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EVALUATION OF FUNGICIDES AGAINST LEAF SPOT DISEASE OF APPLE (*Malus domestica cv. red delicious*) CAUSED BY *Alternaria mali* ROBERTS

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ABSTRACT
Present experiment was conducted to evaluate different fungicides for controlling *Alternaria* leaf spot of apple. Seven different treatments were evaluated *in vitro* and *in vivo* conditions., *In vitro* result revealed that Hexaconazole at concentrations 250,500,750 and 1000 ppm inhibited 100% growth of test pathogen concentration of followed by Difenconazole (72%, 88.9%, 95% and 100%) at 250, 500, 750 and 1000ppm respectively. Carbendazim (100%) at 750 and 1000 ppm. The least percent growth inhibition was found in case of Mancozeb (5.5%) at 250ppm. *In vivo* results revealed that Mancozeb + Carbendazim (0.025%) was found most effective and recorded significantly least disease incidence (20.05%) with per cent yield increased over check (6.03%) followed by Hexaconazole (5.25%), Difenoconazole (5%) and Copper oxy Chloride (4.06%). Hexaconazole is found second most effective for the control of *Alternaria* leafspot of apple.

INTRODUCTION

Apple (*Malus domestica)* is a delicious and an important temperate fruit crop grown throughout the world wherever agro-climatic conditions are met for its cultivation. It belongs to the family *Rosaceae* and is a native of Turkey. In India it is commercially grown in the states of Himachal Pradesh, Jammu & Kashmir, Kumaon hills of Uttarakhand and parts of Sikkim and Arunachal Pradesh on an area of 319770 hectares with annual production of 1884730 tonnes (NHB, 2016). Uttarakhand has 3rd position in area (33910 hectares) with annual production of 91470 tonnes (NHB, 2016). The total value of apple produced in the state is 64281 lakhs at current prices of 2011-12. The area under apple cultivation has increased manifold during the last few decades in India but the production has not increased proportionally and the productivity is quite low in comparison to advanced apple growing countries of the world.

Alternaria leaf spot caused by *Alternaria mali* is an important pathogen on apple in India as it is reported to cause epidemics in Kashmir in 2013(Bhat *et al*., 2015). It can infect up to 85% of leaves on susceptible cultivars, compared with less than 1% on resistant cultivars. It leads to destruction of leaves before the fruits getting maturity that leads to reducing the yield as well as fruit quality in rare cases the fruits are also infected and reduced the value of apple in market. Certain reports based on grower's estimates have reported losses of 40-60% and the disease considerably reduce the market value of apple from grade "A" to grade "C". Primary infection takes place about one month after petal fall. The disease advances rapidly in the optimum temperature range of 25-30^{0}C and wet weather. At optimum temperatures, infection occurs with 5.5h of wetting, and lesions can appear in the orchards two days after infection, causing a serious outbreak (Yoon & Lee, 1987).

The fungus also produces a chemical toxin which increases the severity of disease on susceptible cultivars. Different chemicals are tested against the pathogen in different parts of the world (Yoon & Lee, 1987).

So, by keeping these points in mind an experiment has been conducted on "evaluation of different fungicides against leaf spot disease of apple (*Malus domestica cv*. Red delicious) caused by A*lternaria mali* Roberts". The objective of the present investigation was to evaluate the efficacy of selected fungicides.

MATERIALS AND METHODS

Among the different varieties of apple Red Delicious is among the most susceptible to *Alternaria* leaf spot (Filajdic and Sutton, 1991). The experiment was conducted at fruit research block College of horticulture Bharsar, Uttarakhand during 2016 season.

Isolation and Identification of pathogen

Alternaria mali is isolated from infected leaves of apple by using single spore isolation technique (Johnston and Booth, 1983) and pure culture was maintained by sub-culturing. Pathogenicity test was done by detached leaf experiment (Sofi

et al., 2012) for the confirmation of the pathogen. Re-isolated pathogen is identified by using characters given by Roberts (1924).

In vitro bioassay of fungicides

The efficacy of different contact fungicides viz., Mancozeb instead Copper oxy Chloride, Hexaconazole, Carbendazim, Difenconazole, Flusilazole and Mancozeb + Carbendazim (75% WP) at concentrations of 250, 500, 750 and 1000 ppm were tested by using poisoned food technique (Nene Y L and Thapiyal,1979). Three replication for each concentration was also kept. Radial growth of test pathogen is recorded after 7 days of incubation at 25 ± 1^{0}C.

Percent growth inhibition by fungicides was calculated as follows (Bliss, 1934).

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

Where,

PGI = Percent Growth Inhibition, C = Colony diameter in control (mm), T = Colony diameter in treatments (mm).

In vivo study

Field experiment was conducted in 2016 on red-delicious variety with three replication per treatment. Three prophylactic sprayings of fungicides at 0.25 % (Table 1) were done at 30 days interval. Percent disease index was noted after last spray of fungicides and calculated as follows (Wheeler, 1969).

$$\text{Percent disease index (PDI)} = \frac{\sum \text{All rating grades}}{\text{Total Number of leaves observed}} \times \frac{100}{\text{Maximum grade}}$$

Rating scale

Five plants were selected randomly in each plot and observation on severity of the disease on the foliage was recorded using 0-5 scale developed by Horsefall and Barret, 1945 as given below 0 (Leaves free from infection) , 1 (Small irregular spots covering <5% leaf area), 2 (Small irregular brown spots with concentric rings covering 5.1-10% leaf area) , 3 (Lesions enlarging, irregular brown with concentric rings covering 10.1-25% leaf area) , 4 (Lesions coalesce to form irregular and appears as a typical blight symptom covering 25.1-50% leaf area) and 5 (Lesions coalesce to form irregular and appears as a typical blight symptom covering >50% leaf area).

RESULTS AND DISCUSSION

Invitro study

In *in vitro* study it was revealed that 100% percent inhibition was observed in case of Hexaconazole at all concentrations (250, 500, 750 and 1000 ppm), Difenconazole (1000 ppm), Mancozeb+ Carbendazim (750, 1000 ppm). Then next effective fungicides in controlling radial growth of *A. mali* were Difenoconazole (95% and 88.8%) at 500 and 750 ppm, Copper oxy Chloride (86.67 %) at 1000 ppm, Mancozeb+Carbendazim (85%) at 500 ppm) and Flusilazole (81.11%) at 100 ppm (Table-1).

These results are in agreement with following workers reported earlier that Hexaconazole inhibited radial growth of *Alternaria* spp. at different concentrations *in vitro*. Verma and Verma (2010) reported that Hexaconazole at 250 ppm, 500 pmm , 1000ppm, 2000 ppm and 2500 ppm inhibited radial growth of *Alternaria alternata* causing blight of tomato. Meena and Ratnoo (2014) also reported that Hexaconazole at 1000 ppm reduced mycelial growth of *Alternaria alternata*, *Alternaria macrospora* and *Alternaria gossypina* nearly cent percent (97.64 %, 97.16 % and 97.78 %) causing leaf spot in cotton.

In vivo study

Disease intensity

The least percent disease index (20.05%) as well as highest percent disease (66.58) was shown by Mancozeb + Carbendazim followed by Hexaconazole (27.69% and 53.85%) and Copper oxy Chloride (35.08 and 41.54). The highest PDI (54.61) as well as least percent disease control (9) was found in case of Carbendazim among different fungicides. In control it was observed PDI of 60 (Table-3).

These results are in agreement with Shahzad A (2009) reported that Hexaconazole and copper oxy chloride are the best fungicides for control of leaf spot and fruit spot of apple caused by *A. mali* in field.

Fruit yield

Mancozeb + Carbendazim showed a fruit highest yield of

Table 1: Effect of treatments at 250, 500, 750 and 1000 ppm on radial growth of *Alternaria mali*

Sr. No.	Treatments	Percent Growth Inhibition (PGI) **				Mean
		250 ppm	500 ppm	750 ppm	1000 ppm	
1	Mancozeb	5.5 (13.56) *	30 (33.20)	50 (44.99)	61.11 (51.42)	36.65 (35.79)
2	Copper oxy chloride	16.67 (24.33)	22.22 (28.12)	77.77 (61.87)	86.67 (68.62)	50.83 (45.73
3	Hexaconazole 5% EC	100 (90)	100 (90)	100 (90)	100 (90)	100 (90)
4	Carbendazim 50% WP	17 (23.57)	40 (39.23)	56 (48.44)	72 (58.05)	46.25 (42.32)
5	Difenoconazole 25% EC	72 (58.05)	88.88 (70.53)	95 (77.12)	100 (90)	88.97 (73.92)
6	Mancozeb + Carbendazim	60 (50.76)	85 (67.48)	100 (90)	100 (90)	86.25 (74.56)
7	Flusilazole 40% EC	44 (41.55)	70 (56.79)	72.22 (58.19)	81.11 (64.24)	66.83 (55.19)
	Mean	45.02 (43.12)	62.3 (55.05)	78.71 (67.23)	85.84 (73.19)	66.97 (59.64)
					S.E (d)	C.D @ 5%
Fungicides (F)					0.28	0.79
Concentration (C)					0.21	0.60
F*C					0.56	1.59

*Arc sin transformed values**in comparison with control showing 90.0 mm growth

Table 2: Effect of treatments on disease intensity and fruit yield

Sr. No.	Treatments	Concentration(ppm)	Percent disease index	Fruit yield(t/ha)
1	Mancozeb	500	40.30 (39.38)*	25.80
2	Copper oxy chloride	200	35.08 (36.29)	26.02
3	Hexaconazole	500	27.69 (31.72)	26.25
4	Carbendazim	250	54.61 (47.62)	25.50
5	Difenoconazole	150	37.20 (37.56)	26.08
6	Mancozeb + Carbendazim	250	20.05 (26.57)	26.51
7	Flusilazole	100	42.01 (40.38)	25.95
8	Control	-	60.01 (50.75)	25.00
	C.D. @ 5%		0.16	0.23
	SEm ±		0.05	0.78

*Angular transformed value

26.5 q/ha followed by Difenconazole (26.07 q/ha) and Copper oxy Chloride (26.01) as compared to control (25t/ha).

Kwon *et al*. (2015) also reported the effect of leaf spot diseases in apple and their effect on per fruit weight of apple. He found that individual fruit weight is reduced from 330 gram to 282 g by defoliation of trees ranging from 0.0% to 60% and a reduction of 0.01kg per fruit was noticed in over all fruit weight in different areas of Korea.

Phytotoxicity

All the fungicides tested did not cause any phytotoxicity symptoms in terms of chlorosis, necrosis, wilting, scorching, hyponasty and epinasty on 1, 3, 5, 7 and 10 days after fungicide application.

This result was in agreement with Sahu *et al*. (2014) reported that phytotoxicity symptoms were not found on tomato plants sprayed with newer fungicides.

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