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Bioefficacy of Potential Fungicides and Biocontrol Agents for Efficient Management of Soybean Pod Blight

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ABSTRACT: To know the field efficacy of any new fungicide molecules and bioagents, there is a need to evaluate fungicides *in vitro* which provides useful and preliminary information regarding efficacy against pathogen within a shortest period of time and therefore, serves as a guide for field testing. An experiment was conducted to evaluate the bioefficacy of fungicides and bioagents against *Colletotrichum truncatum* causing pod blight of soybean at Deparment of Plant Pathology, UAS Raichur. Among seven non-systemic fungicides, systemic fungicides and combi fungicides evaluated against *C. truncatum* under *in vitro*, copper oxychloride 50 % WP, hexaconazole 5 % EC, propiconazole 25% EC, carbendazim 50 % WP, thiophanate methyl 70 % WP, and all the combi fungicides were found highly effective in inhibiting the complete mycelial growth of the pathogen with cent per cent mycelial inhibition. Among bioagents evaluated, *T. harzianum* and *T. asperellum* showed maximum mycelial growth inhibition. Effective fungicides and bioagents were selected for integrated management of soybean pod blight under field condition. Seed treatment with *T. harzianum* (5g/ kg seed) followed by spray with tebuconazole + trifloxystrobin (0.05 %) at 60 DAS and spray of *Pseudomonas fluorescens* (1%) at 75 DAS found significantly effective in managing the disease with highest yield and benefit cost ratio.

Keywords: Soybean, pod blight, Fungicides, Per cent inhibition, management.

INTRODUCTION

Soybean (*Glycine max* (L.) Merrill) is a miracle crop renowned for its high protein and oil content. It is also known as "Golden Nugget." It has the highest protein content of all leguminous crops in the Leguminaceae family. Because of its high and virtually unrivalled protein (36 per cent), carbohydrate (35 per cent) (17 per cent of which is dietary fibre) with edible vegetable oil content (19 per cent), minerals (5 per cent), and a variety of other components including vitamins, soybean holds a premier position as a world crop (Ajay *et al*., 2011). Soybean protein quality is comparable to that of meat, milk, and eggs. Because it is the cheapest type of protein, it is known as "poor man's meat." Soybean is the richest, cheapest, and easiest source of high-quality protein and lipids, with a wide range of applications as a meal and and industrial products commonly known as ‘Wonder Crop’. In India, the crop is grown over an area of 11.33 million hectares and production of 13.79 million tonnes with a productivity of 1217 kg/ ha (Anon., 2019). In Karnataka, soybean is grown over an area of 0.29 million hectares with a production of 0.29 million tonnes by recording productivity of about 1008 kg/ha (Anon., 2019).

Plant health is an important factor in ensuring a profitable soybean harvest. One of the most significant obstacles to enhancing soybean yield is its sensitivity to a wide range of illnesses caused by fungi, bacteria, viruses, and nematodes. Anthracnose, caused by Colletotrichum truncatum, caused yield losses of up to 16 per cent in soybeans, followed by frog eye spot (15 per cent), rust (10-90 per cent), downy mildew (8 per cent), and powdery mildew (25-50 per cent) (Sinclair, 1992). Anthracnose (pod blight) produced by *C. truncatum* has been described as the primary restriction in successful soybean cultivation among the major fungal diseases (Khan and Sinclair, 1992 and Mittal *et al.,* 1993). In 1917, Korea was the first country to report the sickness. Backman *et al*. found a 26% yield loss (1982).

C. truncatum also caused 30% yield losses, according to Mahmood and Sinclair (1992).

C. truncatum can infect soybeans at any stage of development, but especially from bloom to pod fullness. Symptoms of the illness emerge as irregularly shaped brown lesions on the stem, pods, leaves, and petioles during the early reproductive stages (Sinclair and Backman, 1989). Necrosis of leaf veins, leaf rolling, petiole canker, and premature defoliation are some of the foliar symptoms. The pod blight phase, on the other hand, is the most harmful (Vyas *et al*. 1997). The pods develop a reddish brown mark that eventually turns black. Fruiting bodies (Acervulli) on infected pods look like miniature pin cushions surrounded by minute blackish brown setae, and infected pods eventually dry up prematurely, resulting in shrivelled and rotten seeds. The plants affected with anthracnose disease are significantly shorter, with fewer pod and seeds with reduced seed weight compared to non- affected plants. Infected seeds often show brown discolouration. Sample with up to 30 per cent seed discoloration had up to 70 per cent seed infection of *C. truncatum* (Hepperely *et al*., 1983).

Integrated plant disease management (IPDM) is a decision-based strategy that entails the coordinated employment of numerous tactics to optimise pathogen control while being environmentally and economically sustainable. It aids in the construction of sturdy structures and the growth of healthy plants. As a long-term answer, promotes bio-based sickness management. Encourages the implementation of more ecologically friendly control procedures, lowering the management's environmental risk. As a result, many fungicides were tested against the pathogen in an attempt to manage the condition.

MATERIAL AND METHODS

A. In vitro evaluation of fungicides and bioagents

Following the poison food technique, the efficacy of seven non-systemic fungicides at concentrations of 0.1, 0.2, and 0.25 per cent, seven systemic fungicides at concentrations of 0.05, 0.1, and 0.15 per cent, and seven combi fungicides at concentrations of 0.1, 0.2, and 0.3 per cent were assessed. By employing a dual culture technique using potato dextrose agar (PDA) as the baseline culture media, the potential species of Trichoderma, Psuedomonas, and Bacillus were investigated in vitro against *C. truncatum.*

Poison food technique

Under aseptic circumstances, the fungicide-affected PDA was poured (15-20 ml/plate) into sanitised Petri plates (90 mm). Each treatment was repeated three times with the same concentration. All treatment plates were inoculated/seeded aseptically after PDA had solidified on Petri plates by putting a 5.0 mm uniform, mycelial disc obtained from a 7- 9 day old culture of *C. truncatum* multiplied on agar plates in the centre (Nene and Thapliyal, 1993). Petri plates containing plain PDA without any fungicide were infected with the test pathogen's 5.0 mm disc and kept as an appropriate untreated control. All of the treatment (inoculated) and control Petri plates were then incubated in a BOD incubator at 27 10°C until the control plates were completely covered with the test's mycelial growth. The efficacy of the fungicides was expressed as per cent inhibition of mycelial growth over control which was calculated by using the formula of Vincent (1947).

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

Where,

I = Per cent inhibition fungal growth

C = Average diameter of fungal growth in control

T = Average diameter of fungal growth in treatment under *in vitro* condition against *C. truncatum.*

Dual culture technique

The efficacy of bioagents was tested using a dual culture approach. *C. truncatum* discs (5 mm) were placed in the middle of one corner of various Petri plates containing solidified PDA media, while discs of *Trichoderma* spp were placed in the opposite corner of various Petri plates. Each bioagent disc was stored in three Petri plates with the disc of *C. truncatum.* A 24 hour old *P. fluorescens* and *Bacillus* spp. culture loop was introduced at 2 cm exactly opposite the pathogen on each plate (Dennis and Webster, 1971). The bioagents efficacy was measured in per cent inhibition of mycelial growth over control, which was computed using the previously cited Vincent (1947) methodology.

B. Integrated management of soybean pod blight

Those fungicides (systemic, non-systemic, and combi products) and bio-control agents (fungal and bacterial) that were found effective against *C. truncatum* during in vitro evaluation were used in an integrated approach for the management of anthracnose/pod blight of soybean under field conditions with the goal of developing an eco-friendly, economical, and holistic disease management practice.

The field trials were conducted during *Kharif* 2020 at Agricultural Research Station, Bidar. The experiment was laid out in randomized block design with three replications and eight treatments including chemicals, bioagents and untreated control. The detail of treatment combinations are given here under.

Experimental details

Variety: JS- 335; Plot size: 3 m × 2.4 m

Treatments	Treatment details
T_1	Seed treatment (ST) with carboxin + thiram (combi product) (2g/ kg seed) followed by spray with thiophanate methyl (0.1 %) at 60 and 75 DAS
T_2	Seed treatment (ST) with carbendazim + mancozeb (combi product) (2g/ kg seed) followed by spray with thiophanate methyl (0.1 %) at 60 and 75 DAS
T_3	Seed treatment (ST) with carboxin + thiram (combi product) (2g/ kg seed) followed by spray with tebuconazole + trifloxystrobin (combi product) (0.05 %) at 60 and 75 DAS
T_4	Seed treatment (ST) with pyraclostrobin + thiophanate methyl (combi product) (2ml/ kg seed) followed by spray with tebuconazole + trifloxystrobin (combi product) (0.05 %) at 60 and 75 DAS
T_5	Seed treatment (ST) with *Trichoderma harzianum* (5g/ kg seed) followed by spray with *Pseudomonas fluorescens* (1 %) at 60 and 75 DAS
T_6	Seed treatment (ST) with *Trichoderma harzianum* (5g/ kg seed) followed by spray with carbendazim (0.1 %) at 60 and 75 DAS
T_7	Seed treatment (ST) with *Trichoderma harzianum* (5g/ kg seed) followed by spray with tebuconazole + trifloxystrobin (combi product) (0.05 %) at 60 DAS and spray of *Pseudomonas fluorescens* (1 %) at 75 DAS
T_8	Control

Ten plants in each plot were scored for disease severity using 0-9 scale given by Mayee and Datar (1986).

Category	Description
0	No lesions/spots/discolouration
1	1 % area covered with lesions/spots/discolouration
3	1.1-10 % area covered with lesions/spots/discolouration
5	10.1-25 % area covered with lesions/spots/discolouration
7	25.1-50 % area covered with lesions/spots/discolouration
9	>50 % area covered with lesions/spots/discolouration

Data were converted into per cent disease index (PDI) by using the formula of Wheeler (1969). Each treatment was harvested separately and yield per plot was recorded further benefit: cost ratio was computed

$$PDI = \frac{\text{Sum of individual disease ratings}}{\text{No. of observations assessed}} \times \frac{100}{\text{Maximum disease rating}}$$

RESULTS AND DISCUSSION

A. Efficacy of non-systemic fungicides against C. truncatum

All seven non-systemic fungicides were shown to be significantly more effective than the control at preventing *C. truncatum* mycelial growth. Copper oxychloride was shown to be significantly superior to all other treatments with (100 per cent) mycelial inhibition at (0.2 and 0.25 per cent) and 98.35 per cent mycelial inhibition at (0.1 per cent), for a total of 99.45 per cent mycelial inhibition. Propineb was the next best fungicide, with (94.88 per cent) mycelial inhibition at (0.25 per cent) and a mean mycelial inhibition of 90.11 per cent, which was shown to be considerably superior. Copper hydroxide (88.74 per cent), captan (86.98 per cent), and mancozeb (86.79 per cent) are all in the same ballpark. Chlorothalonil (75.13 per cent) had the lowest mycelial inhibition, followed by zineb (79.63 per cent) at a concentration of 0.1 per cent, with mean mycelial inhibition of 77.90 and 81.47 per cent, respectively (Table 1 and Plate 1).

Three fungicides, copper oxychloride, probineb, and copper hydroxide, were the most effective with greatest mycelial inhibition among the seven fungicides studied. The non-systemic fungicides performed effectively in preventing mycelial growth at varied concentrations (0.25 per cent) and were found to be considerably superior than propineb and chlorothalonil at (0.2 per cent), while the remaining fungicides were statistically on par between these two concentrations. Fungicides at a concentration of 0.25 per cent were statistically equivalent to all other fungicides at a concentration of 0.1 per cent.

The outcomes of the current study are in accordance with the results of Rajashree *et al.* (2020) who reported that propineb found most effective at all the three concentration and was significantly superior over other fungicides, followed by copper oxychloride, mancozeb and captan.

Table 1: Efficacy of non-systemic fungicides against *C. truncatum*.

Sr. No.	Common name	Inhibition (%)			
		Concentration (%)			Mean
		0.1	0.2	0.25	
1.	Copper oxychloride 50 WP	98.35 (84.60)	100.00 (90.00)	100.00* (90.00)**	99.45 (88.20)*
2.	Copper hydroxide 53.8 WP	86.85 (68.74)	88.88 (70.52)	90.49 (72.04)	88.74 (70.43)
3.	Captan 70 WP	84.72 (66.99)	87.24 (69.07)	89.00 (70.63)	86.98 (68.90)
4.	Mancozeb 80 WP	85.17 (67.35)	86.42 (68.38)	88.77 (70.42)	86.79 (68.72)
5.	Propineb 70 WP	87.23 (68.66)	88.22 (69.15)	94.88 (76.89)	90.11 (71.57)
6.	Zineb 75 WP	79.63 (63.18)	81.82 (64.77)	82.95 (65.62)	81.47 (64.52)
7.	Chlorothalonil 75 WP	75.13 (60.09)	76.69 (61.13)	81.87 (64.80)	77.90 (62.01)
Mean		85.30 (68.52)	87.04 (70.43)	89.71 (72.91)	
		S.Em ±		C.D at 1%	
Fungicides (F)		0.75		2.87	
Concentration (C)		0.49		1.88	
F × C		1.30		4.97	

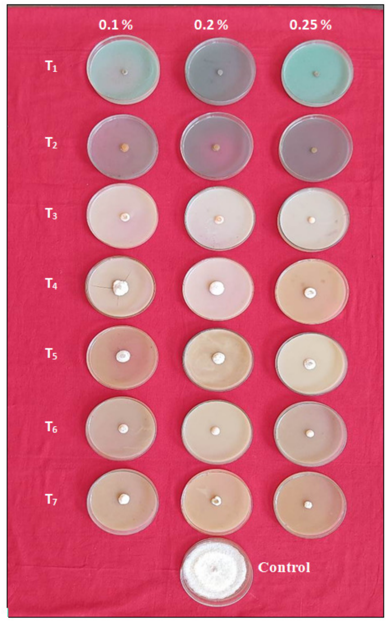
* Mean of three replications

**Values in the parentheses are arc sine transformed values

According to Jagtap *et al*. (2012), mancozeb inhibited mycelial development by 88.45%, followed by propineb, while chlorothalonil inhibited mycelial growth by the least amount (40.01 per cent). Captan inhibited mycelial development by 82.1 per cent, according to Nagaraj (2013). (0.3 per cent). Mancozeb was shown to be the most effective by Mestha (1996) and Hegde (1998). Copper oxychloride and mancozeb, according to Laxman (2006), are the most effective fungicides.

Copper oxychloride is a protective contact fungicide with a broad spectrum of activity. Copper, which has a strong bonding affinity for amino acids and carboxyl groups, interacts with protein and functions as an enzyme inhibitor in target organisms, accounts for its efficiency. By interacting with the sulphahydral groups of some enzymes, copper destroys spores. Spores aggressively collect copper, which inhibits spore germination even at low concentrations.

The next best fungicide is propineb, which interferes at different locations in the metabolism of the fungi on several points of the respiration chain. This multi-site mode of action of propineb prevents development of resistance in the fungi (Thind *et al*., 2018).



T_1: Copper oxychloride 50 WP; T_2: Copper hydroxide 53.8 WP; T_3: Propineb 70 WP; T_4: Chlorothalonil 75 WP; T_5: Zineb 75 WP; T_6: Mancozeb 80 WP; T_7: Captan 70 WP

Plate 1. Effect of non-systemic fungicides against *Colletotrichum truncatum*

B. Efficacy of systemic fungicides against C. truncatum

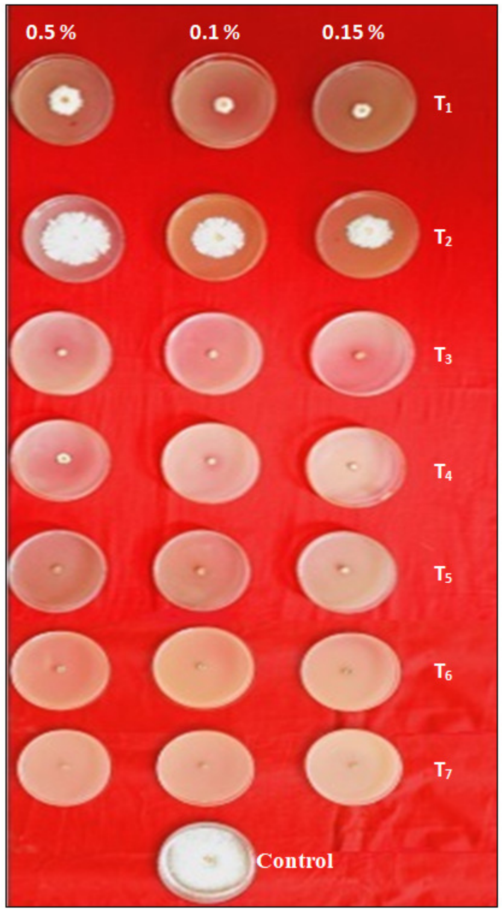
Seven different systemic fungicides were evaluated under laboratory conditions to test their efficacy against *C. truncatum* at three different concentrations (0.05, 0.1 and 0.15 %) through poison food technique as detailed in 'Material and Methods'. The per cent inhibition of mycelial growth of the pathogen over control was calculated. Based on the mycelial growth, the effect of systemic fungicides on inhibition of *C. truncatum* are furnished in Table 2.

Table 2: Efficacy of systemic fungicides against *C. truncatum*.

Sr. No.	Common name	Inhibition (%)			
		Concentration (%)			Mean
		0.05	0.1	0.15	
1.	Difenconazole 25 EC	62.72 (52.37)	77.90 (61.97)	83.83* (66.30)**	74.81 (60.21)
2.	Triadimefon 25 WP	35.50 (36.57)	45.60 (42.48)	52.52 (46.44)	44.54 (41.83)
3.	Tebuconazole 25 EC	87.16 (69.00)	100.00 (90.00)	100.00 (90.00)	95.72 (83.00)
4.	Hexaconazole 5 EC	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
5.	Propiconazole 20 EC	100.00 (90.00)	100.00 (90.000)	100.00 (90.00)	100.00 (90.00)
6.	Carbendazim 50 WP	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
7.	Thiophanate methyl 70 WP	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
Mean		83.63 (73.99)	89.07 (79.21)	90.91 (80.39)	
		S. Em ±		CD at 1%	
Fungicides (F) Concentration (C) F × C		0.13 0.09 0.23		0.50 0.33 0.87	

* Mean of three replications
**Values in the parentheses are arc sine transformed values

The results of the mycelial growth and per cent inhibition tests demonstrated that all seven systemic fungicides were much better than the control at inhibiting *C. truncatum* mycelial growth. Hexaconazole, propiconazole carbendazim, and thiophanate methyl, four fungicides, demonstrated (100 per cent) inhibition at all three concentrations, statistically substantially superior to the other treatments and comparable to each other. Tebuconazole showed 100% inhibition at 0.1 and 0.15 per cent concentrations, but 87.16 per cent mycelial inhibition at 0.05 per cent concentration, with a mean mycelial inhibition of 95.72 per cent, followed by difenconazole (83.83 per cent) at 0.15 per cent concentration, with a mean mycelial inhibition of 74.81 per cent. The fungicide triadimefon showed minimum mycelial inhibition of 35.5 per cent at the concentration of (0.05 %), with mean mycelial inhibition of 44.54 per cent (Plate 2).



T_1: Difenconazole 25 EC; T_2: Hexaconazole 5 EC; T_3: Triadimefon 25 WP; T_4: Tebuconazole 25 EC; T_5: Propiconazole 20 EC; T_6: Carbendazim 50 WP; T_7: Thiophanate methyl 70 WP

Plate 2. Effect of systemic fungicides against *Colletotrichum truncatum.*

In the interaction between fungicides and concentrations, systemic fungicides tested at 0.15 per cent were found to be highly effective in inhibiting mycelial growth and significantly superior to difenconazole and triadimefon at (0.1 per cent) and difenconazole, triadimefon, and tebuconazole at 0.05 per cent, with the remaining fungicides being on par with each other between these two concentrations.

The current findings are consistent with those of Laxman (2006), who investigated eight systemic fungicides in vitro against *C. truncatum* and found that, with the exception of tricyclazole and hexaconazole, all of them completely inhibited mycelial growth at concentrations of 0.05 and 0.1 per cent. According to Kulkarni and Raja (2019), propiconazole, carbendazim, and thiophanate methyl showed complete inhibition at all three concentrations (0.05, 0.1, and 0.15 per cent), followed by hexaconazole (100 per cent) at (0.1 and 0.15 per cent) and difenconazole (100 per cent) at (0.05, 0.1, and 0.15 per cent) (0.15 per cent). According to Agam *et al*. (2019), carbendazim, tebuconazole, and propiconazole inhibited mycelial growth to the greatest extent (0.1 per cent), followed by hexaconazole, and difenconazole.

Nagaraj (2013) reported that, propiconazole showed (100 %) mycelial inhibition at 0.05, 0.1 and 0.15 per cent concentrations. Carbendazim and difenconazole exhibited complete inhibition at (0.1 %) and minimum mycelial inhibition was found in triadimefon (22.10 %). Shovan *et al*. (2008) observed that propiconazole completely inhibited the mycelial growth of *C. truncatum.* Varaprasad (2000) and Madhusudhan (2002) also obtained the similar results.

Hexaconazole, propiconazole, and tebuconazole, among other triazole fungicides, are effective inhibitors of ergo sterol production, the primary membrane sterol of fungus. They inhibit C-14 alpha demethylase, a cytochrome P450-dependent enzyme required for the conversion of lanosterol to ergo sterol. Ergosterol is a component of the cell wall in many fungi, and its absence causes irreversible damage to the cell wall, resulting in the death of the fungal cell. Nene and Thapliyal investigated the effects of triazole fungicides on the sterol biosynthesis pathway in fungi (1993).

C. Efficacy of combi fungicides against C. truncatum

Seven different combi products were evaluated for their efficacy against *C. truncatum* at three different concentrations (0.1, 0.2 and 0.3 %) through poison food technique (Table 3).

Table 3: Efficacy of combi fungicides against *C. truncatum.*

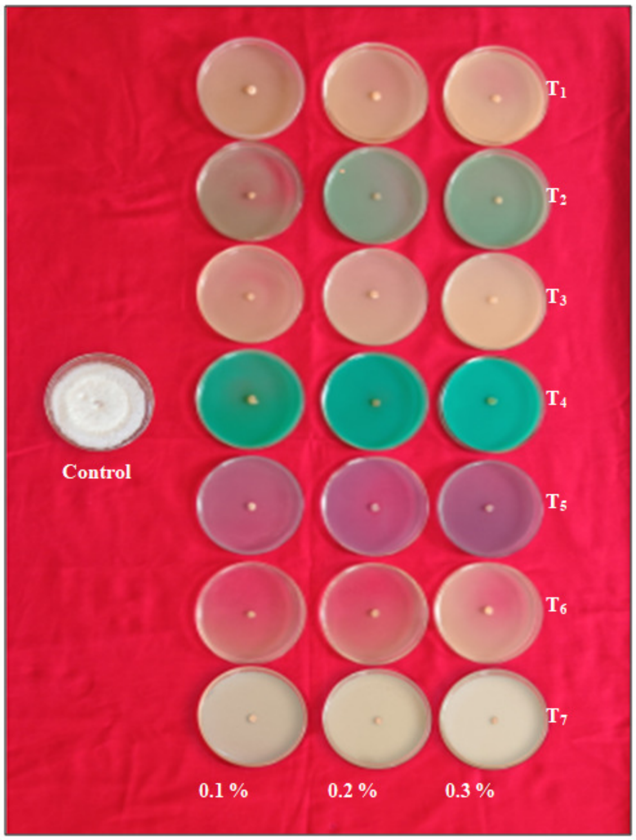
Sr. No.	Common name	Inhibition (%)			
		Concentration (%)			Mean
		0.1	0.2	0.3	
1.	Carboxin 37.5 % + Thiram 37.5 % 75 WP	100.00 (90.00)	100.00 (90.00)	100.00* (90.00)**	100.00 (90.00)
2.	Carbendazim 12 % + Mancozeb 63 % 65 WP	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
3.	Tebuconazole 50 % + Trifloxystrobin 25 % 75 WG	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
4.	Pyraclostrobin 5 % + Thiophanate methyl 45 % FS	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
5.	Iprodione25 % + Carbendazim 25 % 50 WP	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
6.	Captan70 % + Hexaconazole 5 % 75 WP	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
7.	Zineb 68 % + Hexaconazole 4 % 72 WP	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
Mean		100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	
		S.Em ±		C.D at 1%	
Fungicides (F)		NS		NS	
Concentration (C)		NS		NS	
F × C		NS		NS	

* Mean of three replications

**Values in the parentheses are arc sine transformed values

NS: Non significant

All the combi fungicides were found significantly superior over the control in inhibiting the mycelial growth of *C. truncatum* and also, all the combi products were found effective against *C. truncatum* by inhibiting the complete growth of the pathogen with cent per cent mycelial inhibition, at all the three different concentrations (Plate 3).



T_1: Zineb 68 % + Hexaconazole 4 % 72 WP; T_2: Carbendazim 12 % + Mancozeb 63 % 65 WP; T_3: Captan 70 % + Hexaconazole 5 % 75 WP; T_4: Pyraclostrobin 5 % + Thiophanate methyl 45 % FS; T_5: Carboxin 37.5 % + Thiram 37.5 % 75 WP; T_6: Tebuconazole 50 % + Trifloxystrobin 25 % 75 WG; T_7: Iprodione25 % + Carbendazim 25 % 50 WP

Plate 3. Effect of combi fungicides against *Colletotrichum truncatum.*

The outcomes of the present study are in line with the results obtained by Nagaraj (2013) who found that iprodione 25 % + carbendazim 25 %, tebuconazole 50 % WP + trifloxystrobin 25 % WG and carboxin 37.5 % + thiram 37.5 % WP inhibited complete growth of the pathogen at (0.1 %). Kulkarni and Raja (2019) reported that carbendazim 12 % + mancozeb 63 % WP showed (100 %) mycelial growth inhibition at 0.1 and 0.2 per cent concentration.

Shashikumara (2019) reported that, carbendazim 12 % + mancozeb 63 % WP, carboxin 37.5 % + thiram 37.5 % WP and tebuconazole 50 % WP + trifloxystrobin 25 % WG recorded (100 %) at (0.1 and 0.2 %). Rajashree *et al.* (2020) also reported that carbendazim 12 % +

mancozeb 63 % WP recorded (100 %) mycelial growth inhibition at (0.3 %) and captan 70 % + hexaconazole 5 % WP recorded (99.25 %) at (0.3 %). Roopadevi (2014) found that iprodione 25 % + carbendazim 25 % recorded the maximum mycelial growth inhibition (99.5 %) of the fungus subsequently zineb 68 % + hexaconazole 4 % (99.3 %).

Products that contain two or more active components in a single formulation are known as combination products. Various suitable active ingredients with various modes of action are mixed and employed to combat the target infection. Because more than one active ingredient is present, more than one "target" inside pathogen cells is disrupted, boosting the fungicide's efficacy. As a result, the fungicides in combination products are more effective.

D. Bioefficacy of bioagents against C. truncatum

Five different biocontrol agents *viz.,* two strains of fungal (*Trichoderma* sp.) and three strains of bacterial bioagents (two *Bacillus* sp and one *Pseudomonas fluorescens*) were evaluated to explore their possible antagonistic potential against *C. truncatum* by following dual culture technique as detailed in "Material and Methods".

All five biocontrol agents were shown to be effective at greatly inhibiting the pathogen's growth, according to the findings of the study. Among which, *T. harzianum* strain (TUREF59) showed maximum mycelial inhibition of 66.67 per cent, which is on par with T. asperellum strain (Ta- 1) with mycelial inhibition of 66.32 per cent which were significantly superior over other bioagents, subsequently *P. fluorescens* strain (BGREB73) with mycelial inhibition of 60 per cent. *B. subtilis* strain (SE76) showed the least mycelial inhibition (52.01 per cent), followed by *B. thuringiensis* strain (BTG4), which had the same mycelial inhibition (53.47 per cent) (Table 4 and Plate 4).

Table 4: Efficacy of bioagents against *C. truncatum*.

Sr. No.	Bioagents	Per cent inhibition
1.	*Trichoderma harzianum* (TUREF59)	66.67* (54.74)**
2.	*Trichoderma asperellum* (Ta- 1)	66.32 (54.53)
3.	*Bacillus subtilis* (SE76)	52.01 (46.16)
4.	*Bacillus thuringiensis* (BTG4)	53.47 (47.00)
5.	*Pseudomonas fluorescens* (BGREB73)	60.00 (50.78)
S.Em ±		0.95
C.D at 1%		3.94

* Mean of four replications

** Values in the parentheses are arc sine transformed values

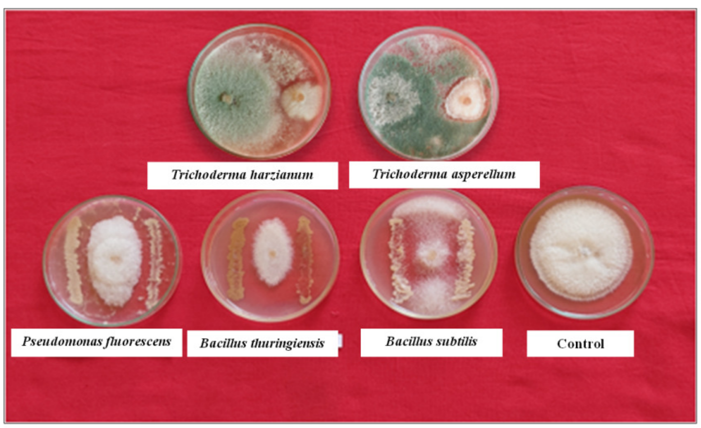


Plate 4. Effect of bioagents against *Colletotrichum truncatum.*

P. fluorescens and *T. harzianum*, according to Jagtap *et al*. (2012), had 69.33 and 64.58 per cent mycelial inhibition, respectively. *T. harzianum* and T. viride were found to impede the growth of *C. lindemuthianum* by Gupta *et al*. (1991). Varaprasad (2000) discovered that *Trichoderma sp* is efficient against *C. dematium*, but Laxman (2006) discovered that *Trichoderma sp* is effective against *C. truncatum.*

Kale and Barhate (2016) reported that *T. viride* and *T. harzianum* exhibited 78.88 and 77.04 per cent mycelial growth inhibition, respectively followed by *T. hamatum* and *P. fluorescens.*

According to Shashikumara (2019), among fungal bioagents, *T. harzianum* was found to be the most effective in inhibiting the mycelial growth of the test pathogen (80.22 per cent), followed by *T. viridae* (72.55 per cent) and bacterial bioagents *P. fluorescens* and *B. subtilis* (46.44 and 36 per cent, respectively). *P. fluorescens* and *T. harzianum* both inhibited mycelial growth (89.11 and 88 per cent, respectively), according to Agam *et al*. (2019). According to Roopadevi (2014), *T. harzianum* had the highest mycelial inhibition (95.38 per cent) and *B. subtilis* had the lowest mycelial growth inhibition (19.11 per cent).

Principal mechanisms *viz.,* mycoparasitism, antibiotics such as, trichodermin, suzukacillin and lamethicin produced by *T. harzianum* may influence morphological or physiological sequences leading to its successful penetration, could release cellular enzymes capable of penetrating and degrading cell walls and causing plasmolysis, such as chitinase, protease, and glucanase enzymes and also, competition for resources and space may be assumed to be the reasons for making *Trichoderma* sp a potential biocontrol agent.

Effectiveness of *P. fluorescens* is may be due the production of antifungal compounds such as pseudobactin, HCN, salicylic acid and 2-hydroxy phenazine which suppress growth of fungi.

Integrated management of soybean pod blight. For evaluating the success in managing the disease, the per cent disease index (PDI), yield, and cost benefit ratio were calculated (Table 5). The results showed that all of the therapies were significantly better than the control group. Treatment T7 (Seed treatment with *T. harzianum* (5g/ kg seed) followed by spray with tebuconazole + trifloxystrobin (0.05 per cent) at 60 DAS and spray of *P. fluorescens* (1 per cent) at 75 DAS) has a minimum PDI of 12.62, which is significantly effective in managing the disease, followed by treatment T6 (Seed treatment with *T. harzianum* (5g/ kg seed) followed by spray with carbendazim (0.1 %) at 60 and 75 DAS) with PDI of 14.20 which are on par with each other and significantly superior over all other treatments. T4 (Seed treatment with pyraclostrobin + thiophanate methyl (2ml/ kg seed) followed by spray with tebuconazole + trifloxystrobin (0.05 per cent) at 60 and 75 DAS) and T3 (Seed treatment with carboxin + thiram (2g/ kg seed) followed by spray with tebuconazole + trifloxystrobin (0.05 per cent) at 60 and 75 DAS) were the next best treatments and are on par with each other and untreated control had a maximum PDI of 33.91.

Table 5: Integrated management of soybean pod blight.

Sr. No.	Treatments details	Pod blight (PDI)	Yield q/ha	BCR
1.	Seed treatment (ST) with carboxin + thiram (combi product) @ 2g/kg seed + spray with thiophanate methyl @ 0.1% at 60 and 75 DAS	22.96 (28.63)*	16.24	1.41
2.	Seed treatment (ST) with carbendazim + mancozeb (combi product) @ 2g/kg seed + spray with thiophanate methyl @ 0.1% at 60 and 75 DAS	24.49 (29.66)	15.20	1.32
3.	Seed treatment (ST) with carboxin + thiram (combi product) @ 2g/kg seed + spray with tebuconazole + trifloxystrobin (combi product) @ 0.05% at 60 and 75 DAS	17.87 (25.00)	16.99	1.35
4.	Seed treatment (ST) with pyraclostrobin + thiophanate methyl (combi product) @ 2ml/kg seed + spray with tebuconazole + trifloxystrobin (combi product) @ 0.05 % at 60 and 75 DAS	17.20 (24.5)	18.05	1.44
5.	Seed treatment (ST) with *Trichoderma harzianum* @ 5g/kg seed + spray with *Pseudomonas fluorescens* @ 1 % at 60 and 75 DAS	28.41 (32.21)	14.58	1.21
6.	Seed treatment (ST) with *Trichoderms harzianum* @ 5g/kg seed + spray with carbendazim @ 0.1 % at 60 and 75 DAS	14.2 (22.14)	19.42	1.64
7.	Seed treatment (ST) with *Trichoderma harzianum* @ 5g/kg seed + spray with tebuconazole + trifloxystrobin (combi product) @ 0.05 % at 60 DAS and spray of *Pseudomonas fluorescens* @ 1% at 75 DAS	12.62 (20.81)	20.33	1.76
8.	Control	33.91 (35.61)	11.33	1.02
	S.Em ±	0.88	0.19	
	C.D at 5%	2.66	0.58	

* Values in the parentheses are arc sine transformed

T7 (Seed treatment with *T. harzianum* (5g/ kg seed) followed by spray with tebuconazole + trifloxystrobin (0.05 per cent) at 60 DAS and spray of *P. fluorescens* (1 per cent) at 75 DAS) had the highest yield (20.33 q/ ha), which was significantly higher than all other treatments. T6 (Seed treatment with *T. harzianum* (5g/ kg seed) followed by spray with carbendazim (0.1 %) at 60 and 75 DAS) with mean yield (19.42 q/ ha). Minimum yield was documented in untreated control (11.33 q/ ha).

Maximum benefit: cost ratio (BCR) was obtained in (Seed treatment with *T. harzianum* (5g/ kg seed) followed by spray with tebuconazole + trifloxystrobin (0.05 %) at 60 DAS and spray of *P. fluorescens* (1 %) at 75 DAS) (1.76) followed by treatment seed treated with *T. harzianum* (5g/ kg seed) followed by spray with carbendazim (0.1 %) at 60 and 75 DAS (1.64) and seed treatment with pyraclostrobin + thiophanate methyl (2ml/ kg seed) followed by spray with tebuconazole + trifloxystrobin (0.05 %) at 60 and 75 DAS (1.44) and the minimum in untreated control (1.02) followed by treatment seed treated with *T. harzianum* (5g/ kg seed) followed by spray with *P. fluorescens* (1 %) at 60 and 75 DAS (1.21).

Pawan *et al*. (2018) found that seed treatment with carboxin + thiram (combi product) (2g/ kg seed) followed by spray with thiophanate methyl (0.1 per cent) at 55 DAS and 75 DAS was superior, with the lowest per cent pod infection and the highest yield of 1183.3 and 811.6 kg/ha in 2016 and 2017, respectively, compared to seed treatment with carbendazim + mancozeb (2g/kg seed) followed by spray with thiophanate methyl (0.1 %) at 55 and 75 DAS with yield of 1080.0 and 798.3 kg/ ha during 2016 and 2017, respectively.

Wasule *et al*. (2018) found that treatment seed treated with carboxin + thiram (combi product) (2g/ kg seed) followed by spray with thiophanate methyl (0.1 per cent) at 55 DAS and 75 DAS had the lowest per cent disease (5.72) and the highest grain production (1246 kg/ ha) subsequently treatment seed treated with carbendazim + mancozeb (2g/ kg seed) followed by spray with thiophanate methyl (0.1 %) at 55 and 75 DAS had per cent disease (7.09) with grain yield of (1139 kg/ ha) and concluded that, seed treatment with carboxin + thiram (2g/ kg), carbendazim + mancozeb (2g/ kg seed) and *Trichoderma* (5g/ kg seed) followed by foliar application of thiophanate methyl (0.1 %) at 55 and 75 DAS resulted in increase in seed germination, least disease index, minimum per cent pod infection and increased 100 seed weight which have contributed for higher seed yield as compared to control and other treatments.

Gawade *et al*. (2009) discovered that carbendazim (0.1%) efficiently controlled the disease, had the lowest mean pod blight (9.64%), the highest seed output (2605 kg/ ha) and test weight (14.33 g), and was the most cost-effective, with a B:C ratio of 1:13.55.

Nagaraj (2013) revealed that, at Dharwad, least PDI (9.94) was found in treatment seed treated with carboxin + thiram (2g/ kg seed) subsequently foliar spray of tebuconazole + trifloxystrobin (0.1 %) at 55 DAS. Maximum seed yield was recorded in treatment (foliar spray of tebuconazole + trifloxystrobin) followed by treatment (seed treatment with carboxin + thiram subsequently foliar spray of tebuconazole + trifloxystrobin) and concluded that, maximum net income was obtained in foliar spray of tebuconazole + trifloxystrobin at 55 DAS followed by seed treatment with carboxin + thiram followed by foliar spray of tebuconazole + trifloxystrobin.

Madhusudhan (2002) claimed that carbendazim seed treatment and foliar spray greatly improved disease severity and seed output in soybeans infected with anthracnose.

CONCLUSION

Copper oxychloride 50 per cent WP, hexaconazole 5 per cent EC, propiconazole 25 per cent EC, carbendazim 50 per cent WP, thiophanate methyl 70 per cent WP, and all combi fungicides were found to be highly effective in inhibiting the pathogen's complete mycelial growth with 100 per cent mycelial inhibition, according to the results of this study. Seed treatment with *T. harzianum* (5g/ kg seed), followed by sprays with tebuconazole + trifloxystrobin (0.05 per cent) at 60 DAS and sprays of *Pseudomonas fluorescens* (1 per cent) at 75 DAS, were found to be significantly more effective than all other treatments in managing soybean pod blight, with a minimum disease index (PDI) of 12.62 and the highest yield (20.33 q/ ha) and BC ratio of 1.76.

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