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## Efficacy of fungicides against *Phyllosticta zingiberi*, causing *Phyllosticta* leaf spot of ginger in *In vitro* conditions

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### Abstract

*Phyllosticta* leaf spot (*Phyllosticta zingiberi*) of ginger is an important disease, causing severe damage to the foliage, which ultimately results in significant yield losses of ginger. Therefore, present *in vitro* studies were undertaken to evaluate the bioefficacy of seven each systemic fungicides and non-systemic/combiprod fungicides, to assess their potential against *P. zingiberi*. Results indicated that Among systemic fungicides evaluated Carbendazim 50% WP, Propiconazole 25% EC, Hexaconazole 5% EC, Thiophanate methyl 70% WP and Difenconazole 25% EC (each @ 500 and 1000 ppm) resulted with cent per cent (100.00%) mycelial growth inhibition of the test pathogen, over untreated control. These were followed by Azoxystrobin 23% SC (73.33 and 83.33%), respectively @ 500 and 1000 ppm. Among non-systemic/combiprod fungicides evaluated Carbendazim 12% + Mancozeb 63% WP, Carboxin 37.5 + Thiram 37.5% WP and Tebuconazole 50% + Trifloxystrobin 25% WP (each @ 2000 and 2500 ppm) resulted with cent per cent (100.00%) mycelial growth inhibition of the test pathogen, over untreated control. These were, followed by Mancozeb 75% WP (73.33 and 80.00%), Metalaxyl M 8% + Mancozeb 64% WP (51.10 and 62.22%) and Propineb 70% WP (42.22 and 68.88%), respectively @ 2000 and 2500 ppm.

Thus, judicious use of these fungicides can be recommended to combat *Phyllosticta* leaf spot of ginger.

**Keywords:** *Zingiber officinale*, *Phyllosticta zingiberi*, fungicides, systemic, non-systemic

### Introduction

Ginger (*Zingiber officinale*), an herbaceous perennial plant belonging to the family Zingiberaceae. It is probably the native of South-East Asia and can also be grown in other parts of the world. India is the world's leading producer of ginger (Medhi *et al.*, 2012) [7]. In India, the area under ginger cultivation was 164 thousand ha., with average production of 1788 thousand MT and average productivity of 10.90 MT / ha. (Anonymous, 2019) [1].

Ginger grows well in warm, humid climate, moderate rainfall and well-drained soils like sandy loam rich in humus. Ginger crop suffers from many insect pests and biotic / abiotic stresses. Among biotic stresses, the diseases caused by fungi, bacteria, viruses and nematodes, are the major constraints in ginger production and productivity. Ginger crops suffers from various foliage diseases, Among various foliage diseases caused by *Phyllosticta*, *Helminthosporium*, *Cercospora*, *Pyricularia*, *Rhizoctonia* and *Septoria*, *Phyllosticta* leaf spot caused by *P. zingiberi* had been reported as most destructive and of common occurrence in the ginger growing states / areas of the country (Sohi *et al.*, 1964; Sood and Dohroo, 2005; Singh, 2015) [12, 13, 11].

*Phyllosticta* leaf spot (*Phyllosticta zingiberi*) of ginger is an important disease, causing severe damage to the foliage, which ultimately results in significant yield losses of ginger. Therefore, considering economic importance of the crop as well as destructive nature of the disease, present *in vitro* studies were undertaken to evaluate the bioefficacy of seven each systemic fungicides and non-systemic/combiprod fungicides, to assess their potential against *P. zingiberi* at Department of Plant Pathology, College of Agriculture, Latur.

### Materials and Methods

Seven systemic (each @ 500, 1000 ppm), three non-systemic and four combiprod (each @ 2000, 2500 ppm) fungicides were evaluated *in vitro* against the highly virulent isolate: Pz-Kh of *P. zingiberi* and using PDA as basal culture medium and by applying Poisoned food technique (Nene and Thapliyal, 1993) [8].

Based on active ingredient, requisite quantity of the all test fungicides was calculated, dispensed separately and mixed thoroughly with autoclaved and cooled (45 °C) PDA medium

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in glass conical flasks (250 ml capacity) and prepared their desired concentrations. This PDA medium separately amended with the test fungicides was then poured (20 ml/plate) aseptically in sterile glass Petri plates (90 mm dia.) and allowed to solidify at room temperature. For each test fungicide and its test concentration, three PDA plates per treatment per replication were maintained and replicated thrice.

After solidification of the fungicides amended PDA medium, all these plates were inoculated aseptically by placing in the centre a 5 mm culture disc of the test fungus, obtained from actively growing 7 days aged pure culture of the test pathogen isolate. PDA plates without fungicide and inoculated with pure culture disc of the test pathogen were maintained as

untreated control. Both treated and untreated PDA plates were incubated in an inverted position in BOD incubator ( $27 \pm 2$  °C). The experimental details were as given below.

#### List of systemic fungicides

Tr. No.	Treatments	Tr. No.	Treatments
T <sub>1</sub>	Carbendazim 50% WP	T <sub>5</sub>	Thiophanate methyl 70% WP
T <sub>2</sub>	Propiconazole 25% EC	T <sub>6</sub>	Difenconazole 25% EC
T <sub>3</sub>	Hexaconazole 5% EC	T <sub>7</sub>	Metalaxyl-M 35% WS
T <sub>4</sub>	Azoxystrobin 23% SC	T <sub>8</sub>	Control (untreated)

#### List of non-systemic and four combiproduct fungicides

Tr. No.	Treatments	Tr. No.	Treatments
T <sub>1</sub>	Propineb 70% WP	T <sub>5</sub>	Carboxin 37.5% + Thiram 37.5% WP
T <sub>2</sub>	Mancozeb 75% WP	T <sub>6</sub>	Metalaxyl M 4% + Mancozeb 64% WP
T <sub>3</sub>	Chlorothalonil 75% WP	T <sub>7</sub>	Tebuconazole 50% + Trifloxystrobin 25% WG
T <sub>4</sub>	Carbendazim 12% + Mancozeb 63% WP	T <sub>8</sub>	Control (untreated)

Observations on radial mycelial growth / colony diameter (mm) of the test pathogen, at an interval of 24 hrs. of incubation were recorded and continued upto seven days or till the untreated PDA plates were fully covered with mycelial growth of the test pathogen. Based on cumulative data, per cent mycelial growth inhibition of the test pathogen with the test fungicides, over untreated control was calculated by applying the following formula (Vincent, 1927) [15].

$$C - T$$

$$\text{Per cent Inhibition (I)} = \frac{C - T}{C} \times 100$$

#### Where

C = Growth (mm) of the test fungus in untreated control plates.

T = Growth (mm) of the test fungus in treated plates.

#### Results and Discussion

Based on symptomatology and pathogenicity test attributes (incubation period, lesion frequency, lesion size and per cent disease severity), *P. zingiberi* (Isolate Pz-Kh), causing leaf spot of ginger were selected to evaluate *in vitro* efficacy of the fungicides against them and the results thereof are described under following sub-heads.

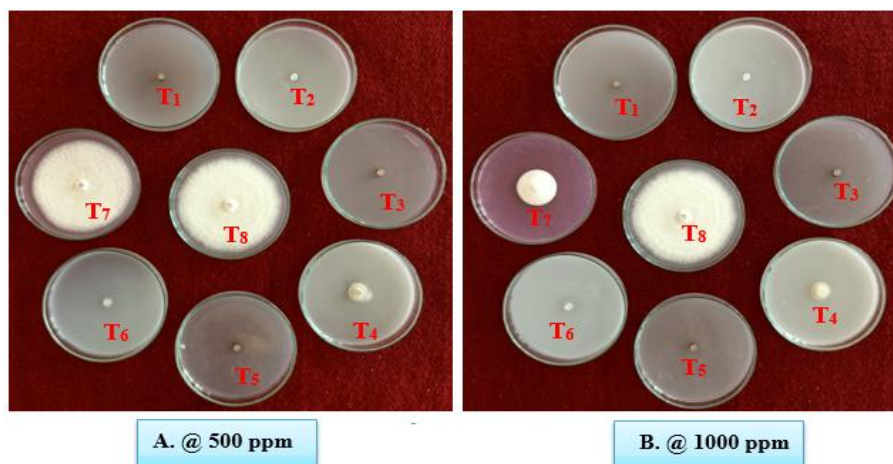
#### *In vitro* efficacy of systemic fungicides against *Phyllosticta zingiberi*, causing leaf spot of ginger

All of the seven systemic fungicides evaluated *in vitro* (each @ 500, 1000 ppm) exhibited antifungal activity against *P. zingiberi* (isolate Pz-Kh), which numerically influenced mycelial growth and its corresponding inhibition, over untreated control (Table 1).

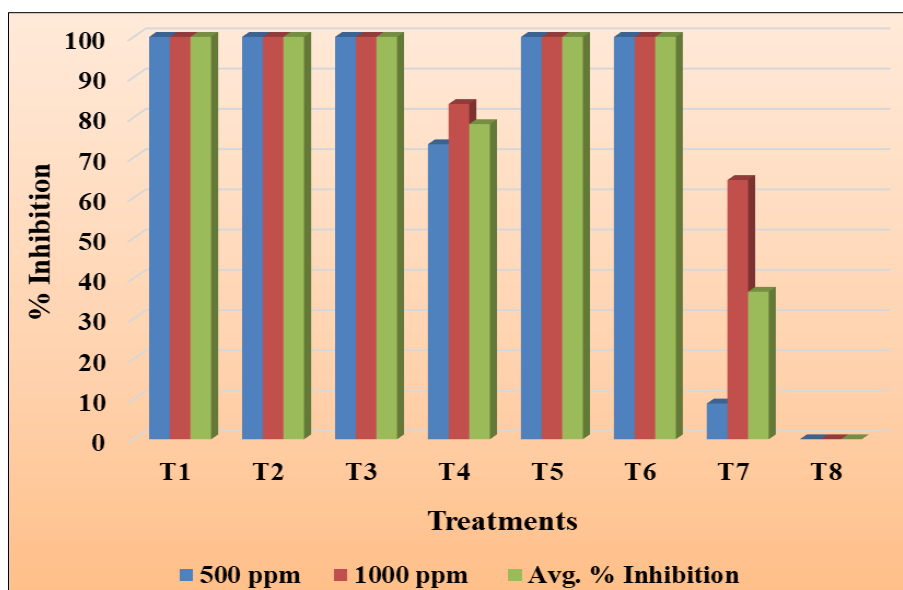
**Table 1:** *In vitro* efficacy of systemic fungicides against *Phyllosticta zingiberi* (isolate Pz-Kh), causing leaf spot of ginger

Tr. No.	Treatments	Colony Diam.* At ppm (mm)		Av. (mm)	% Inhibition* At ppm		Av. Inhibition (%)
		500	1000		500	1000	
T <sub>1</sub>	Carbendazim 50% WP	0.00	0.00	0.00	100 (90.00)	100 (90.00)	100 (90.00)
T <sub>2</sub>	Propiconazole 25% EC	0.00	0.00	0.00	100 (90.00)	100 (90.00)	100 (90.00)
T <sub>3</sub>	Hexaconazole 5% EC	0.00	0.00	0.00	100 (90.00)	100 (90.00)	100 (90.00)
T <sub>4</sub>	Azoxystrobin 23% SC	24.00	15.00	19.50	73.33 (58.89)	83.33 (65.88)	78.33 (62.38)
T <sub>5</sub>	Thiophanate methyl 70% WP	0.00	0.00	0.00	100 (90.00)	100 (90.00)	100 (90.00)
T <sub>6</sub>	Difenconazole 25% EC	0.00	0.00	0.00	100 (90.00)	100 (90.00)	100 (90.00)
T <sub>7</sub>	Metalaxyl 35% WS	82.00	32.00	57.00	8.88 (17.24)	64.44 (53.37)	36.66 (35.30)
T <sub>8</sub>	Control (untreated)	90.00	90.00	90.00	00.00 (00.00)	00.00 (00.00)	00.00 (00.00)
	S.E. ±	0.57	0.28	-	0.64	0.32	-
	C.D. (P=0.01)	1.74	0.87	-	1.94	0.96	-

\*Mean of three replications. Figures in parentheses are arcsine transformed values. Diam.: Diameter, Av.: Average



**Plate 1:** *In vitro* efficacy of systemic fungicides against *P. zingiberi*, causing leaf spot of ginger



**Fig 1:** *In vitro* efficacy of systemic fungicides against *P. zingiberi*, causing leaf spot of ginger

#### Effect on mycelial growth

Result (Table 1, Fig 1, Plate 1) revealed that the test systemic fungicides exhibited a wide range of radial mycelial growth of *P. zingiberi*, which was found to be decreased drastically with increase in concentrations of systemic fungicides. However, Carbendazim 50% WP, Propiconazole 25% EC, Hexaconazole 5% EC, Thiophanate methyl 70% WP and Difenconazole 25% EC (each @ 500 and 1000 ppm) showed none of the mycelial growth of the test pathogen. This was followed by Azoxystrobin 23% SC (24.00 and 15.00 mm) and Metalaxyl 35% WS (82.00 and 32.00 mm) respectively each @ 500 and 1000 ppm, as against maximum mycelial growth (90.00 mm) in untreated control.

Average radial mycelial growth recorded with the test systemic fungicides, ranged from 00.00 mm to 57.00 mm. However, it was least with Azoxystrobin 23% SC (19.50 mm), followed by Metalaxyl 35% WS (57.00 mm).

#### Effect on mycelial growth inhibition

The results (Table 1) revealed that all the systemic fungicides tested (each @ 500 and 1000 ppm) numerically inhibited mycelial growth of *P. zingiberi*, over untreated control and it was directly proportional to concentrations of the test fungicides.

Among systemic fungicides tested, Carbendazim 50% WP,

Propiconazole 25% EC, Hexaconazole 5% EC, Thiophanate methyl 70% WP and Difenconazole 25% EC (each @ 500 and 1000 ppm) resulted with cent per cent (100%) mycelial growth inhibition. These were followed by Azoxystrobin 23% SC (73.33 and 83.33%) and Metalaxyl 35% WS (8.88 and 64.00%) respectively each @ 500 and 1000 ppm.

Average mycelial growth inhibition recorded with the test systemic fungicides ranged from 36.66 to 100 per cent. However, it was numerically highest and cent per cent with Carbendazim 50% WP, Propiconazole 25% EC, Hexaconazole 5% EC, Thiophanate methyl 70% WP and Difenconazole 25% EC (100%), followed by Azoxystrobin 23% SC (78.33%) and Metalaxyl 35% WS (36.66%).

These results are in conformity to the findings of several earlier workers. Systemic fungicides *viz.*, Carbendazim 50% WP, Propiconazole 25% EC, Hexaconazole 5% EC, Azoxystrobin 23% SC, Thiophanate methyl 70% WP, Difenconazole 25% EC and Metalaxyl 35% WS were reported as potential antifungal compounds with significant maximum mycelial growth inhibition of *P. zingiberi*, causing leaf spot of ginger and other crop hosts. (Arunakumara and Satyanarayana, 2015; Bandyopadhyay *et al.*, 2015; Ravikumara *et al.*, 2015; Singh, 2015; Hedge, 2018; Maheshwari *et al.*, 2007) [2, 3, 9, 4, 11, 6].

**In vitro efficacy of non-systemic and combiproduct fungicides against *Phyllosticta zingiberi*, causing leaf spot of ginger**

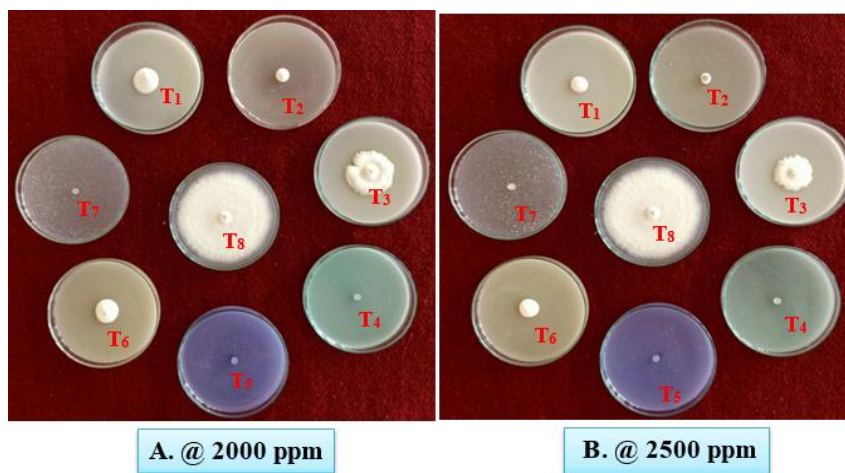
All of the seven non-systemic and combiproduct fungicides

evaluated *in vitro* (each @ 2000 and 2500 ppm) exhibited antifungal activity against *P. zingiberi* (isolate Pz-Kh), which numerically influenced mycelial growth and its corresponding inhibition, over untreated control (Table 2)

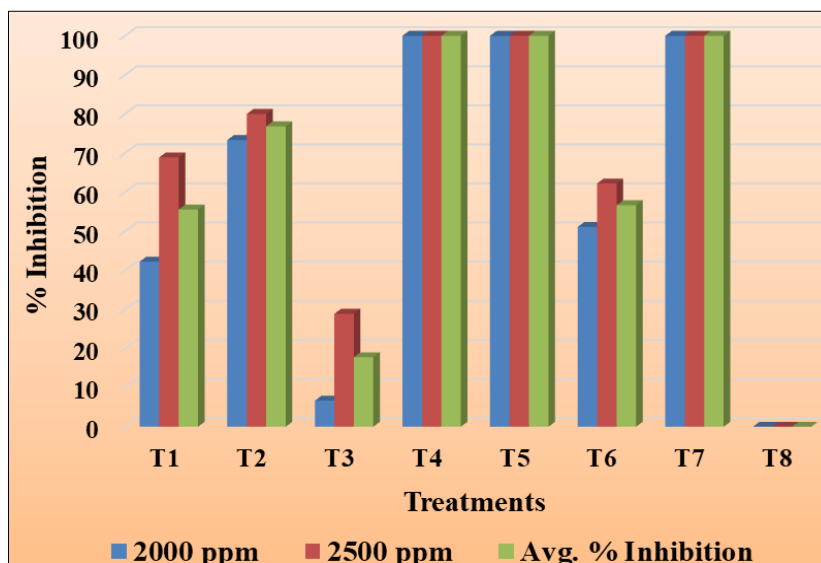
**Table 2:** *In vitro* efficacy of non-systemic and combiproduct fungicides against *P. zingiberi* (isolate Pz-Kh), causing leaf spot of ginger

Tr. No.	Treatments	Colony Diam.* At ppm (mm)		Av. (mm)	% Inhibition* At ppm		Av. Inhibition (%)
		2000	2500		2000	2500	
T <sub>1</sub>	Propineb 70% WP	52.00	28.00	40.00	42.22 (40.50)	68.88 (56.07)	55.55 (48.28)
T <sub>2</sub>	Mancozeb 75% WP	24.00	18.00	36.00	73.33 (58.89)	80.00 (63.67)	76.84 (61.28)
T <sub>3</sub>	Chlorothalonil 75% WP	84.00	64.00	74.00	6.66 (14.81)	28.88 (32.49)	17.77 (23.65)
T <sub>4</sub>	Carbendazim 12% + Mancozeb 63% WP	0.00	0.00	0.00	100 (90.00)	100 (90.00)	100 (90.00)
T <sub>5</sub>	Carboxin 37.5% + Thiram 37.5% WP	0.00	0.00	0.00	100 (90.00)	100 (90.00)	100 (90.00)
T <sub>6</sub>	Metalaxyl M 4% + Mancozeb 64% WP	44.00	34.00	39.00	51.10 (45.61)	62.22 (52.05)	56.66 (48.83)
T <sub>7</sub>	Tebuconazole 50% + Trifloxystorbin 25% WP	0.00	0.00	0.00	100 (90.00)	100 (90.00)	100 (90.00)
T <sub>8</sub>	Control (untreated)	90.00	90.00	90.00	00.00 (00.00)	00.00 (00.00)	00.00 (00.00)
	S.E. ±	0.81	0.40	-	0.90	0.41	-
	C.D. (P=0.01)	2.46	1.23	-	2.74	1.25	-

\*Mean of three replications. Figures in parentheses are parentheses are arcsine transformed values. Diam.: Diameter, Av.: Average



**Plate 2:** *In vitro* efficacy of non-systemic fungicides and combiproduct fungicides against *P. zingiberi*, causing leaf spot of ginger



**Fig 2:** *In vitro* efficacy of non-systemic and combiproduct fungicide against *P. zingiberi*, causing leaf spot of ginger

### Effect on mycelial growth

Result (Table 2, Fig 2, Plate 2) revealed that all the test non-systemic and combiproduct fungicides exhibited a wide range of *P. zingiberi*, mycelial growth, which was found to be decreased drastically with increase in concentrations of the test fungicides. However, Carbendazim 12% + Mancozeb 63% WP, Carboxin 37.5% + Thiram 37.5% WP and Tebuconazole 50% + Trifloxystorbin 25% WP (each @ 2000 and 2500 ppm) did not showed mycelial growth of the test pathogen. These were followed by Mancozeb 75% WP (24.00 and 18.00 mm), Metalaxyl M 8% + Mancozeb 64% WP (44.00 and 34.00 mm), Propineb 70% WP (52.00 and 28.00 mm) and Chlorothalonil 75% WP (84.00 and 64.00 mm) respectively each @ 2000 and 2500 ppm, as against maximum mycelial growth (90.00 mm) in untreated control. Average radial mycelial growth recorded with the test non-systemic and combiproduct fungicides ranged from 00.00 mm to 74.00 mm. However, it was least with Mancozeb 75% WP (36.00 mm), followed by Metalaxyl M 8% + Mancozeb 64% WP (39.00 mm), Propineb 70% WP (40.00 mm) and Chlorothalonil 75% WP (74.00 mm).

### Effect on mycelial growth inhibition

Result (Table 2) revealed that all the non-systemic and combiproduct fungicides tested (each @ 2000 and 2500 ppm) significantly inhibited mycelial growth of *P. zingiberi*, over untreated control and it was directly proportional to concentrations of the fungicides tested.

Among non-systemic and combiproduct fungicides tested, Carbendazim 12% + Mancozeb 63% WP, Carboxin 37.5 + Thiram 37.5% WP and Tebuconazole 50% + Trifloxystorbin 25% WP (each @ 2000 and 2500 ppm) resulted with cent per cent (100%) mycelial growth inhibition. These were followed by Mancozeb 75% WP (73.33 and 80.36%) and Metalaxyl M 8% + Mancozeb 64% WP (51.10 and 62.22%), Propineb 70% WP (42.22 and 68.88%) and Chlorothalonil 75% WP (6.66 and 28.88%) respectively each @ 2000 and 2500 ppm and these fungicides were significantly superior to each other.

Average mycelial growth inhibition recorded with the test non-systemic and combiproduct fungicides ranged from 17.77 to 100 per cent. However, it was numerically highest and cent per cent (100%) with Carbendazim 12% + Mancozeb 63% WP, Carboxin 37.5% + Thiram 37.5% WP and Tebuconazole 50% + Trifloxystorbin 25% WP, followed by Mancozeb 75% WP (76.84%), Metalaxyl M 8% + Mancozeb 64% WP (56.66%), Propineb 70% WP (55.55%) and Chlorothalonil 75% WP (17.77%).

These results are in conformity with the findings of several earlier workers. Non-systemic and combiproduct fungicides viz., Propineb 70% WP, Mancozeb 75% WP, Chlorothalonil 75% WP, Carbendazim 12% + Mancozeb 63% WP, Carboxin 37.5% + Thiram 37.5% WP, Metalaxyl M 8% + Mancozeb 64% WP and Tebuconazole 50% + Trifloxystorbin 25% WP were reported as most antifungal compounds with significant mycelial growth inhibition of *P. zingiberi*, causing leaf spot of ginger and other crop hosts (Sharma and Acharya, 2004; Sood and Dohroo, 2005; Kamhawy, 2006; Thammaiah *et al.*, 2009; Arunakumara and Satyanarayana, 2015; Bandyopadhyay *et al.*, 2015)<sup>[10, 13, 5, 2, 3, 14, 13]</sup>.

### Conclusion

It is concluded from above result that, among the fungicides (systemic, non-systemic and combiproduct) evaluated *in vitro*, Carbendazim 50% WP, Propiconazole 25% EC, Difenconazole 25% EC, Hexaconazole 5% EC (systemic); Mancozeb 75% WP (non-systemic); Carbendazim 12% +

Mancozeb 63% WP, Carboxin 37.5% + Thiram 37.5% WP, Tebuconazole 50% + Trifloxystorbin 25% WP (combiproduct) fungicides were found most prominent in significantly inhibiting mycelial growth of *P. zingiberi*. Thus, judicious use of these fungicides can be recommended to combat *Phyllosticta* leaf spot of ginger.

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