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RESEARCH PAPER

Evaluation of different fungicides against *Alternaria solani* causing early blight disease of potato under *in vitro* conditions

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Abstract: Potato (*Solanum tuberosum*) is a widely cultivated starch crop known for its edible underground storage organ, providing essential nutrients globally. Early blight, caused by the fungal pathogen *Alternaria solani* can lead to significant yield losses in potato crop, reports indicated that average annual yield losses due to early blight range from approximately 50 per cent to 75 per cent of production. Chemical protection have been a prominent approach in controlling early blight. Under *in vitro* evaluation fungicides, among non-systemic fungicides, mancozeb 75% WP exhibited the highest mean mycelial growth inhibition at 96.25 per cent, followed by thiram 75% WP with a mean mycelial growth inhibition of 92.51 per cent. Among six systemic fungicides evaluated, hexaconazole 5% EC and tebuconazole 25.9% EC recorded cent per cent mycelial growth inhibition at all four concentrations (0.025, 0.05, 0.1, 0.15%) tested which were significantly superior over rest of fungicides followed by difenconazole 25% EC. Among seven combi product fungicide tested, (captan 70% + hexaconazole 5%) 75% WP showed the mycelial growth inhibition of 100 per cent at all three concentrations tested, significantly over other combi-product fungicides.

Key words: *Alternaria solani*, Early blight, Percent mycelial growth inhibition, Potato

Introduction

The potato (*Solanum tuberosum* L.) ranks third in global significance as a human food crop, trailing only rice and wheat. Originating in the Andean highlands of South America, it spread to other continents through European dissemination. There is evidence suggesting that it was likely introduced to India in the early 17th century, possibly by Portuguese traders or British missionaries (Pushkarnath, 1976). Revered as the "king of vegetables," the potato holds a vital place in India's vegetable markets. Thanks to its high dry matter and edible protein content, the potato stands out as a nutritionally superior vegetable and one of the world's essential food crops (Bansode *et al.,* 2018).

The potato is a carbohydrate-rich food that provides energy while being low in fat. Although its protein content is relatively modest, it boasts an excellent biological value of 90-100. Rich in vitamin C, several B vitamins and potassium, potatoes offer valuable nutritional benefits. The skins, in particular provide a good source of dietary fiber. Additionally, potatoes contain various compounds that contribute to antioxidant activity (Camire, 2009).

Despite its potential for higher production, the potato crop faces challenges due to attacks from various phytopathogens, which limit its productivity. The potato plant is susceptible to several fungal diseases, including Anthracnose (*Colletotrichum phomoides*), Early blight (*Alternaria solani*), Fusarium wilt (*Fusarium oxysporum*), Late blight (*Phytophthora infestans*), and Leaf blight (*Alternaria alternata*) (Sharma *et al*., 2020).

Early blight, caused by the fungal pathogen *Alternaria solani*, can lead to significant yield losses in potato crops. Reports indicate that average annual yield losses due to early blight range from approximately 50 per cent to 75 per cent of production (Murmu *et al.,* 2017). However, values in the literature for measured crop losses due to early blight vary widely, ranging from 5 per cent to 78 per cent (Waals *et al*., 2004).

Early blight is characterized by the appearance of dark brown to black lesions with concentric rings, resembling a target board, which can significantly reduce the market quality of the potatoes. The affected leaves develop circular to angular dark brown lesions, typically measuring 3-4 mm in diameter. The disease tends to affect stressed or senescing plants, with the oldest foliage being the first to show symptoms. Severely affected leaves turn yellow and start drooping. Infected tubers may display a dark, corky dry rot, with brown, round to irregular, depressed lesions on the surface. The flesh beneath these lesions also turns dry, brown and corky (Sharma *et al.,* 2020).

Material and methods

The research on early blight disease of potato was conducted during the period of 2022-23 at the Department of Plant Pathology, College of Agriculture, University of Agricultural Sciences, Dharwad. In the experiment effectiveness of systemic, non-systemic and combi fungicide products were evaluated using the poison food technique. Before conducting the experiment pathogen, *Alternaria solani* was cultured on Potato Dextrose Agar (PDA) medium in Petri plates for a duration of ten days.

Poisoned food technique

The poisoned food technique (Nene and Thapliyal, 1993) was followed to evaluate the efficacy of fungicides in inhibiting the mycelium growth of *Alternaria solani*. The PDA medium was prepared and melted. The fungicidal suspension was added to the melted media to obtain the required concentrations about 20 ml of poisoned medium was poured in each sterilized Petri Plates. Suitable check was maintained without addition of fungicide. Eight mm mycelia disc was cut with the help of the sterilized cork borer and it was placed aseptically in the middle of Petri plate and incubated at room temperature for 7 days. Three replications were maintained for each treatment. The diameter of the colony was measured after reaching maximum growth in control plates. The Percent growth inhibition of the pathogen was calculated using the formula given by Vincent (1947) as follows

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

Where,

I= Per cent inhibition

C = Growth of mycelium in control

T = Growth of mycelium in treatment.

Results and discussion

In the laboratory evaluation, a total of six non-systemic and six systemic fungicides, along with seven combi product fungicides, were tested at varying concentrations for their effectiveness against *Alternaria solani*. In the evaluation of fungicides, all the tested fungicides showed significant superiority over the control in terms of mycelial inhibition.

Table 1. Efficacy of non-systemic fungicides on inhibition of mycelial growth of *Alternaria solani*

Fungicides	Percent mycelial growth inhibition			Mean
	Concentrations (%)			
	0.1	0.2	0.3	
Mancozeb 75%WP	88.76 (70.42) *	100 (89.71) *	100 (89.71) *	96.25 (83.29) *
Propineb 70% WP	73.03 (58.72)	78.65 (62.49)	84.26 (66.64)	78.65 (62.61)
Cupper oxy chloride 50% WP	85.39 (67.54)	86.52 (68.46)	86.52 (68.46)	86.14 (68.16)
Zineb 78%WP	39.33 (38.84)	53.93 (47.26)	84.27 (66.64)	59.18 (50.91)
Chlorothalonil 75%WP	21.35 (27.52)	24.72 (29.80)	32.58 (34.80)	26.22 (30.71)
Thiram75%WP	77.53 (61.71)	100 (89.71)	100 (89.71)	92.51 (80.38)
Mean	64.23 (54.12)	73.97 (64.57)	81.27 (69.33)	73.16 (62.67)
	S.Em.±	C.D. at 1%		
Fungicides (F)	0.17	0.69		
Concentration (C)	0.123	0.49		
F×C	0.35	1.20		

*Angular transformed values

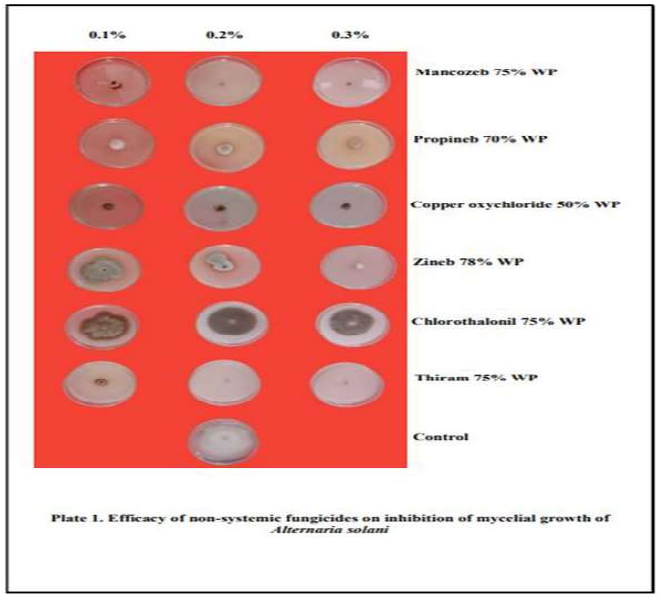

Plate 1. Efficacy of non-systemic fungicides on inhibition of mycelial growth of *Alternaria solani*

Among the non-systemic fungicides tested at three concentrations (0.1, 0.2 and 0.3%), treatments using mancozeb 75% WP exhibited the highest mycelial growth inhibition at all three concentrations (88.76%, 100%, 100%, respectively), which was significantly superior to other treatments. Following this, thiram 75% WP showed mycelial growth inhibition at 77.53 percent (at 0.1%), 100 percent (at 0.2%) and 100 percent (at 0.3%). On the other hand, chlorothalanil 75% WP showed the lowest mycelial growth inhibition at 21.35 per cent (at 0.1%).Regardless of the tested fungicide concentrations, mancozeb 75% WP recorded the highest mean mycelial growth inhibition (96.25%), followed by thiram 75% WP (92.51%), while chlorothalanil 75% WP exhibited the least mean mycelial growth inhibition at 26.22 per cent. The detailed data tabulated in Table 1, illustrated on Plate 1 and presented in Fig.1

Mancozeb 75% WP functions by inactivating the sulfhydryl groups of amino acids there by interrupting the enzymatic activities within the fungal cell. This action prevents fungal spores from germinating and disrupts lipid metabolism and respiration. It also enhances plant immunization and partially or completely degrades the cell wall of the fungal pathogen. These combined effects are likely the reason for the observed inhibition in the growth of the test fungus.

Similarly, Deshmukh *et al.* (2020) observed cent per cent inhibition of mycelial growth of *Alternaria solani* by mancozeb 75% WP at 0.2 per cent. Chapei *et al.* (2019) reported that mancozeb 75% WP at 0.2 per cent concentration inhibited 93.33 per cent of mycelial growth. Arunakumara and Satyanarayana (2018), Yadav *et al.* (2018) and Misba *et al.* (2022), reported effectiveness of mancozeb 75% WP against *Alteranria solani* in lab conditions.

In the evaluation of systemic fungicides at four different concentrations (0.025, 0.05, 0.1, 0.15%) both hexaconazole 5% EC and tebuconazole 25.9% EC demonstrated cent percent mycelial growth inhibition at all tested concentrations, which was significantly superior to the other fungicides.

Table 2. Efficacy of systemic fungicides on inhibition of mycelial growth of *Alternaria solani*

Fungicides	Percent mycelial growth inhibition				Mean
	Concentrations (%)				
	0.025	0.05	0.1	0.15	
Thiophanate methyl 70%WP	21.35(27.52)*	26.97(31.28)*	41.57(40.15)*	71.91 (58.09)*	40.45(39.26)*
Difenconazole 25%EC	95.25(77.85	96.25(80.87)	98.76(83.00)	100(89.71)	97.56(82.86)
Propiconazole 25%EC	92.50975.465	94.3878.24	97.64(82.08)	98.76(81.95)	95.82(79.43)
Carbendazim 50% WP	43.82(41.45)	44.94(42.09)	55.05(47.90)	60.67 (51.17)	51.12(45.65)
Tebuconazole 25.9% EC	100(89.71)	100(89.71)	100(89.71)	100(89.71)	100.00(89.71)
Hexaconazole 5%EC	100(89.71)	100(89.71)	100(89.71)	100(89.71)	100(89.71)
Mean	75.48(66.95)	77.09(68.64)	82.17(72.09)	88.55(76.72)	80.82(71.1)
	S.Em.±	C.D. at 1%			
Fungicides (F)	1.24	4.96			
Concentration (C)	1.03	4.05			
F×C	2.89	9.92			

*Angular transformed values

Difenconazole 25% EC also showed cent percent inhibition at 0.15 per cent concentration. Thiophanate methyl 70% WP exhibited the least mycelial growth inhibition at different concentrations, with values of 21.35 per cent (at 0.025%), 26.97 per cent (at 0.05%), 41.57 per cent (at 0.1%) and 71.91 percent (at 0.15%). Irrespective of the concentrations, the mean mycelial growth inhibition was highest in hexaconazole 5% EC and tebuconazole 25.9% EC (100%), followed by difenconazole 25% EC (97.56%) and propiconazole 25% EC (95.82%). The detailed data tabulated in Table 2 and is illustrated on Plate 2.

Triazoles belong to a potent group of fungicides with strong inhibitory action on ergosterol synthesis. They block cytochrome P-450 dependent enzymes and C-14 alpha de-methylase, essential for converting lanosterol to ergosterol. Ergosterol is critical for forming fungal cell walls. The lack of normal sterol production hinders fungal growth, preventing further infection or invasion of host tissues. This mode of action makes triazoles effective in controlling fungal diseases by disrupting essential processes vital for the fungus survival and spread.

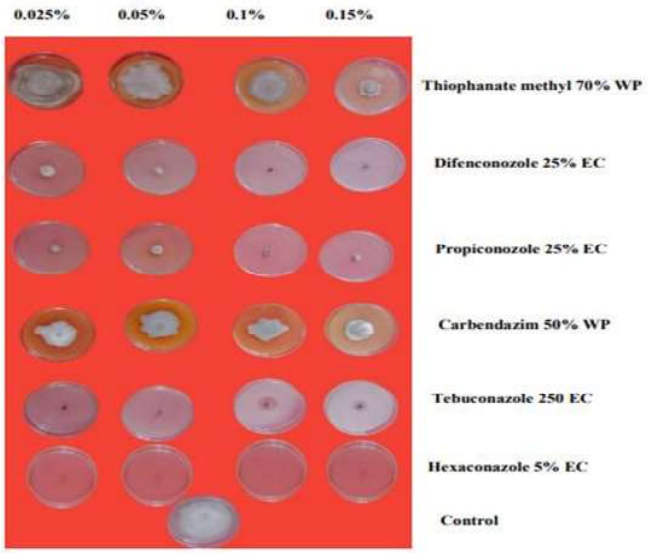

Plate 2. Efficacy of systemic fungicides on inhibition of mycelial growth of *Alternaria solani*

Similar observations were reported by Yadav *et al.*(2018) and Chapei *et al.*(2019) found hexaconazole is effective against *Alternaria solani* . Dhaka and Choudary (2022) and Misba *et al.* (2022), reported effectiveness of propiconazole and tebuconazole. The obtained results are in accordance with the findings made by Blessina *et al.*(2022), Gule *et al.*(2021), Sudarshan *et al.* (2020) and Sowmya and Chandra (2021) observed that tebuconazole was the most effective fungicide followed by difenconazole. Herle 2014, who reported that triazole fungicides were effective in inhibiting the growth of *Alternaria solani.*

In the evaluation of combi-products at three different concentrations (0.1, 0.2 and 0.3%), (captan 70% + hexaconazole 5%) 75% WP exhibited mycelial growth inhibition of 100 per cent at all three concentrations and it was significantly superior to the other combi product fungicides tested. Followed by (tebuconazole 50% + trifloxystrobin 25%) 75% WG and (azoxystrobin 18.2% + difenoconazole 11.4%) SC also showed mycelial growth inhibition of 97.08 per cent and 96.33 per cent

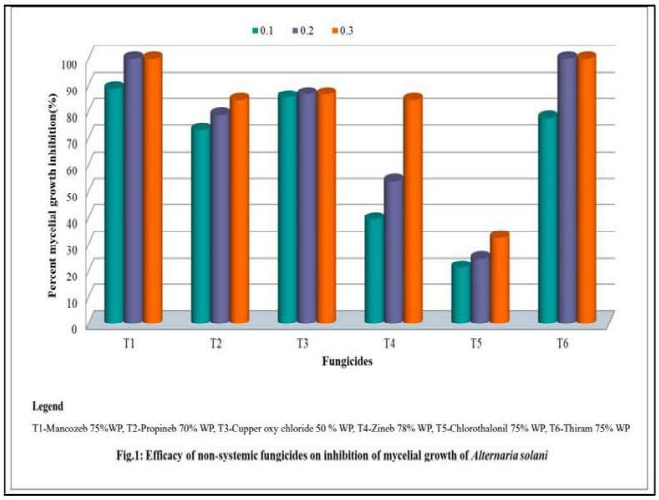

Fig.1: Efficacy of non-systemic fungicides on inhibition of mycelial growth of *Alternaria solani*

Table 3. Efficacy of combi product fungicides on inhibition of mycelial growth of *Alternaria solani*

Fungicides	Percent mycelial growth inhibition			Mean
	Concentrations (%)			
	0.1	0.2	0.3	
(Zineb 68 % + Hexaconazole 4 %) 72% WP	85(67.38)*	85.5(67.71)*	88.25(70.29)*	86.25(68.46)*
(Captan 70%+Hexaconazole 5%) 75%WP	100(89.71)	100(89.71)	100(89.71)	100(89.71)
(Metiram 55% +Pyraclostrobin 5%) 60%WG	56.18(48.57)	59.55(50.53)	66.29(54.53)	60.67(51.18)
(Carbendazim 12% + Mancozeb 63 %) 75%WP	85.00(67.58)	86.25(68.35)	87.07(69.43)	86.11(68.45)
(Carbondazim 25%+mancozeb 50%) 75% WS	78.65(62.70)	85.00(67.41)	86.25(68.41)	83.30(66.18)
(Tebuconazole 50 % + Trifloxytrobin 25%) 75%WG	96.25(78.72)	97.25(80.45)	98.76(82.35)	97.08(80.51)
(Azoxystrobin18.2%+Difeconazole11.4%)29.26% SC	95.25(79.42)	96.05(78.54)	98.02(83.36)	96.33(80.44)
Mean	85.19(70.58)	87.08(71.82)	89.23(74.01)	87.17(72.14)
	S.Em.±	C.D. at 1%		
Fungicides (F)	1.24	4.97		
Concentration (C)	0.81	3.25		
F×C	2.51	8.61		

*Angular transformed values

respectively. Regardless of the concentration (metiram 55% + pyraclostrobin 5%) 60% WG demonstrated the least mean mycelial growth inhibition of 60.67 per cent. The detailed data tabulated in Table 3 and is illustrated on Plate 3.

The results are in agreement with Dhaka and Choudhary (2022) also reported (captan 70% + hexaconazole 5%) 75% WP with 79.40 per cent mycelia inhibition against *A. solani*.

The mode of action of captan involves its multisite contact activity, meaning it affects multiple sites within the fungal cells. It targets various enzymes, including those containing SH (sulfhydryl) groups. Additionally, captan interferes with the oxidative process, specifically Co-carboxylase. The combination of multisite activity and low resistance risk makes captan an effective and sustainable fungicide for disease management in agriculture.

Combi-product fungicides offer a strategy to mitigate the development of resistance in pathogens against systemic fungicides. Systemic fungicides typically target only one or a few specific functions in fungal physiology making it easier for pathogens to overcome their effects through a single mutation or the selection of resistant individuals in the population. On the other hand, non-systemic protectant fungicides affect multiple functions in fungal physiology, requiring numerous gene changes for the fungus to develop resistance.

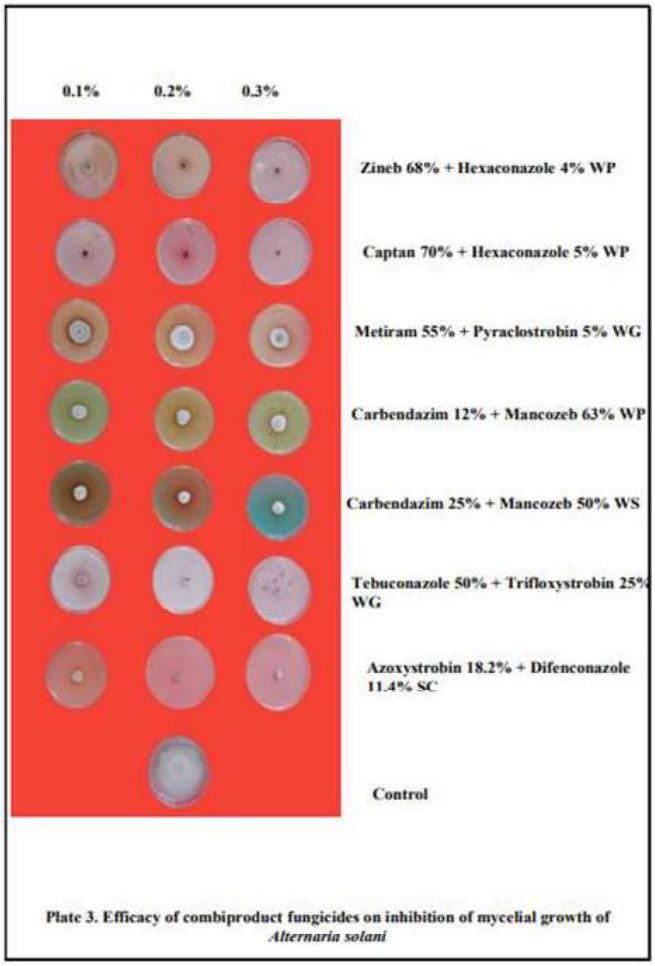

Plate 3. Efficacy of combiproduct fungicides on inhibition of mycelial growth of *Alternaria solani*

Conclusion

Among six non-systemic fungicides tested mancozeb 75% WP resulted in the highest mean mycelial growth inhibition of 96.25 per cent, followed by thiram 75% WP with a mean mycelial growth inhibition of 92.51 per cent. Among six systemic fungicides hexaconazole 5% EC and tebuconazole 25.9% EC showed the highest mean mycelial growth inhibition of 100 per cent followed by difenconazole 25% EC with a mean inhibition of 97.56 per cent, while propiconazole 25% EC exhibited mean inhibition of 95.82 per cent. Among seven combi product fungicide tested, (captan 70% + hexaconazole 5%) 75% WP demonstrated the highest mycelial growth inhibition of 100 per cent at all three concentrations tested, significantly superior than other combi-product fungicides.

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