In-vitro evaluation of botanicals and fungicides against *Cercospora capsici* causing leaf spot of chilli

Abstract

One of the most significant spice crops worldwide is the chilli. It is also the most significant spice crop in India and belongs to the Solanaceae family, which has nutritional benefits and is particularly rich in vitamin C. The low yield of the country's chilli crop is caused by a variety of reasons, but fungi-induced diseases constitute a major contributor. One of the main issues with chilli cultivation in India is the fungal disease Cercospora leaf spot. In the present investigation, five botanicals *viz.*, *Azadirachta indica* leaf, *Alium cepa* bulb, *Citrus lemon* leaf, *Datura fastussa* leaf and *Mamordica charantia* leaf and five fungicides *viz.*, Mancozeb 75% WP, Carbendazim 50% WP, Difenoconazole 25% EC, Fluciazole - 40%EC and Azoxystrobin 23% SC were evaluated against the growth of *Cercospora capsici. Azadirachta indica* leaf *and Datura fastussa* leaf were found effective among all the botanicals and Difenoconazole 25% EC was found most effective among all the fungicides.

Key word: *Cercospora capsici,* botanicals, fungicides, mycelial growth, chilli leaf spot

Introduction

The chilli (*Capsicum annuum* L.), a plant belonging to the solanaceae family, is a valuable vegetable crop and spice from India. Indians had been accustomed to the chilli for about 400 years by the time the Portuguese introduced this crop to the country towards the end of the 15th century. Its cultivation became more well-liked in the 17th century. It is presently planted all over India on an approximate total area of 9,53,800 hectares. Chilli has a variety of practical uses in business. When the chilli fruit is green or ripe, which is grown as a valuable cash crop, it is used to flavour cuisine (when it is dried). Green chiles are a rich source of minerals, vitamins A and C, and protein. India is the largest producer of chillies in the world, and it distributes them to a wide wide range of countries, including Malaysia, Singapore, Germany, Saudi Arabia, the United States, Canada, and many others (Chandra Nayaka et al., 2009). The two largest states in India where chillies are cultivated are Andhra Pradesh (1.7 lakh ha) and Karnataka (0.69 lakh ha), with Maharashtra, Uttar Pradesh, Punjab, Tamil Nadu, Rajasthan, Orissa, West Bengal, and Madhya Pradesh following. The successful production of chiles is impacted by a variety of biotic limitations, infections being the most significant. Viruses like the cucumber mosaic virus, tomato spotted wilt virus, tobacco

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mosaic virus, etc., as well as fungal diseases like damping off, wilt, Cercospora leaf spot, anthracnose, powdery mildew, and others, commonly affect chillies. These diseases are in addition to the bacterial diseases (bacterial wilt and soft rot).The fungus that causes leaf spots on chilli plants belongs to the Phylum Ascomycota, Family Dematiaceae, Order Moniliales, Class Deuteromycetyes, and Family Dematiaceae. The causal organism of disease is *Cercospora capsici* and the perfect stage of pathogen is *Mycosphaerella capsici*. The term "botanical pesticides" refers to drugs that use plant extracts to kill insects and diseases. Botanical insecticides come from plants that have been shown to have pest-repelling properties. Additionally, because the bacteria in most soils are chemically very similar to the plants from which they are derived, they swiftly destroy them. Botanical pesticides can be used as substitutes for chemical pesticides. Botanical pesticides have several positive qualities, including being natural, inexpensive, target-specific, and biodegradable. These offer less of a risk to both people and the environment since, when exposed to sunlight for several hours or days, they break down into harmless chemicals.

Materials and methods

An *in-vitro* assessment of the effectiveness of plants against *Cercospora capsici*

The effectiveness of five plant extracts, including *Alium cepa* bulb, *Azadirachta indica* leaf, *Citrus lemon* leaf, *Mamordica charantia* leaf, and *Datura fastussa* leaf, *on Cercospora capsici* mycelial growth under in vitro conditions was examined in a lab experiment using the poisoned food technique described by Nene and Thapliyal (1979). The experiment was conducted at the ITM University, Gwalior's School of Agriculture and Department of Plant Pathology. Four replications of the experiment were executed in a completely randomised design. After being brought into the lab, all plants were rinsed under running water. They were surface sterilised with a 40% ethyl alcohol solution, and then cut into small pieces separately using a sterile, sharp knife. The following step involved independently grinding and homogenising each leaf sample in a mechanical grinder with an equal amount of sterile distilled water (1:1, w : v). The resulting homogenate was then strained through two layers of muslin cloth, and the filtrate was then collected and passed through Whatman No. 1 filter paper using volumetric flasks (50 ml capacity). The stock solution was made up entirely of the obtained clear leaf extracts. The stock solution of 100% was made from the obtained clear leaf extracts. To achieve the necessary concentration (10%) of extract of botanicals, an appropriate quantity of leaf extracts was added separately in the

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molten and cooled PDA medium in conical flask (250 ml capacity). After the PDA had been amended with plant extracts, sterilized Petriplates (90 mm in diameter) were filled with 15 to 20 ml per plate. All the plates were aseptically inoculated after PDA had solidified in Petriplates by introducing a 5.0 mm homogeneous mycelial disc derived from an isolated pathogen culture that had been multiplied on agar plates at the centre of each plate. Petriplates with PDA and no botanicals were inoculated with a 5.0 mm disc of the test pathogen and kept as (appropriate) untreated control. All the inoculated plates were then incubated at 25±1°C temperature in a BOD incubator until the test pathogen's mycelial growth completely covered the control plates. Data on mycelial growth for all the botanicals and control was recorded at 7 and 14 Days after inoculation (DAI). The per cent mycelium inhibition was calculated by using following formula

$$PMH = \frac{a-b}{A} \times 100$$

Where,

PMH = Per cent mycelium inhibition

a= Mycelium growth in control

b= Mycelium growth in botanical

Evaluation of fungicides against *Cercospora capsici* under *In-vitro* condition

In the present investigation, the effectiveness of fungicides against *Cercospora capsici* was evaluated using the approach of poisoned food (Nene and Thapliyal, 1979). All of the fungicides, including Mancozeb 75% WP, Carbendazim 50% WP, Difenoconazole 25% EC, Fluciazole - 40%EC and Azoxystrobin 23% SC were evaluated at a concentration of 500 ppm. The experiment was carried out in Completely Randomized Design with four replications. The required quantity of each fungicide was added separately to molten, sterile Potato dextrose agar before it cooled in order to reach the proper fungicide concentration. In sterilized Petriplates, approximately 20 ml of the poisoned medium was then added. A fungus culture that was actively growing was placed in the centre of each plate, with mycelial discs measuring 5 mm in diameter. Control was maintained without the use of any fungicide in the medium. A BOD incubator was used to incubate the inoculated petri plates at a temperature of 25±1 °C. At 7 and 14 DAI, data on the mycelial development was gathered from all the

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fungicides and the control. The per cent mycelium inhibition was calculated by using following formula

$$PMH = \frac{a-b}{a} \times 100$$

Where,

PMH = Per cent mycelium inhibition

a= Mycelium growth in control

b= Mycelium growth in fungicide

RESULT AND DISCUSSION

Evaluation of botanicals against *Cercospora capsica* under *in-vitro* condition

An experiment was conducted in the laboratory of Plant Pathology for evaluation of five botanicals against *Cercospora capsici*. Data presented in table-1 showed that all the botanical significantly inhibited the mycelial growth of *Cercospora capsici* at 7 days after inoculation. Significantly minimum mycelial growth of pathogen was recorded in *Azadirachta indica* leaf (20.50 mm) which was found at par with *Datura fastussa* leaf (23.25 mm).This was followed by *Alium cepa* bulb (27.00 mm), *Citrus lemon* leaf (36.50 mm) and *Mamordica charantia* leaf (38.50 mm), while maximum mycelial growth was recorded in Control (47.75 mm). At 14 days after inoculation, minimum mycelial growth of pathogen was recorded in *Azadirachta indica* leaf (36.25 %) which was found at par with *Datura fastussa l*eaf (38.50 mm). *Alium cepa* bulb (43.00 %) was the next best botanical followed by *Citrus lemon* leaf (51.75 mm) and *Mamordica charantia* leaf (59.00 %), while maximum growth of pathogen was recorded in control (64.25 mm). Neindow *et al.* (2020) reported that the aqueous extracts of desert date seed at 100 g/l strongly suppressed the highest mycelial growths The incidence, severity, and defoliation of disease in the field were all significantly reduced by three of the four aqueous extracts (desert date seed, neem seed, and jatropha seed) at 100 g/l while yield was also raised. According to Ambang *et al*. (2011), *Cercospora* decreased in rate of spread as *Thevetia peruviana* seed extract content was increased. According to Haller *et al*. (2013), *P. juliflora* and *D. metel* leaf extracts recorded 50.34 and 31.66 percent less mycelial development than the control, respectively. *Allium* sp. extract completely inhibited mycelial growth, they found. Alam *et al.* (2010) reported that the leaf

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and seed extracts of *A. indica* both exhibited strong inhibitory of *F. oxysporum* f. sp. *capsici*, *R. artocarpi*, and *A. tenuis*

Table 1: Evaluation of botanicals against *Cercospora capsica* under *in-vitro* condition

Botanicals	7 DAI		14 DAI	
	Mycelial growth (mm)	Mycelium inhibition (%)	Mycelial growth (mm)	Mycelium inhibition (%)
Azadirachta indica leaf	20.50	57.07	36.25	43.58
Alium cepa bulb	27.00	43.46	43.00	33.07
Citrus lemon leaf	36.50	23.56	51.75	19.46
*Datura fastussa l*eaf	23.25	51.31	38.50	40.08
Mamordica charantia leaf	38.50	19.37	59.00	8.17
Control	47.75	0.00	64.25	0.00
SE.m±	1.18	-	1.31	-
C.D. at 5 %	3.51	-	3.89	-

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4.6: Evaluation of fungicides against *Cercospora capsica* under *in-vitro* condition

An experiment was conducted in the laboratory of Plant Pathology for evaluation of five fungicides against *Cercospora capsici*. At 7 days after inoculation, minimum mycelial growth of pathogen was recorded in Difenoconazole 25% EC (9.25 mm) which was found at par with Fluciazole 40%EC (10.25 mm) followed by Carbendazim 50% WP (12.75 mm), Azoxystrobin 23% SC (14.25 mm) and Mancozeb 75% WP (17.00 mm), while maximum mycelial growth of pathogen was recorded in control (46.50 mm). At 14 days after inoculation, minimum mycelial growth was recorded in Difenoconazole 25% EC (14.25 mm) which was found significantly superior over other fungicides. Fluciazole 40%EC (19.50 mm) was the next best fungicides followed by Carbendazim 50% WP (22.50 mm), Azoxystrobin 23% SC (27.25 mm) and Mancozeb 75% WP (33.00 mm), while highest mycelial growth was recorded in Control (68.50 mm). Pun *et al*. (2020) reported that Hexaconazole and carbendazim completely prevented the fungus's mycelial growth. A field experiment was done to determine the best effective treatment for the chilli plant disease Cercospora leaf

spot. The findings showed that the lowest percent disease index was achieved when hexaconazole 5 percent SC. Singh *et al*. (2011) reported that the Bavistin significantly suppressed the radial growth of pathogenic fungus

Table 2: Evaluation of fungicides against *Cercospora capsica* under *in-vitro* condition

Fungicides	7 DAI		14 DAI	
	Mycelial growth (mm)	Mycelium inhibition (%)	Mycelial growth (mm)	Mycelium inhibition (%)
Mancozeb 75% WP	17.00	63.44	33.00	51.82
Carbendazim 50% WP	12.75	72.58	22.50	67.15
Difenoconazole 25% EC	9.25	80.11	14.25	79.20
Fluciazole - 40%EC	10.25	77.96	19.50	71.53
Azoxystrobin 23% SC	14.25	69.35	27.25	60.22
Control	46.50	0.00	68.50	0.00
SE.m±	0.91	-	1.26	-
C.D. at 5 %	2.71	-	3.75	-

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Conclusion

In the present investigation, botanicals and fungicides were evaluated against *Cercospora capsici* under in-vitro condition. *Azadirachta indica* leaf *and Datura fastussa* leaf were found effective among all the botanicals and Difenoconazole 25% EC was found most effective among all the fungicides.

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