

Comparative effectiveness of fungicides, botanicals, nano-formulations and bio-control agents against *Colletotrichum siamense* and *Neopestalotiopsis* sp. causing anthracnose disease of guava

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ABSTRACT

Anthracnose is one of the most serious and widespread diseases limiting guava farming success. Infection of growing flowers, immature and developed fruits results in qualitative and quantitative fruit loss. Keeping in view, an attempt has been made to manage the disease by exploiting the systemic and non-systemic fungicides, botanicals, nano-formulations and bio-control agents under *in vitro* conditions. The comparative efficacy of the fungicides, botanicals and bio-control agents for the management of anthracnose disease of guava was also investigated using foliar spray at University Seed Farm (USF), Ladowal, Ludhiana, in 2021 and 2022. Among various treatments evaluated for their inhibitory effect on the mycelial growth of *Colletotrichum siamense* and *Neopestalotiopsis* sp., 100 per cent mycelial growth inhibition was achieved with azoxystrobin 18.2% + difenoconazole 11.4% SC and tebuconazole 50% + trifloxystrobin 25% WG at 50 and 100 ppm concentrations under laboratory conditions. Azoxystrobin 18.2% + difenoconazole 11.4% SC and tebuconazole 50% + trifloxystrobin 25% WG exhibited low per cent disease index and provided significantly higher disease control as compared to other treatments under field conditions.

Key words: Guava, anthracnose, *Colletotrichum siamense*, *Neopestalotiopsis* sp., fungicides, botanicals, bio-control agents, nano-formulations, management

Guava (*Psidium guajava*) is an important commercial fruit crop of India belonging to Myrtaceae family (Bose and Mitra, 1990). It is well grown in tropical and subtropical areas of the world (Adhau and Salvi, 2014) and has potential to adapt well to various ecological conditions, including wastelands and soil with higher pH values (8.6 to 9.6) (Gautam *et al.*, 2010). India is a major producer of guava, harvesting 25 metric tons accounting for 45 per cent of the world's guava production. In India, guava ranks fifth and fourth with respect to area and fruit production, respectively. Guava is an important fruit crop in Punjab, ranking second after Kinnnow. Various diseases have been reported in guava, of which anthracnose is considered as the second most important disease (Rahman *et al.*, 2003). Guava anthracnose receives more attention as it can affect

young developing flowers, immature and ripened fruits leading to qualitative and quantitative loss of the fruit (Hossain and Meah, 1992). The characteristic symptoms on matured fruit consist of sunken, dark colored, necrotic lesions. Under humid conditions, the necrotic lesions become covered with pinkish spore masses. Keeping in view the devastating potential of the disease, the present study was conducted to manage guava anthracnose by using various fungicides, botanicals, nano-formulations and bio-control agents. The research trials were carried out both under *in vitro* and *in vivo* conditions.

MATERIALS AND METHODS

***In vitro* efficacy of fungicides, botanicals, nano-formulations and bio-control agents against *Colletotrichum siamense* and *Neopestalotiopsis* sp.**

Two fungi isolated from anthracnose affected fruit samples of guava were identified as *Colletotrichum*

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siamense and *Neopestalotiopsis* sp. after molecular confirmation. The ITS, *ACT* and *TUB2* sequences were deposited in GenBank as accession numbers OR002118, OR004846 and OR025590 for isolate no. C-2 and OR005054, OR025591 and OR025592 for isolate no. C-15, respectively. Similarly, molecular phylogenetic analysis of ITS region of the isolate no. N-5 was done and the ITS sequence was deposited in GenBank as accession number OR105072 (Rabia, 2023).

To check mycelial growth inhibition of these two fungi *C. siamense* and *Neopestalotiopsis* sp., five systemic fungicides viz., tebuconazole 50% + trifloxystrobin 25% WG, azoxystrobin 18.2% + difenoconazole 11.4% SC, carbendazim 12% + mancozeb 63% WP, propiconazole 25% EC and zineb 68% + hexaconazole 4% WP; three non-systemic fungicides viz., zineb 75 WP, mancozeb 75 WP and copper oxychloride 50 WP; three botanicals viz., neem extract (leaves, tender branches and fruits), garlic extract (clove) and mustard oil (seeds); two different metal and metal oxide nano-formulations viz., copper and silver and lastly two bio-control agents viz., *Trichoderma harzianum* and *Pseudomonas fluorescens* were evaluated under *in vitro* conditions. The bio-control agents, botanicals and nano-formulations were procured from Department of Plant Pathology, School of Organic Farming and Department of Soil Science, PAU, Ludhiana, respectively. *In vitro* evaluation of the chemicals was done using poisoned food technique (Nene and Thapliyal, 1993). While the bio-control agents were evaluated for their efficacy using dual culture technique (Skidmore and Dickinson, 1976). Systemic fungicides were tested at a series of concentrations viz., 5, 10, 25, 50 and 100 ppm and non-systemic fungicides were tested at 50, 100, 200, 500 and 1000 ppm concentrations. Similarly, botanicals were evaluated at different concentrations (5, 10 and 15%) against both the fungi. Nano-formulations (copper and silver) were evaluated at 100, 200, 300, 400, 500, 600, 700, 800, 900 and 1000 ppm concentrations. Different concentrations of test chemicals were mixed (each) with 100 ml of double-strength potato dextrose agar (PDA) medium. After the mixing of agar and test chemicals, approximately 15-20 ml of poisoned PDA medium was poured into Petri plates of 90 mm diameter. Circular bits (5 mm) were cut from the periphery of the actively growing fungal culture and were placed aseptically in the

center of each Petri plate. The Petri plates having non-poisoned PDA medium served as control. After inoculation, the Petri plates were incubated at $25\pm 2^{\circ}\text{C}$ and the colony growth of pathogens were recorded until the growth in the control Petri plate was full (90 mm). Per cent growth inhibition in colony was calculated at each concentration by the formula given by Vincent (1947).

The data obtained were analysed by using CPCS software to determine the efficacy of chemicals. In case of *in-vitro* evaluation of fungicides, ED_{50} and ED_{90} values were also determined by probit analysis.

Field evaluation of fungicides, botanicals and bio-control agents against anthracnose disease of guava

For *in vivo* efficacy of the chemicals and bio-control agents against anthracnose disease of guava, the research trial was conducted in a randomized block design on guava cultivar Allahabad safeda at University Seed Farm (USF), Ladowal, Ludhiana during 2021 and 2022. Five systemic fungicides viz., tebuconazole 50% + trifloxystrobin 25% WG (0.1%), azoxystrobin 18.2% + difenoconazole 11.4% SC (0.1%), carbendazim 12% + mancozeb 63% WP (0.2%), propiconazole 25% EC (0.1%) and zineb 68% + hexaconazole 4% WP (0.1%); three non-systemic fungicides viz., zineb 75 WP (0.2%), mancozeb 75 WP (0.2%) and copper oxychloride 50 WP (0.3%); three botanicals viz., neem extract (15%), garlic extract (15%) and mustard oil (15%) and two bio-control agents viz., *T. harzianum* (1.5%) and *P. fluorescens* (1.5%) were further evaluated as foliar spray against guava anthracnose. As the *in vitro* efficacy of nano-formulations was very less against both the pathogens as compared to fungicides, botanicals and bio-control agents, therefore, field efficacy of nano-formulations was not evaluated for managing the disease. The treatment without any application of chemical served as control. Each treatment was replicated thrice by keeping three guava plants per replication. Three sprays of each treatment were given starting at flower bud emergence, second 15 days after first spray and third 15 days after second spray. The Per cent Disease Index (PDI) was recorded on the basis of 0-4 disease scale (0- No infection, 1- 1 to 25%, 2- 26 to 50%, 3- 51 to 75%, 4- >75%) given by Prabakar *et al.* (2008). The PDI was calculated using the following formula (Wheeler, 1969):

$$\text{PDI} = \frac{\text{Sum of numerical ratings}}{\text{Number of fruits examined} \times \text{Maximum grade}} \times 100$$

The data obtained was analysed by using CPCS software to determine the efficacy of chemicals.

RESULTS AND DISCUSSION

In vitro efficacy of fungicides, botanicals, nano-formulations and bio-control agents against *Colletotrichum siamense* and *Neopestalotiopsis* sp.

It is evident from the data presented in Table 1 and 2 that among the systemic fungicides evaluated for their inhibitory effect on the mycelial growth of *C. siamense* and *Neopestalotiopsis* sp., 100 per cent mycelial growth inhibition was achieved with azoxystrobin 18.2% + difenoconazole 11.4% SC and tebuconazole 50% + trifloxystrobin 25% WG at 50 and 100 ppm concentrations and with carbendazim 12% + mancozeb 63% WP at 100 ppm concentration. Comparing all the fungicide treatments at a particular concentration revealed that azoxystrobin 18.2% + difenoconazole 11.4% SC showed highest per cent growth inhibition of both the fungi at all the tested concentrations in contrast to zineb 68% + hexaconazole 4% WP which showed least growth inhibition at all concentrations. ED₅₀ value of all the evaluated systemic fungicides against both the fungi were < 5ppm except zineb + hexaconazole. Azoxystrobin + difenoconazole and tebuconazole + trifloxystrobin were observed to be highly effective having ED₉₀ values of 15.85 and 20.42, respectively, against *C. siamense* and 15.13

and 22.38, respectively, against *Neopestalotiopsis* sp. These findings were in conformity with those of Sundravada *et al.* (2007) who reported complete inhibition of mycelial growth of *C. gloeosporioides* by azoxystrobin. Similarly, Ranjitha *et al.* (2019) observed that all the systemic fungicides including tebuconazole + trifloxystrobin, difenoconazole, propiconazole and zineb + hexaconazole reduced mycelial growth of *C. gloeosporioides* from 85.06 to 100 per cent. These results are also in accordance with the findings of Biju *et al.* (2018) who reported that carbendazim, propiconazole and carbendazim + mancozeb completely inhibited hyphal growth of *Neopestalotiopsis clavispora* at all the concentrations assessed.

Among non-systemic fungicides, copper oxychloride 50% WP inhibited the mycelial growth of *C. siamense* and *Neopestalotiopsis* sp. up to 72.80 and 70.97 per cent, respectively, at 1000 ppm (Table 3 and 4). Zineb 75% WP proved to be the least effective in inhibiting the colony growth of both the fungi at all concentrations. ED₅₀ and ED₉₀ values of copper oxychloride against *C. siamense* were 262.27 and more than 1000 ppm, respectively and 254.12 and more than 1000 ppm, respectively, against *Neopestalotiopsis* sp. These findings corroborate with Mathews *et al.* (2009) who reported the significant efficacy of copper oxychloride against *C. gloeosporioides* at 1000 ppm as compared to the other test concentrations. They also reported that mancozeb was the least effective fungicide in reducing mycelial growth of the fungus up to 61.91 per cent at 1000 ppm. Similarly, Biju *et*

Table 1. *In vitro* efficacy of systemic fungicides against *Colletotrichum siamense*

Fungicide	Per cent mycelial growth inhibition at different concentrations (ppm)*						
	5	10	25	50	100	ED ₅₀	ED ₉₀
Propiconazole 25% EC	60.23 (50.88)	65.81 (54.20)	78.03 (62.03)	85.23 (67.37)	94.80 (76.81)	<5	66.07
Carbendazim 12% + mancozeb 63% WP	59.36 (50.37)	64.57 (53.45)	76.42 (60.95)	82.74 (65.45)	100 (89.97)	<5	72.63
Azoxystrobin 18.2% + difenoconazole 11.4% SC	71.72 (57.85)	80.65 (63.94)	93.52 (75.24)	100 (89.97)	100 (89.97)	<5	15.85
Zineb 68% + hexaconazole 4% WP	47.74 (43.69)	59.56 (50.49)	64.71 (53.53)	78.68 (62.48)	91.00 (72.53)	6.61	95.82
Tebuconazole 50% + trifloxystrobin 25% WG	60.21 (50.87)	71.99 (58.03)	90.77 (72.29)	100 (89.97)	100 (89.97)	<5	20.42

*Mean of three replications

CD (p=0.05) : Fungicides = 0.69; Concentrations = 0.69; Fungicides × Concentrations = 1.53

Figures in parentheses represent growth mycelial inhibition in arc sine transformed values

Table 2. *In vitro* efficacy of systemic fungicides against *Neopestalotiopsis* sp.

Fungicide	Per cent mycelial growth inhibition at different concentrations (ppm)*						
	5	10	25	50	100	ED ₅₀	ED ₉₀
Propiconazole 25% EC	59.19 (50.28)	63.58 (52.87)	77.97 (61.98)	83.67 (66.14)	93.20 (74.86)	<5	90.28
Carbendazim 12% + mancozeb 63%	57.44 (49.26)	62.91 (52.46)	74.45 (59.64)	81.06 (64.18)	100 (89.97)	<5	58.66
Azoxystrobin 18.2% + difenoconazole 11.4% SC	73.09 (58.73)	81.71 (64.69)	94.39 (76.27)	100 (89.97)	100 (89.97)	<5	15.13
Zineb 68% + hexaconazole 4% WP	45.81 (42.58)	61.28 (51.50)	66.04 (54.33)	78.07 (62.05)	89.95 (71.49)	6.60	>100
Tebuconazole 50% + trifloxystrobin 25% WG	60.47 (51.03)	70.03 (56.78)	89.85 (71.40)	100 (89.97)	100 (89.97)	<5	22.38

*Mean of three replications

CD (p=0.05): Fungicides = 0.63; Concentrations = 0.63; Fungicides × Concentrations = 1.41

Figures in parentheses represent mycelial growth inhibition in arc sine transformed values

al. (2018) also reported comparatively low efficacy of Bordeaux mixture against *N. clavispora* at all the test concentrations i.e., 0.05, 0.125, 0.25, 0.5 and 1.0 per cent.

The results of *in vitro* efficacy of botanicals (Table 5 and 6) indicated significant variation among all three plant extracts in terms of per cent growth inhibition of *C. siamense* and *Neopestalotiopsis* sp.. Maximum growth inhibition of *C. siamense* was achieved with neem extract (38.17%) which was followed by mustard oil (27.74%) at 15 per cent concentration. The least growth inhibition was recorded in garlic extract (18.80%). The results are similar as has been observed by Venkataravanappa *et al.* (2005), neem leaf extract was the most effective in inhibiting mycelial growth (30.62 %) of *C. gloeosporioides*, the incitant of blossom blight of mango. Garlic extract was the most effective in inhibiting mycelial growth of *Neopestalotiopsis* sp.

with 35.30 per cent growth inhibition at 15 per cent concentration. This treatment was followed by neem extract which suppressed the fungal growth in terms of growth inhibition of 25.76 per cent at 15 per cent concentration. The fungal growth inhibition with mustard oil increased by 3.7-folds when increasing its concentration from 5 to 15 per cent. The present finding is in agreement with the work of Sangma *et al.* (2017) who demonstrated fungicidal activity of garlic extract against *Pestalotiopsis versicolor*, a pathogen of guava and observed that garlic extract was significantly superior to all other botanicals.

Nano-formulations, in comparison to fungicides and botanicals, were very less effective in inhibiting the mycelial growth of both the fungi (Table 7 and 8). Minimal fungal growth inhibition was observed even at highest concentration of nano-formulations (1000ppm). Silver nano-formulations could not suppress the mycelial growth of *C. siamense* and

Table 3. *In vitro* efficacy of non-systemic fungicides against *Colletotrichum siamense*

Fungicide	Per cent mycelial growth inhibition at different concentrations (ppm)*						
	50	100	200	500	1000	ED ₅₀	ED ₉₀
Copper oxychloride 50 % WP	23.69 (29.06)	30.30 (33.41)	46.19 (42.79)	65.78 (54.18)	72.80 (58.56)	262.27	>1000
Mancozeb 75 % WP	17.37 (24.62)	27.09 (31.35)	38.23 (38.17)	52.42 (46.37)	63.36 (52.72)	446.68	>1000
Zineb 75 % WP	11.92 (20.19)	16.39 (23.87)	20.62 (26.99)	26.24 (30.80)	36.43 (37.10)	>1000	>1000

*Mean of three replications

CD (p=0.05): Fungicides = 0.70; Concentrations = 0.54; Fungicides × Concentrations = 1.21

Figures in parentheses represent mycelial growth inhibition in arc sine transformed values

Table 4. *In vitro* efficacy of non-systemic fungicides against *Neopestalotiopsis* sp.

Fungicide	Per cent growth inhibition at different concentrations (ppm)*						
	50	100	200	500	1000	ED ₅₀	ED ₉₀
Copper oxychloride 50% WP	22.05 (27.99)	28.31 (32.16)	45.80 (42.54)	63.79 (52.96)	70.97 (57.26)	254.12	>1000
Mancozeb 75% WP	15.93 (23.51)	25.97 (30.62)	36.80 (37.33)	51.80 (46.01)	62.41 (52.17)	467.73	>1000
Zineb 75% WP	10.51 (18.90)	14.75 (22.57)	18.88 (25.73)	24.79 (29.84)	34.96 (36.22)	>1000	>1000

*Mean of three replications

CD (p=0.05): Fungicides = 0.73; Concentrations = 0.57; Fungicides × Concentrations = 1.26

Figures in parentheses represent mycelial growth inhibition in arc sine transformed values

Neopestalotiopsis sp. up to 300 and 200 ppm concentration, respectively. Copper nano-formulation was better in contrast to silver nano-formulation in inhibiting growth of both the pathogens. Copper nano-formulation was found 3.77-folds and 3.52-folds superior in reducing the mycelial growth of *C. siamense* and *Neopestalotiopsis* sp., respectively in comparison to silver nano-formulation at 1000 ppm concentration. Similarly, Dong *et al.* (2023) conducted a study to assess the *in vitro* efficacy of silver, copper and mixed silver-copper nano solution at different concentrations against *C. gloeosporioides*. They observed fungal growth inhibition for nano silver at 125 ppm, for nano copper at 75 ppm and for silver-copper nano solution at 50 ppm. Jagana *et al.* (2017) also reported inhibition of spore germination of *C. musae* isolated from banana with copper, silver, nickel, and magnesium formulations extracted from the leaves of the medicinal plant ajwain (*Trachyspermum ammi*).

Among the bio-control agents, *T. harzianum* was found superior in controlling the growth of *C. siamense* as compared to *P. fluorescens* expressing

59.62 per cent growth inhibition over 29.47 per cent recorded with *P. fluorescens* (Table 9). These findings are in conformity with those of Patil *et al.* (2009) who reported 58.06 per cent growth inhibition of *C. gloeosporioides* by *T. harzianum*. Similarly, Galindez *et al.* (2017) also reported that *T. harzianum* significantly reduced the mycelial growth (59.16%) of *C. gloeosporioides*. These results corroborate the finding of Singh *et al.* (2020) who observed 57.35 per cent growth inhibition of *C. gloeosporioides* in the presence of *T. harzianum*. The effectiveness of two potential bio-control agents (*T. harzianum* and *P. fluorescens*) was also investigated against *Neopestalotiopsis* sp. under *in vitro* conditions. The data presented in Table 10 showed that in dual culture with *T. harzianum* against *Neopestalotiopsis* sp., the radial growth of the fungus was 3.11 cm as compared to control (7.5 cm). *T. harzianum* proved to be better bio-control agent suppressing the growth of fungal pathogen with per cent growth inhibition of 58.53 over *P. fluorescens* which provided fungal growth inhibition of 25.97 per cent. Similarly, Saju *et al.* (2011) reported significant differences among the

Table 5. *In vitro* efficacy of botanicals against *Colletotrichum siamense*

Botanical	Per cent mycelial growth inhibition at different concentrations (%)*		
	5	10	15
Mustard oil	8.40 (16.84)	18.52 (25.47)	27.74 (31.77)
Neem extract	11.30 (19.63)	22.86 (28.55)	38.17 (38.14)
Garlic extract	5.19 (13.16)	13.29 (21.35)	18.80 (25.68)

*Mean of three replications

CD (p=0.05): Fungicides = 0.69; Concentrations = 0.69; Fungicides × Concentrations = 1.20

Figures in parentheses represent mycelial growth inhibition in arc sine transformed values

Table 6. *In vitro* efficacy of botanicals against *Neopestalotiopsis* sp.

Botanical	Per cent growth inhibition at different concentrations (%) *		
	5	10	15
Neem extract	9.32 (17.76)	17.50 (24.72)	25.76 (30.48)
Garlic extract	9.98 (18.40)	19.97 (26.53)	35.30 (36.42)
Mustard oil	4.54 (12.25)	10.17 (18.59)	16.81 (24.19)

*Mean of three replications

CD (p=0.05): Fungicides = 0.97; Concentrations = 0.97; Fungicides × Concentrations = 1.68

Figures in parentheses represent mycelial growth inhibition in arc sine transformed values

Table 7. *In vitro* efficacy of nano-formulations against *Colletotrichum siamense*

Nano-formulation	Per cent growth inhibition at different concentrations (ppm)*									
	100	200	300	400	500	600	700	800	900	1000
Silver	0 (0.00)	0 (0.00)	0 (0.00)	0.18 (2.41)	0.46 (3.86)	1.34 (6.65)	2.46 (9.01)	3.39 (10.60)	4.02 (11.53)	4.79 (12.64)
Copper	3.52 (10.81)	6.11 (14.31)	7.37 (15.74)	7.51 (15.89)	9.22 (17.65)	10.91 (19.28)	11.51 (19.82)	13.01 (21.14)	13.40 (21.46)	18.07 (25.14)

*Mean of three replications

CD (p=0.05): Fungicides = 0.27; Concentrations = 0.61; Fungicides × Concentrations = 0.87

Figures in parentheses represent mycelial growth inhibition in arc sine transformed values

bio-control agents in inhibiting *Pestalotiopsis* sp., the incitant of leaf streak disease of large cardamom (*Amomum subulatum*). Observations on mycelial growth of the pathogen after 7 days of incubation indicated that *Bacillus subtilis* showed maximum growth inhibition (62.6 %) against *Pestalotiopsis* sp. followed by *T. viride* (50.9 %), *P. fluorescens* (41.3 %) and *T. harzianum* (30.4 %). These results are also in accordance with the findings of Sangma *et al.* (2017) who reported that *T. harzianum* exerted the maximum inhibition (77.40%) of mycelial growth of *Pestalotiopsis versicolor*, a pathogen of guava followed by *T. viride* and *P. putida* with an inhibition of 72.96 and 52.59 per cent, respectively. Similar results have been observed by Barman *et al.* (2015)

who identified the antagonist *P. fluorescens* isolates 1 and 2 as poor inhibitor with the growth inhibition of only 27.8 and 35.4 per cent, respectively against *P. theae* causing grey blight of tea. Whereas, the antagonist *T. viride* was found most effective and exerted 74.3% inhibition of mycelial growth over the control.

Field evaluation of fungicides, botanicals and bio-control agents against anthracnose disease of guava

The pooled analysis of two years data (Table 11) revealed that all the fungicides were found significantly effective in managing anthracnose disease of guava as compared to untreated control.

Table 8. *In vitro* efficacy of nano-formulations against *Neopestalotiopsis* sp.

Nano-formulation	Per cent mycelial growth inhibition at different concentrations (ppm)*									
	100	200	300	400	500	600	700	800	900	1000
Silver	0 (0.00)	0 (0.00)	0.13 (2.03)	0.34 (3.30)	1.89 (7.88)	2.51 (9.08)	3.63 (10.92)	4.50 (12.22)	5.03 (12.91)	5.50 (13.53)
Copper	3.66 (10.93)	5.79 (13.83)	6.22 (14.42)	6.46 (14.71)	9.73 (18.16)	11.13 (19.47)	13.18 (21.27)	14.06 (22.00)	15.17 (22.91)	19.39 (26.11)

*Mean of three replications

CD (p=0.05): Fungicides = 0.47; Concentrations = 1.06; Fungicides × Concentrations = 1.50

Figures in parentheses represent mycelial growth inhibition in arc sine transformed values

Table 9. *In vitro* efficacy of bio-control agents against *Colletotrichum siamense*

Bio-control agent	Pathogen growth (cm)	Antagonist growth (cm)	Inhibition zone (cm)	Control (cm)	Growth inhibition of the pathogen (%)
<i>Trichoderma harzianum</i>	3.23	4.5	-	8.0	59.62
<i>Pseudomonas fluorescens</i>	5.36	-	0.34	7.6	29.47

Table 10. *In vitro* efficacy of bio-control agents against *Neopestalotiopsis* sp.

Bio-control agent	Pathogen growth (cm)	Antagonist growth (cm)	Inhibition zone (cm)	Control (cm)	Growth inhibition of the pathogen (%)
<i>Trichoderma harzianum</i>	3.11	3.61	-	7.5	58.53
<i>Pseudomonas fluorescens</i>	5.33	-	0.3	7.2	25.97

The treatment azoxystrobin + difenoconazole proved significantly superior to other fungicides by reduction in the per cent disease index. In azoxystrobin + difenoconazole (1 ml/litre water) treated plants PDI recorded on fruits was 9.05 which was considerably lower when compared with corresponding control with 45.19 per cent disease index. Maximum disease control of 79.97 per cent was exhibited by this combination of fungicide. Tebuconazole + trifloxystrobin was the next best treatment in minimizing the PDI (9.76) and providing 78.40 per cent disease control. Superiority of azoxystrobin 18.2% + difenoconazole 11.4% SC and tebuconazole 50% + trifloxystrobin 25% WG in reducing per cent

disease index and providing significantly higher disease control may be due to their better ability to provide longer protective action on the tree canopy and to prevent fresh infection by air-borne conidia which may serve as reservoir for secondary spread of anthracnose disease. Carbendazim + mancozeb and propiconazole were statistically at par with each other for their efficacy against anthracnose disease of guava. Among non-systemic fungicides copper oxychloride was also effective in controlling the disease up to 59.53 per cent. Zineb proved the least effective among all the fungicides as it resulted into 28.68 PDI along with 36.53 per cent disease control. Botanicals and bio-control agents were less effective

Table 11. Field evaluation of fungicides, botanicals and bio-control agents for managing guava anthracnose during 2021 and 2022 at USF, Ladowal, Ludhiana

Treatment	Concentration (%)	PDI			Disease control (%)
		2021	2022	Mean	
Propiconazole 25% EC	0.1	14.36	12.52	13.44	70.26
Azoxystrobin 18.2% + difenoconazole 11.4% SC	0.1	9.84	8.26	9.05	79.97
Carbendazim 12% + mancozeb 63% WP	0.2	13.39	12.18	12.78	71.72
Tebuconazole 50% + trifloxystrobin 25% WG	0.1	10.18	9.34	9.76	78.40
Zineb 68% + hexaconazole 4% WP	0.1	16.88	15.97	16.42	63.66
Copper oxychloride 50 WP	0.3	18.41	18.17	18.29	59.53
Mancozeb 75 WP	0.2	20.18	19.33	19.75	56.29
Zineb 75 WP	0.2	29.22	28.15	28.68	36.53
Neem leaf extract	15	30.79	29.75	30.27	33.01
Mustard oil	15	31.33	31.84	31.60	30.07
Garlic extract	15	30.59	29.43	30.01	33.59
<i>Pseudomonas fluorescens</i>	1.5	33.61	32.08	32.84	27.33
<i>Trichoderma harzianum</i>	1.5	32.05	30.61	31.33	30.67
Control	-	44.88	45.51	45.19	-
CD (p=0.05)		4.04	2.34	3.19	

as compared to fungicides in controlling the disease under field conditions. Among botanicals, maximum disease control was achieved with garlic clove extract up to 33.59 per cent. Whereas *T. harzianum* reduced the disease by 30.67 per cent and *P. fluorescens* was the least effective treatment with only 27.33 per cent disease control.

These results corroborate the finding of Sundravadana *et al.* (2007) who observed complete control of mango anthracnose with azoxystrobin. Azoxystrobin at 1, 2 and 4 ml/litre suppressed the development of both panicle and leaf anthracnose. Adhikary *et al.* (2013) also reported maximum reduction of anthracnose disease with azoxystrobin up to 75.29 per cent. The results are in agreement with the findings of Pandey *et al.* (2016) who reported superiority of azoxystrobin over other fungicides against mango anthracnose as it exhibited PDI up to 14.7 per cent. These findings are in conformity with those of Chaudhari and Gohel (2016) who reported that two foliar sprays of tebuconazole + trifloxystrobin 75 WG at 15 days interval starting from initiation of disease can efficiently manage the anthracnose disease of mungbean. These results are also in accordance with Singh *et al.* (2008) who reported that sprays of the systemic fungicides were very effective for the management of anthracnose of guava as compared to the non-systemic fungicides. Similarly, Sharma *et al.* (2022) reported that fungicides viz., Nativo, Follicur and Saaf; bio-agent *T. harzianum* and aqueous extracts of botanical i.e., *Melia azadirachtin* were found to be effective against *C. lindemuthianum*. Two foliar sprays of trifloxystrobin + tebuconazole 75 WG @ 1 g/litre water and *T. harzianum* @ 10 g/litre water at 45th and 60th day after sowing were highly effective and resulted in lower disease severity and higher seed yield. Similar results were reported by Patel (2017) that different isolates of *T. harzianum* reduced chilli anthracnose up to 40 per cent.

The results revealed that azoxystrobin 18.2% + difenoconazole 11.4% SC and tebuconazole 50% + trifloxystrobin 25% WG exhibited low per cent disease index and provided significantly higher disease control as compared to other treatments. Therefore, the present investigation indicates that guava anthracnose can be effectively managed by giving three sprays of azoxystrobin 18.2% +

difenoconazole 11.4% SC or tebuconazole 50% + trifloxystrobin 25% WG starting from flower bud emergence at 15 days interval.

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