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EFFECTIVITY OF DIFFERENT FUNGICIDES AGAINST FOLIAR LEAF SPOT PATHOGENS OF POPLAR UNDER *IN-VITRO* AND *IN-VIVO* CONDITIONS

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ABSTRACT : The incidence of leaf spots of *Drechslera specifera* and *Curvularia lunata* during April-June was observed to the extent of 72.68 to 88.55 and 11.45 to 27.32 per cent, respectively, whereas, *Alternaria* and *Cercospora* leaf spots were absent. The disease incidence of *Alternaria alternata* was maximum in the month of July-September ranging 19.11 to 29.23 per cent and that of *Cercospora populina* was higher in October-November than July-September being 18.60 to 28.70 per cent. *In vitro* testing of five fungicides namely Dithane M-45, Kavach, Blitox-50, Bayleton and Bavistin against *D. specifera, C. lunata* and *A. alternata* revealed Dithane M-45 to be most effective, whereas, Bayleton and Bavistin were least effective against all these fungi. The *in vivo* testing of fungicides in nursery proved Dithane M-45 at 0.20 per cent concentration best against *D. specifera, C. lunata, A. alternata* and *C. populina* as it totally inhibited the disease under field condition. Next effective fungicides were Kavach and Blitox-50, however, Bayleton and Bavistin were least effective.

Keywords : *Poplar, leaf spot, fungicide, in-vitro in-vivo.*

Poplar (*Populus deltoides* Marsh.) is one of the most important tree species of agroforestry and grown either in blocks or on the boundaries of fields in India. It is also known as American cotton wood is an economically important fast growing tree species, which belongs to family Salicaceae. It is easily growing soft wood tree, raised through stem cuttings and suitable for commercial use like manufacture of matches, furniture, packing cases, plywood, sport goods, pulp and paper, etc. It is most preferred by the farmers because of its fast growing, short rotation and more compatibility with intercrops. Some clones, viz., G-3, G-48 and PL1 to PL7 have proved their adaptability in India. Large number of diseases and insects affect the poplars. Out of these, diseases are one of the major limiting factors in its cultivation (Singh and Singh, 8). It is seriously affected by fungal leaf spots and blights both in nursery and plantations (Anon., 1). This study comprises the incidence of various leaf spots caused by different pathogens in different growing seasons of poplars and their management.

MATERIALS AND METHODS

Collection and isolation of leaf spot pathogens: Isolations of leaf spot fungi were made from diseased leaves of Poplar collected from different agro-climatic zones of Punjab on Potato Dextrose Agar (PDA) medium. Culture of pathogens was purified by single spore isolation technique. Pathogenicity tests were conducted for eaSNch isolated fungus and Koch's postulates were proved (Singh *et al.*, 9).

Incidence of leaf spots of poplar: Incidence of leaf spot pathogens was recorded thrice during main growing seasons of Poplar, i.e. April-June, July-September and October-November from all three agro-climatic zones of Punjab. The following formula was used to calculate per cent disease incidence (Gupta *et al.*, 3 and 4).

$$\text{Per cent disease incidence} = \frac{\text{No. of spots of pathogens}}{\text{Total No. of spots examined}} \times 100$$

***In vitro* evaluation of fungicides:** Poisoned food technique (Nene and Thapliyal, 7) was employed to study the relative efficacy of different fungicides namely, Dithane M-45, Kavach, Blitox-50, Bavistin and Bayleton on mycelial growth of the fungi. Several concentrations (5, 10, 25, 50, 100, 200 µg/ml) of fungicides each on *a.i.* (active ingredient) basis were tested under *in vitro* conditions against *Drechslera specifera* (Bainier) V. Arx., *Curvularia lunata* (Wakker) Boedijen and *Alternaria alternata* (Fr.) Keissl. isolated from leaf spots of Poplar. The weighed quantities of fungicides were dissolved in the measured volumes of sterilized double strength potato dextrose agar (PDA) medium, followed by subsequent pouring into sterilized Petri plates. After solidification of medium, 8 mm disc from 10 days old culture of the test fungus were cut and

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Table 1: Effect of different fungicides on mycelial growth of *Drechslera specifera in vitro* (Average colony diameter (mm) after 6 days of incubation).

Fungicide	CG in control (mm)	Concentration (µg/ml)											
		5		10		25		50		100		200	
		CG (mm)	I (%)	CG (mm)	I (%)	CG (mm)	I (%)	CG (mm)	I (%)	CG (mm)	I (%)	CG (mm)	I (%)
DithaneM-45	90	—	—	81.6	9.3	—	—	64.3	28.5	52.3	42.2	20.3	77.4
Kavach	90	—	—	86.3	4.1	—	—	67.0	25.5	56.0	37.7	41.4	54.1
Blitox-50	90	—	—	87.0	3.33	—	—	71.6	20.4	66.6	26.0	62.3	30.7
Bayleton	90	90	0	89.0	1.11	—	—	73.0	18.8	70.0	22.2	—	—
Bavistin	90	—	—	—	—	90	0	74.0	17.7	70.3	21.8	68.0	24.4
C.D.(P=0.05)				3.07				4.71		4.14		5.15	

Table 2: Effect of different fungicides on mycelial growth of *Curvularia lunata* in *vitro* (Average colony diameter (mm) after 6 days of incubation).

Fungicide	CG in control (mm)	Concentration (µg/ml)											
		5		10		25		50		100		200	
		CG (mm)	I (%)	CG (mm)	I (%)	CG (mm)	I (%)	CG (mm)	I (%)	CG (mm)	I (%)	CG (mm)	I (%)
DithaneM-45	90	—	—	80.0	11.11	—	—	59.6	33.77	49.60	44.88	14.00	84.40
Kavach	90	—	—	83.0	7.77	—	—	67.0	25.50	53.30	40.70	40.00	55.50
Blitox-50	90	—	—	85.0	5.55	—	—	72.0	20.00	62.30	30.70	59.00	34.40
Bayleton	90	90	0	87.0	3.33	—	—	70.0	22.22	65.30	27.40	—	—
Bavistin	90	—	—	—	—	90	0	72.3	19.60	67.60	24.80	64.00	28.80
C.D.(P=0.05)				5.15				5.61		3.41		3.76	

Table 3: Effect of different fungicides on mycelial growth of *Alternaria alternata in vitro* (Average colony diameter (mm) after 5 days of incubation).

Fungicide	CG in control (mm)	Concentration (µg/ml)											
		5		10		25		50		100		200	
		CG (mm)	I (%)	CG (mm)	I (%)	CG (mm)	I (%)	CG (mm)	I (%)	CG (mm)	I (%)	CG (mm)	I (%)
DithaneM-45	90	—	—	79.0	12.2	—	—	62.3	30.4	49.6	44.8	17.3	80.7
Kavach	90	—	—	81.0	10.0	—	—	64.0	28.8	50.3	44.1	38.3	57.4
Blitox-50	90	—	—	84.6	6.0	—	—	70.0	22.22	59.0	34.4	56.0	37.7
Bayleton	90	90	0	87.0	3.33	—	—	69.3	23.0	67.3	25.2	—	—
Bavistin	90	—	—	—	—	90	0	68.0	24.4	67.3	25.7	66.3	26.3
C.D.(P=0.05)				2.87				4.19		4.32		3.76	

CG = Mean colony growth of three replications in mm, I (%) = Inhibition in per cent, - = Not tested

placed in inverted position for the direct contact with the when Petri plates kept as control were completely filled (Gupta and Agarwal, 2). The per cent inhibition of growth in each treatment was calculated by the formula devised by Vincent (10).

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

Pii = per cent inhibition of vegetative growth

C = colony diameter in control

T = colony diameter in treatment

Testing of fungicides against leaf spots in nursery: The fungicides found effective during laboratory screening test, were also tested in nursery under field conditions. The leaves of young nursery plants were inoculated with spore suspension of isolated pathogen and sprayed with the fungicides suspension after 24 hours of inoculation. Each treatment was replicated three times. Data on disease intensity were recorded after 10 days. The disease intensity was rated on the basis of leaf area covered by the infection spot for each leaf as given below:

Disease rating scale: The rating scale was used as 0 - leaves free from infection, 1-5 per cent leaf area affected, 6-20 per cent leaf area affected, 21-40 per cent leaf area affected, 41-70 per cent leaf area affected, 71-100 per cent leaf area affected.

The disease intensity in each plot was calculated by the formula as employed by McKinney (6).

$$\text{Per cent disease index} = \frac{\text{Sum of all numerical rating}}{\text{No. of leaves observed} \times \text{Maximum rating}} \times 100$$

The following formula was used to calculate per cent disease inhibition.

$$\text{Per cent disease inhibition} = \frac{\text{Disease index in control} - \text{Disease index in treatment}}{\text{Disease index in control}} \times 100$$

RESULTS AND DISCUSSION

All of the infected tissues of the diseased leaves yielded different pathogens, viz. *Cercospora populina, Curvularia lunata, Drechslera specifera* and *Alternaria alternata.* In present investigation the incidence of various leaf spots have been observed, as the highest incidence of *Drechslera specifera* and *Curvularia lunata* was during April-June, 1997 varied from 72.68 to 88.56 per cent and 11.45 to 27.32 per cent, respectively, whereas other two pathogens were absent during this period. The incidence of *Drechslera specifera* leaf spots during July-September 1997 varied from 31.00 to 50.58 per cent and that of *Curvularia lunata* from 10.22 to 23.43 per cent, *Alternaria alternata* from 19.11 to 29.23 per cent and *Cercospora populina* from 12.11 to 25.70 per cent. A perusal of October-November, 1997 reveals that the disease incidence of *Drechslera specifera* and *Alternaria alternata* and *C. lunata* was lowest, i.e. 27.87 to 48.42 per cent and 12.00 to 28.20 per cent 12.8 to 22.90 per cent, respectively. Similarly, *Cercospora populina,*which was absent an April-June, picked up during July-September but was highest during the last growing period i.e. October-November and ranged from 18.60 to 28.70 per cent. The results are in conformity with various findings of different workers as they also reported the prevalence and incidences of these pathogens on poplar clones (Gupta *et al.,* 3, and Singh, *et al.*, 9).

***In-vitro* evaluation of fungicides:** To determine the comparative efficacy and optimum effective concentration of different selected fungicides namely, Dithane M-45, Kavach, Blitox-50, Bavistin and Bayleton against Poplar leaf spot pathogens, viz. *Alternaria alternata, Curvularia lunata* and *Drechslera specifera*, an experiment was conducted by employing poisoned food technique. The data in term of per cent inhibition of radial growth were recorded after 6 days in case of *Drechslera specifera* and *Curvularia lunata* and after 5 days in case of *Alternaria alternate* (Table 1 to 3).

***Drechslera specifera*:** Results presented in Table 1 revealed that among all the fungicides tested against *Drechslera specifera,* Dithane M-45 proved to be most effective. At 200 µg/ml concentration maximum growth inhibition was obtained with Dithane M-45 followed by Kavach, Blitox-50 and Bayleton. Similarly, at 50 and at 100 µg/ml concentrations Dithane M-45 was best; however, Kavach did not differ significantly with Dithane M-45 at both these concentrations. Bavistin proved to be least effective at all the concentrations tested. At 200 µg/ml concentration Dithane M-45 gave maximum colony growth inhibition of 77.4 per cent, where as Bavistin was least effective with 22.4 per cent inhibition only.

***Curvularia lunata*:** It is clear from the results (Table 2) that Dithane M-45 was significantly superior to other fungicides at 50, 100 and 200 µg/ml concentrations followed by Kavach. At 10 µg/ml concentration 7 Kavach was at par with Dithane M-45.

Table 4: Effect of different fungicides on development of *Drechslera specifera* leaf spots on poplar under field condition.

Fungicide	Concentration (%)	Per cent disease index	Per cent inhibition
Dithane M-45	0.10	18.66 (25.54)	39.13
	0.15	9.33 (17.66)	69.56
	0.20	0.00 (0.00)	100.00
Kavach	0.10	18.66 (25.54)	39.13
	0.15	10.66 (18.95)	65.23
	0.20	2.66 (9.17)	91.32
Blitox - 50	0.10	23.99 (29.24)	21.72
	0.15	15.99 (23.51)	47.81
	0.20	6.66 (14.75)	78.27
Bayleton	0.02	29.33 (32.74)	4.33
	0.05	26.66 (31.01)	13.04
	0.10	23.99 (29.24)	21.72
Bavistin	0.02	27.99 (31.91)	8.67
	0.05	26.66 (31.01)	13.04
	0.10	23.99 (29.24)	21.22
Control	—	30.66 (33.59)	
C.D.(P= 0.05)	—	4.01	—

Table 5: Effect of different fungicides on development of *Curvularia lunata* leaf spots on poplar under field condition.

Fungicide	Concentration (%)	Per cent disease index	Per cent inhibition
Dithane M-45	0.10	6.66 (14.82)	58.37
	0.15	5.33 (13.04)	66.68
	0.20	0.00 (0.00)	100.00
Kavach	0.10	9.33 (17.66)	41.68
	0.15	6.66 (14.82)	58.37
	0.20	0.00(0.00)	100.00
Blitox - 50	0.10	10.66 (18.95)	33.37
	0.15	9.33 (17.66)	41.68
	0.20	5.3 (13.04)	66.68
Bayleton	0.02	14.66 (22.41)	8.37
	0.05	11.99 (20.18)	25.00
	0.10	10.66 (18.95)	33.37
Bavistin	0.02	14.66 (22.41)	8.37
	0.05	11.99 (20.18)	25.00
	0.10	10.66 (18.95)	33.37
Control	—	15.99 (23.51)	
C.D. (P = 0.05)	—	4.28	—

Observations are based on three replications

Data in parentheses are arc sine transformed values.

At 200 µg/ml concentration Dithane M-45 inhibited 84.4 per cent colony growth, whereas Bavistin at this concentration inhibited only 28.8 per cent colony growth of pathogen. Bayleton at 5 µg/ml and Bavistin at 25 µg/ml showed no inhibition of colony growth.

***Alternaria alternata*:** It is clear from the results (Table 3) that all the fungicides at all the concentrations tested, except Bayleton at 5 µg/ml and Bavistin at 25 µg/ml, were significantly superior over control. At 200 µg/ml Dithane M-45 proved to be most effective followed by Kavach, whereas Bavistin was least effective. At 10, 50 and 100 µg/ml Dithane M-45 and Kavach were significantly at par in inhibiting colony growth of the fungus. Blitox-50 and Bayleton at 10 µg/ml concentration, Blitox-50, Bayleton and Bavistin at 50 µg/ml and Bayleton and Bavistin at 100 µg/ml were statistically at par and least effective.

Evaluation of fungicides against leaf spot pathogens of poplar *in-vivo*

***Drechslera specifera*:** The data presented in Table 4 showed that Dithane M-45 gave good disease inhibition at all concentration tested with 100 per cent inhibition at 0.20 per cent and was closely followed by Kavach and then Blitox-50 in inhibition of disease. The performance of Bayleton and Bavistin at all concentrations tested was much poor as compared to these non-systemic fungicides, although disease control in these two was also significant as compared to control. By judging the relative efficacy of fungicides, Dithane M-45 and Kavach were statistically at par in controlling the disease. Disease was totally inhibited with Dithane M-45 at 0.20 per cent concentration. The third best fungicide in order of efficacy was Blitox-50.

***Curvularia lunata*:** The data given in Table 5 showed that Dithane M-45 and Kavach were most effective fungicides with 100 per cent control at 0.20 per cent concentration, Blitox-50 comes second with 66.68 per cent disease inhibition at the concentration of 0.20 per cent. Bayleton and Bavistin (systemic) fungicides were much poorer in their performance as compared to above mentioned fungicides but were significant in inhibition at 0.10 per cent concentrations as compared to control and these also controlled the disease to some extent.

***Alternaria alternata*:** The data given in Table 6 indicated that Dithane M-45, Kavach and Blitox-50 at all concentrations tested and Bayleton at 0.05 and Bavistin at 0.10 per cent concentration were statistically superior over control. At 0.20 per cent concentration both Dithane M-45 and Kavach gave 100 per cent disease inhibition followed by Blitox-50 which gave 73.70 per cent disease control at 0.20 per cent concentration. However, the disease inhibition in Bayleton and Bavistin was only up to 42.12 per cent at highest concentration of 0.20 per cent tested for these fungicides.

***Cercospora populina*:** It is clear from the data (Table 7) that all the three fungicides tested, i.e. Dithane

Table 6: Effect of different fungicides on development of *Alternaria alternata* leaf spots on poplar under field condition.

Fungicide	Concentration (%)	Per cent disease index	Per cent inhibition
Dithane M-45	0.10	13.33 (21.34)	47.37
	0.15	7.99 (16.27)	68.41
	0.20	0.00 (0.00)	100.00
Kavach	0.10	15.99 (22.51)	36.83
	0.15	10.66 (18.95)	57.91
	0.20	0.00(0.00)	100.00
Blitox - 50	0.10	17.33 (24.49)	31.58
	0.15	11.99 (20.18)	52.61
	0.20	6.66 (14.82)	73.70
Bayleton	0.02	22.66 (28.33)	10.54
	0.05	18.66 (25.54)	26.33
	0.10	14.66 (22.41)	42.12
Bavistin	0.02	22.66 (28.33)	10.54
	0.05	17.33 (24.49)	31.58
	0.10	14.66 (22.41)	42.12
Control	—	25.55 (30.30)	
C.D. (P = 0.05)	—	4.46	—

Table 7: Effect of different fungicides on development of *Cercospora populina* leaf spots on poplar under field condition.

Fungicide	Concentration (%)	Per cent disease index	Per cent inhibition
Dithane M-45	0.10	21.33 (27.40)	42.86
	0.15	11.99 (20.18)	67.85
	0.20	0.00 (0.00)	100.00
Kavach	0.10	23.99 (29.24)	35.70
	0.15	21.33 (27.40)	42.86
	0.20	2.66 (9.17)	92.87
Blitox - 50	0.10	30.66 (33.59)	17.86
	0.15	19.99 (26.52)	46.42
	0.20	7.99 (16.27)	78.56
Control	—	37.36 (37.64)	
C.D. (P = 0.05)	—	3.60	

*Observations are based on three replications

**Data in parentheses are arc sine transformed values.*

M-45, Kavach and Blitox-50 were significantly superior over control at all the concentrations tested. At 0.15 and 0.20 per cent concentration Dithane M-45 was most effective followed by Kavach and Blitox. At 0.20 per cent concentration per cent disease inhibition with Dithane M-45 was hundred per cent, whereas it was 92.87 per cent with Kavach and 78.56 per cent with Blitox-50.

Likewise, Khara and Sahni (5) found that blitox-50, captan and difolitan were effective against *C. lunata* under laboratory conditions. Gupta *et al.*, (3) reported that Dithane M-45 was effective fungicide followed by Dithane M-45 and Blitox-50 against leaf spot pathogen of poplar.

REFERENCES

1. Anonymous (1996). *Annual Report*. Department of Forestry and Natural Resources, P.A.U., Ludhiana pp. 149.
2. Gupta, A. K., and Agarwal, V. K. (2003). *Phytophthora cactorum*: A new test fungus for estimation and detection of tebuconazole from treated seeds of wheat. *Ann. Pl. Protec. Sci.,* **11**(1): 170-172.
3. Gupta, A. K., Kushwaha, K.P.S. and Singh, S. (2001a). Evaluation of some fungicides against foliage pathogen of Poplar. *Ann. Pl. Protec. Sci.,* **9**(1): 150-153.
4. Gupta, A. K., Khara, H. S. and Singh, S. (2001b). Incidence of leaf spots of poplar in three agro-climatic zones of Punjab. *Pl. Dis. Res.,* **16** (1): 134-136.
5. Khara, H.S. and Sahni, S.K. (1984). Curvularia leaf spot disease of Palmarosa in the Punjab. *J Mycol.Pl. Path.* **14**: 131-133.
6. McKinney, H.N. (1923). Influence of soil temperature and moisture on infection of wheat seedlings by Helminthosporium sativum. J. Agric. Res., **26**: 195-217.
7. Nene, Y.L. and Thapliyal, P.N. (1979). Fungicide in Plant Disease Control. 2nd edition, Oxford and IBH Publ. Co., New Delhi, India, pp. 507.
8. Singh, P. and Singh, S. (1986). *Insect Pests and Diseases of Poplar*. Publ. Forest Research Institute Press, Dehradun, pp. 74.
9. Singh, S.; Pandey, P.C.; Singh, A.; Bahadur, R. and Mukherjee, S.N. (1991). Studies on foliage diseases of Poplars. *Indian For.* **117**: 135-42.
10. Vincent, J.H. (1947). Distortation of fungal hyphae in the presence of certain inhibitiors. *Nature (London). 159: 850.*

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