Int.J.Curr.Microbiol.App.Sci (2020) *9*(12): 1895-1902

International Journal of Current Microbiology and Applied Sciences
ISSN: 2319-7706 **Volume 9 Number 12 (2020)**
Journal homepage: http://www.ijcmas.com

Original Research Article https://doi.org/10.20546/ijcmas.2020.912.225

Evaluation of Strobilurin and Triazole Fungicides against Target Leaf Spot caused by (*Corynespora cassiicola*) *in vitro* Condition

Vivek Kumar Vishwakarma*, Moly Saxena and D. R. Saxena

Department of Plant Pathology, RVSKVV College of Agriculture, Sehore (M.P.), India

**Corresponding author*

Keywords

Corynespora cassiicola, Strobilurin and Triazole Fungicides

Article Info

Accepted:
14 November 2020
Available Online:
10 December 2020

A B S T R A C T

Target leaf spot caused by (*Corynespora cassiicola*). The most important fungal diseases of Soybean [*Glycine max* (L.) Merrill]. *In vitro* evaluation of eight fungicides at three concentrations *viz*. 5 0 ppm, 100ppm and 200 ppm. Mix fungicides *viz*., prochloraz + tebuconazole, azoxystrobin + tebuconazole + prochloraz, pyraclostrobin + mefentrifluconazole + fluxapyroxad, carbendazim + mancozeb and hexconazole, completely inhibited the growth of *C. cassiicola*, at lower concentration of 50ppm. At 100ppm, tebuconazole + sulphur reduced the growth of *C. cassiicola* 73%, tebuconazole 61.21% and pyraclostrobin 51.71%. With the increase of concentration to 200 ppm, tebuconazole + sulphur reduced 82.89% mycelial growth, tebuconazole 79.08% and pyraclostrobin 70.34%.

Introduction

Soybean [*Glycine max* (L.) Merrill] is a legume crop and is the second largest after groundnut oilseed in India. It is growing in diverse agro-climatic conditions. Soybean ranks first among the oilseeds in the world and contributes for nearly 25% of the world's total oil and fats production. The USA leads in terms of area and production of soybean, while India ranks fourth in area and fifth in production in the world. USA, Argentina, Brazil, China and India are the major producers of soybean accounting for 90 percent of world production. Productivity of soybean in India (830 kg/ha) is less than global (2800 kg/ha) average due to abiotic and biotic stresses (Anno, 2019).

Strobilurins are an important class of fungicides that come from the discovery of *Stroblilurus tenacellus*, the mushroom fungus that causes wood-rotting. This isolated natural fungicide is thought to be used to protect the fungus against microbes in the decomposition of the wood. The discovery of strobilurins led scientist to isolate and produce synthetic strobilurins by chemically altering the compound to be able to tolerate sunlight (Vincelli, 2012).

Mix fungicides, pyraclostrobin + epoxiconazole, Azoxystrobin + cyproconazole, pyraclostrobin + epoxiconazole + fluxapyroxad and procymidone showed significant reduction in mycelial growth with increased rates of

fungicides from 10 and 100 mg for all isolates of *C.cassicola* (Cabral *et al.,* 2016).The inhibitory concentration (IC50) studies for different fungicides specific to *C. cassiicola* in soybean are scarce, yet it is very useful in carrying out research and sensitivity monitoring, especially in areas where the control of this disease is not being efficient (Avozani *et al.,* 2014).

Materials and Methods

The laboratory experiments were carried out at the Department of Plant Pathology, R.A.K. College of Agriculture, Sehore (M.P.).

Isolation, purification and identification of pathogen

Small pieces of infected tissue (2-3mm in length) target leaf spot were cut at the junction of diseased and healthy portion with the help of disinfected blade after surface sterilizing with alcohol. These bits were surface sterilized in 0.1 per cent mercuric chloride solution ($HgCl_2$) for 30 seconds followed by three washing with sterilized distilled water in Petri plates under aseptic conditions using laminar air flow. These bits were then dried by placing on sterilized blotting paper. Five bits were transferred aseptically to the sterile Petri plates containing potato dextrose agar (PDA) medium. Inoculated Petri plates were incubated at 25 ± 2℃ for five to seven days and examined at frequent intervals to see the growth of the fungus/conidia developing from different pieces. (Ahmad *et al.,* 2013)

The appearing fungus (*Corynespora cassiicola*) was observed after 72 hours and isolations were made from developing colonies for further study. The pathogen was further purified by hyphal tip method and sub-cultured on PDA slants kept at 4 ºC for further study (Dhingra and Sinclair,1985). Isolated fungus (*Corynespora cassiicola*) was identified according to morphological characters and cultural behaviour (Ellis and Subramanian, 1971).

In vitro evaluation of fungicides

The poison food technique (Nene and Thapliyal, 1979) was followed to evaluate the efficacy of fungicides in inhibiting the mycelial growth of *Corynespora cassiicola.* Strobilurin and Triazole fungicides used in the present investigation are three concentrations i.e., 50ppm, 100ppm, 200ppm of each fungicides were used. Three replications were kept for each concentration. *Corynespora cassiicola* was grown on PDA medium for 15 days prior to setting up the experiment. The PDA medium was prepared and melted. Required quantity of fungicides was added to the melted medium to obtain the required concentration on the basis of active ingredient present in the chemical. Little amount of streptomycin was added in each flask before plating to avoid bacterial contamination. Twenty ml of poisoned medium was poured in each sterilized Petri plates. Suitable check was maintained without addition of fungicides. The plates were then inoculated as described earlier and incubated at 25 ±1 ºC. The mycelium growth as colony diameters was measured after 3 days, 6 days and 9 days of inoculation. The inhibition percentage of each fungicide on *Corynespora cassiicola* was determined by using the formula given by Vincent (1947).

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

Where,

I = Per cent inhibition in growth of test pathogen
C = Radial growth (mm) in control
T = Radial growth (mm) in treatment

Results and Discussion

Evaluation of strobilurin and triazole fungicides against caused by *C. cassiicola in vitro* condition

In vitro radial growth of *Corynespora cassiicola* was recorded by poison food technique using eight fungicides at three concentrations *viz*. 50ppm, 100ppm and 200ppm (Plate-1& 2). The data presented in the table 1, 2 & 3 and fig.1, 2 & 3 indicates that the growth of *C. cassiicola* was significantly inhibited at 50ppm, 100ppm, and 200ppm concentrations of fungicides as compared to control.

Table.1 Radial growth of *Corynespora cassiicola* at 50ppm concentration of fungicides

S .No	Treatment	Mean radial growth (mm)*on			Final Inhibition (%)
		3rd day	6th day	9th day	
1	Pyraclostrobin	14.33	25.00	61.00	30.42
2	Tebuconazole	11.33	20.66	42.33	51.71
3	Prochloraz + Tebuconazole	0.00	0.00	0.00	100
4	Azoxystrobin + Tebuconazole + Prochloraz	0.00	0.00	0.00	100
5	Pyraclostrobin + Mefentrifluconazole+ Fluxapyroxad	0.00	0.00	0.00	100
6	Tebuconazole + Sulphur	7.66	14.33	28.66	67.30
7	Carbendazim + Mancozeb	0.00	0.00	0.00	100
8	Hexaconazole	0.00	0.00	0.00	100
9	Control	46.33	77.66	87.66	_
	SE(m) ± 1	**0.40**	**0.43**	**0.49**	_
	CD at 5%	**1.20**	**1.28**	**1.48**	_

*Average of three replications

Table.2 Radial growth of *Corynespora cassiicola* at 100ppm concentration of fungicides

S. No	Treatment	Mean radial growth (mm)*on			Final Inhibition (%)
		3rd day	6th day	9th day	
1	Pyraclostrobin	12.33	24.00	42.33	51.71
2	Tebuconazole	10.33	18.00	34.00	61.21
3	Prochloraz + Tebuconazole	0.00	0.00	0.00	100
4	Azoxystrobin + Tebuconazole +Prochloraz	0.00	0.00	0.00	100
5	Pyraclostrobin+Mefentrifluconazole+ Fluxapyroxad	0.00	0.00	0.00	100
6	Tebuconazole + Sulphur	5.66	12.33	23.66	73.00
7	Carbendazim + Mancozeb	0.00	0.00	0.00	100
8	Hexaconazole	0.00	0.00	0.00	100
9	Control	46.33	77.66	87.66	_
	SE(m) ± 1	**0.31**	**0.41**	**0.47**	_
	CD at 5%	**1.05**	**1.24**	**1.41**	_

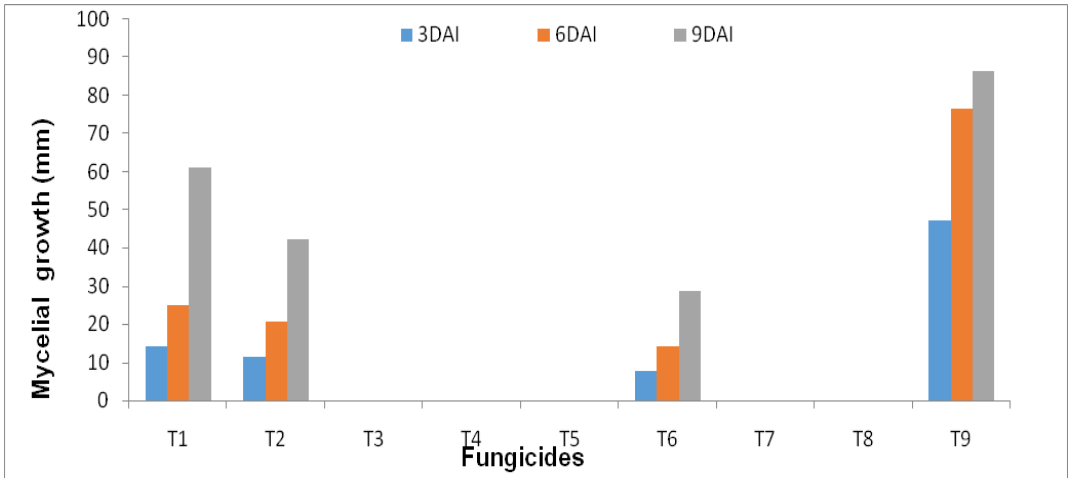
*Average of three replications

Table.3 Radial growth of *Corynespora cassiicola* at 200ppm concentration of fungicides

S .No	Treatment	Mean radial growth (mm)*on			Final Inhibition (%)
		3rd day	6th day	9th day	
1	Pyraclostrobin	11.00	17.33	26.00	70.34
2	Tebuconazole	8.00	14.66	18.33	79.08
3	Prochloraz + Tebuconazole	0.00	0.00	0.00	100
4	Azoxystrobin + Tebuconazole +Prochloraz	0.00	0.00	0.00	100
5	Pyraclostrobin + Mefentrifluconazole+ Fluxapyroxad	0.00	0.00	0.00	100
6	Tebuconazole + Sulphure	4.00	10.33	15.00	82.89
7	Carbendazim + Mancozeb	0.00	0.00	0.00	100
8	Hexaconazole	0.00	0.00	0.00	100
9	Control	46.33	77.66	87.66	_
	SE(m) ± 1	**0.29**	**0.35**	**0.36**	_
	CD at 5%	**0.88**	**1.05**	**1.10**	_

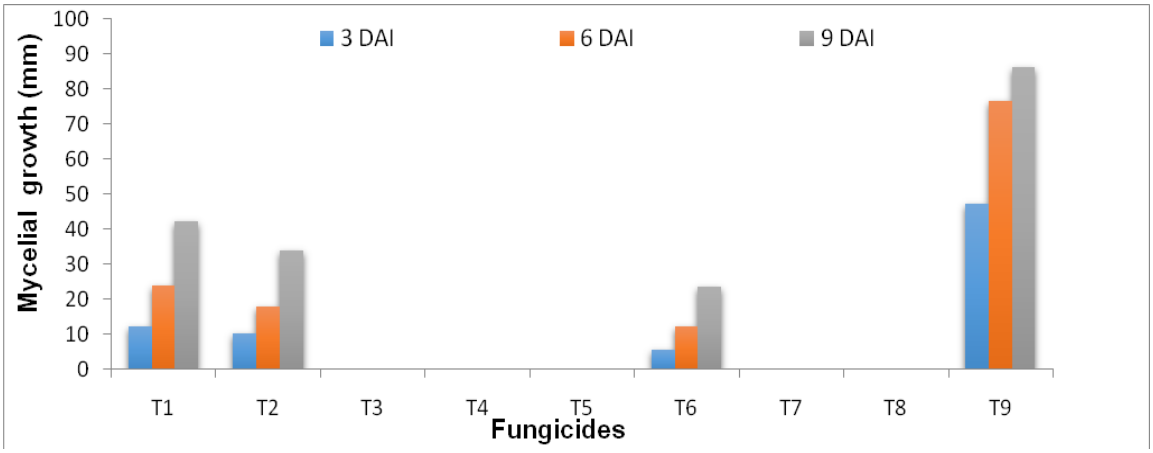
*Average of three replications

Fig.1 Mycelial growth of *Corynespora cassiicola* at 50ppm concentration of different fungicides



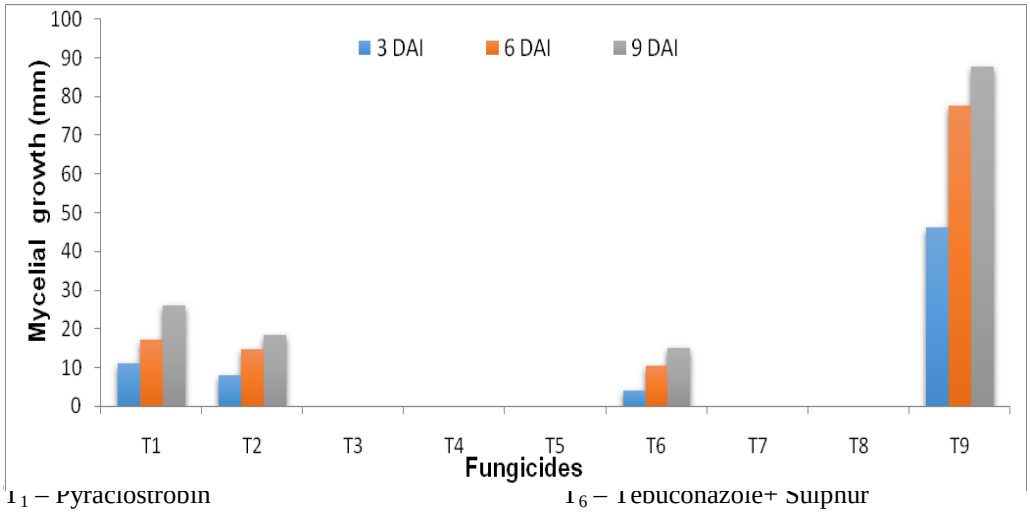
T_1 – Pyraclostrobin
T_2 – Tebuconazole
T_3 – Prochloraz+Tebuconazole
T_4 – Azoxystrobin+ Tebuconazole+ Prochloraz
T_5 – Pyraclostrobin+ Mefentrifluconazole+ fluxapyroxad
T_6 – Tebuconazole+ Sulphur
T_7 – Carbendazim+ Mancozeb
T_8 – Hexaconazole
T_9 – control

Fig.2 Mycelial growth of *Corynespora cassiicola* at 100ppm concentration of different fungicides



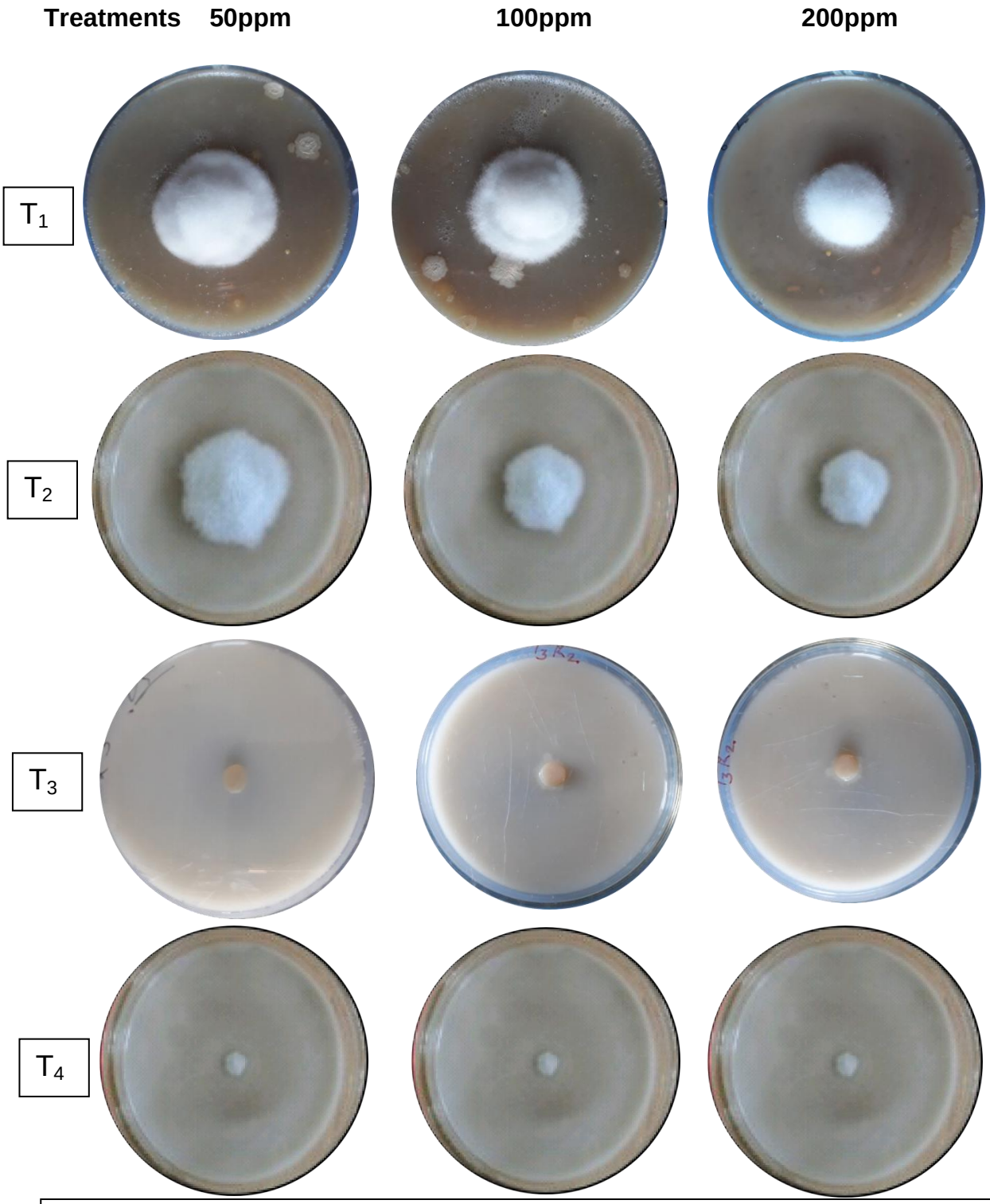
T_1 – Pyraclostrobin
T_2 – Tebuconazole
T_3 – Prochloraz+Tebuconazole
T_4 – Azoxystrobin+ Tebuconazole+ Prochloraz
T_5 – Pyraclostrobin+ Mefentrifluconazole+ fluxapyroxad
T_6 – Tebuconazole+ Sulphur
T_7 – Carbendazim+ Mancozeb
T_8 – Hexaconazole
T_9 – control

Fig.3 Mycelial growth of *Corynespora cassiicola* at 200ppm concentration of different fungicides



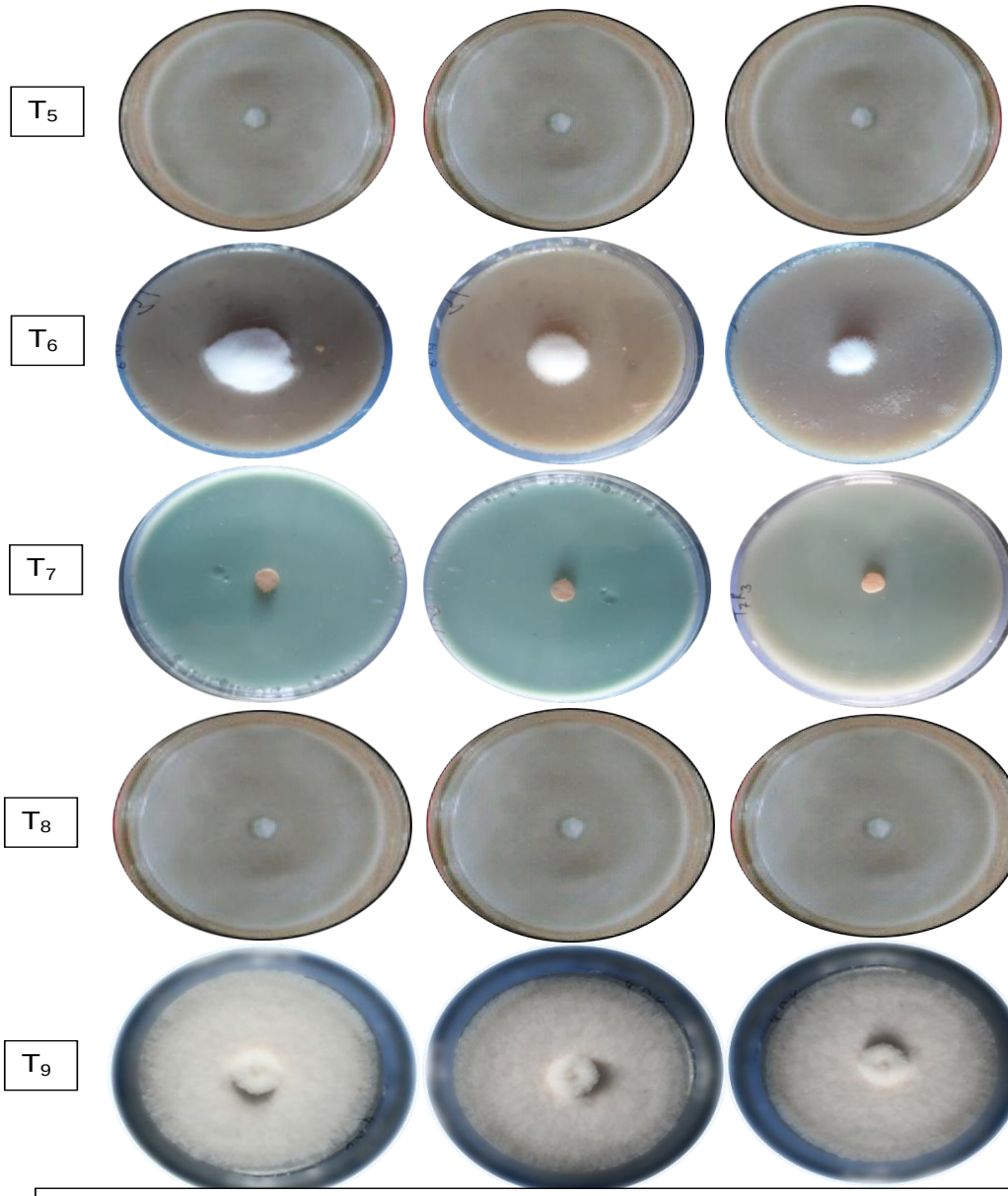
T_1 – Pyraclostrobin
T_2 – Tebuconazole
T_3 – Prochloraz+Tebuconazole
T_4 – Azoxystrobin+ Tebuconazole+ Prochloraz
T_5 – Pyraclostrobin+ Mefentrifluconazole+ fluxapyroxad
T_6 – Tebuconazole+ Sulphur
T_7 – Carbendazim+ Mancozeb
T_8 – Hexaconazole
T_9 – control

Plate.1 Mycelial growth of *Corynespora cassiicola* at 50ppm, 100ppm, 200ppm concentration of different fungicides



T_1= Pyraclostrobin T_2= Tebuconazole $T_{3=}$ Prochloraz+Tebuconazole T_4= Azoxystrobin+ Tebuconazole+ Prochloraz

Plate.2 Mycelial growth of *Corynespora cassiicola* at 50ppm, 100ppm, 200ppm concentration of different fungicides



In the present investigation, mix fungicides *viz*., prochloraz + tebuconazole, azoxystrobin + tebuconazole + prochloraz, pyroclostrobin + mefentrifluconazole + fluxapyroxad, carbendazim + mancozeb and hexconazole, completely inhibited the growth of *C. cassiicola*, at lower concentration of 50ppm. At 100ppm, Tebuconazole + sulphure reduced 73%, tebuconazole 61.21% and Pyraclostrobin 51.71%. With the increase of concentration to 200 ppm, Tebuconazole + sulphur reduced 82.89% mycelia growth, tebuconazole 79.08% and Pyraclostrobin 70.34%.

Cabral *et al.*, (2016), also demonstrated that mix fungicides, pyraclostrobin + epoxiconazole, Azoxystrobin + cyproconazole, pyraclostrobin + epoxiconazole + fluxapyroxad and procymidone significantly reduced mycelial growth of *C.cassiicola* with increase rates of fungicides from 10 and 100 mg. Similarly in 2014, Aozani found Tebuconazole at 1.89 mg/l inhibiting the mycelial growth by 50%.

Kurre *et al.*, (2017) reported complete inhibition of C. *cassiicola* by fungicides fluxapyroxad, propiconazole, tebuconazole and hexaconazole and minimum inhibition of mycelial growth was recorded by mancozeb (30.59%) and pyraclostrobin (75.37%).

References

Ahmed, F.A., Alam, N. and Khair, A. 2013. Incidence and biology of *Corynespora cassiicola* (berk. & curt.) wei. disease of okra in Bangladesh. *Bangladesh J. Bot*., 42 (2): 265-272.

Anonymous 2019. Online Agriculture Statics. Http/www.faostal.org.

Avozani, A., Reis, E.M. and Tonin, R.B. 2013. Sensitivity loss by *Corynespora cassiicola*, isolated from soybean, to the fungicide carbendazim. Summa *Phytopathol.*, 40(2): 273-276.

Cabral, W.C.; Campos, H. D.; Costa, L. S. A. S. and Simon, G. A. 2016. *In vitro* susceptibility of *Corynespora cassiicola* isolate from Brazil fields to fungicide. *Afr. J. Agric. Res*, Vol. 11(19), pp: 1699-1711.

Dhingra, O.D. and Sinclair, J.B. 1985. "Culture of Pathogens," Basic Plant Pathology Methods, CRC Press, Boca Raton, pp: 11-47.

Ellis, M. B. and Holiday, P. 1971. *Corynespora cassiicola* (Berk. &Curt.) Wei. Common wealth Mycological Institute Descriptions of Fungi and Bacteria, 31: 303.

Kurre, A. K.; Dewangan, M. and Dantre, R.K. 2017. Evaluation of fungicides against Target leaf spot caused by *Corynespora cassiicola in vitro* condition. *Int. J. Curr. Microbiol. App. Sci.* 6(8): 462-467

Nene, Y.L. and Thapliyal, P.N. 1979. Fungicides in plant disease control. Oxford and JBH Publishing Co., New Delhi. pp: 413.

Vincelli, P. 2012. QoI (Strobilurin) Fungicides: Benefits and Risks. *APS net Features*.doi:10.1094/PHI-I-2002-0809-02.

Vincent, J. M. 1947. Distribution of fungal hyphae in per cent of certain inhibitors Nature, 96: 596.

How to cite this article:

Vivek Kumar Vishwakarma, Moly Saxena and Saxena, D. R. 2020. Evaluation of Strobilurin and Triazole Fungicides against Target Leaf Spot caused by (*Corynespora cassiicola*) *in vitro* Condition. *Int.J.Curr.Microbiol.App.Sci.* 9(12): 1895-1902.
doi: https://doi.org/10.20546/ijcmas.2020.912.225