides are proved to be good for recovery of the plant and for production of good biomass and yield. Thus, the use of botanicals and bio-control agents provide an option for management of blight.

Conflicts of interest

The authors declare no conflicts of interest.

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