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Evaluation of In Vitro Efficacy of Commercial Fungicides Against Alternaria Alternata Isolates Causing Leaf Spot on Populus Deltoides

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ABSTRACT

Poplar is the one of the domesticated forest trees in India and has better synergy with agriculture system than forestry operations. Populus deltoides have shown great promise in north-western part of India. Many a times, single clone of poplars have been propagated extensively. Ninety per cent of the total planted poplar comprises of clones G-48, WSL-22, WSL-39, Udai, WSL-32, Wimco81 and S7C15 in the states of Punjab, Haryana, Uttarakhand and Uttar Pradesh. High incidence of Alternaria leaf spot was noticed on different commercial clones of P. deltoides (G-48, Udai, WSL-22 and WSL-39) during surveys in poplar nurseries (2009-11). The most common method of managing plant diseases is use of fungicides. In vitro efficacy of two fungicides, namely, propiconazole (systemic) and chlorothalonil (non-systemic) was tested against Alternaria alternata isolates using poisoned food technique. It was observed that propiconazole was more effective in suppressing the growth of the fungus than chlorothalonil as cent per cent inhibition was achieved for all the isolates at 40ppm. On the other hand, cent per cent inhibition of growth was achieved for only four isolates, no. A15, A24, A41 and A47 at highest concentration of 400ppm of chlorothalonil. The study indicated that propiconazole proved to be effective at a very low concentration of 40ppm against A. alternata under in vitro conditions.

Keywords: Alternaria Alternata, Clones Disease, Fungicides, Inhibition

I. INTRODUCTION

Populus deltoides, commonly known as cottonwood, is the only species of polar that is planted on a significant scale in India. Its share of about 312,000 ha outside the forest constitutes the backbone of agro-forestry in irrigated plains of northern India having the share of

(ICFRE, 2011). It belongs to genus Populus and family Salicaceae. Around two dozen clones are commercially grown in India out of which, clone G48 (36.2%), WSL22(18.8%), Udai (11.9%), WSL39 (9.1%), WSL32(2.9%), WIMCO81 (5.3 %) and S7C15 (5.8 %) together constitute over 90 percent of the total planted poplar in Punjab, Haryana, U.K. and U.P. states of the country (Dhiman and Gandhi, 2012). Of late, many clones (G-48, Udai, WSL- 22, WSL- 39, etc.) have been found to be infected with a number of pathogens like Alternaria, Bipolaris, Curvularia, Sclerotium, etc. Infection of Alternaria spp. was epidemic in nurseries of poplar in Uttarakhand and U.P. states. The disease symptoms are characterized by irregular, necrotic greyish brown lesions surrounded by a chlorotic halo on leaves with a characteristic bull's eye. Many species of Alternaria are saprophytes or a weak pathogen especially of damaged plants that are commonly found in soil or on decaying plant (Holliday, 1989). Aletrnaria alternata cause severe diseases in different ornamental crops, including trees and shrubs (Chase, 1998; Masangkay et al., 1999; Jones, 2001). A great number of species were recorded for the genus Alternaria infecting different crops causing world-wide economic loss (Kirk, 2008).

II. METHODS AND MATERIAL

The surveys were conducted at R&D Center of Wimco Seedlings, Bagwala (Rudrapur) in Udham Singh Nagar (located at latitude: 29° 30' N and longitude: 79° 28' E), Maheshwari and Paniyala (Roorkee) in Hardwar (29° 52' N and 77° 53' E), Thana Chappar (Yamuna Nagar) Haryana (located at 30°7'N and 77°18'E.). The different commercial clones of *P. deltoides* viz. G-48, Udai, WSL-22 and WSL-39 were screened for *A. alternata* infection on leaves.

Isolation of the disease causing fungi was made from collected leaf samples and subsequently pathogenic cultures were purified. The isolates of *A. alternata* were tested against two fungicides, Chlorothalonil or Kavach (non-systemic) and Propiconazole or Tagzol (systemic) using poisoned food technique (Nene and Thapliyal, 1979). The experiment was conducted using 4 concentrations of Chlorothalonil, i.e., 100, 200, 300 and 400 ppm and 10, 20, 30 and 40ppm of Propiconazole. The required quantity of the fungicides were weighed and added into pre- sterilized PDA followed by pouring into the sterilized Petriplates. Control plates were also maintained without adding any fungicides into them. Three replicates were maintained for each isolate tested. Mycelial disc of about 1mm was cut from the actively growing culture plate with the aid of sterilized borer and subsequently kept in the middle of the each poisoned agar plate. Then each plate was kept in BOD incubator at 30° C. Radial growth was recorded for each isolate when control plates were completely filled. Radial growth inhibition percentage was calculated by using the formula given by Vincet (1947):

$$\text{Percentage inhibition} = (\text{Control} - \text{treatment} / \text{Control}) \times 100$$

Data for different parameters were analyzed with the help of GENSTAT 5 Release 3.22. Two-way analysis was used for fungicidal sensitivity data. Treatments means were compared at 5 per cent level of significance.

III.RESULTS AND DISCUSSION

Propiconazole, irrespective of concentrations, inhibited maximum and significantly higher growth of isolate no. A64 (97.7%; Table1 & Fig.1). Minimum and significantly low suppression of fungal growth was observed for isolate no. A40 (87.3%). There was significant reduction of growth of *A. alternata* isolates as the concentration of fungicide increased from 10 to 40ppm ignoring isolates.

When the interactions between isolate and concentration (I x C) were studied, it was observed that cent per cent inhibition was achieved for all the

isolates at 40ppm. Barring two isolates, no. A12 (90.2%) and A40 (91.7%) remaining isolates had complete growth inhibition at 30ppm. Growth suppression of five isolates, no. A13, A32, A51, A64 and A65 were cent per cent from 20ppm to the highest concentration tested. On the contrary, minimum and significantly less growth inhibition was observed for isolate no. A40 (73.6%) at the lowest concentration of 10ppm. Irrespective of Chlorothalonil concentrations, maximum and significantly high growth inhibition was recorded for isolate no. A15 (68.1%) which was at par with isolate no. A41(67.9%; Table2;Fig2.). Significantly low suppression of growth was quantified for isolate no. A65(39.3%). Irrespective of isolates, there was a linear trend of growth inhibition over concentrations of the fungicide. For example, minimum growth inhibition of 35.1 per cent was recorded at 100ppm and highest of 82.1 per cent at 400ppm. Cent percent inhibition of growth was achieved by three isolates A15, A24, A41 and A47 at highest concentration of 400ppm, when interactions between isolate and concentration (I x C) were studied. On the other extreme, minimum and significantly less growth suppression was observed for isolate no. A65 at 100ppm (14.3%). All the isolates had significant suppression of growth over the fungicidal concentrations used.

Table 1. Effect of different concentrations of Propiconazole on the growth of *A.alternata isolates*

Isolate no.	**Fungicidal concentration (ppm)/Inhibition (%)**					**Mean**
	Control	**10**	**20**	**30**	**40**	
A7	0.0	77.9	85.5	100.0	100.0	90.8
A12	0.0	79.5	83.1	90.2	100.0	88.2
A13	0.0	90.0	100.0	100.0	100.0	97.5
A15	0.0	83.3	90.9	100.0	100.0	93.6
A16	0.0	74.8	85.5	100.0	100.0	90.1
A24	0.0	84.5	89.3	100.0	100.0	93.5
A25	0.0	80.0	90.2	100.0	100.0	92.6
A32	0.0	87.9	100.0	100.0	100.0	97.0
A40	0.0	73.6	84.1	91.7	100.0	87.3
A41	0.0	87.6	92.1	100.0	100.0	95.0
A47	0.0	88.8	91.2	100.0	100.0	95.0
A51	0.0	88.3	100.0	100.0	100.0	97.1
A52	0.0	90.2	91.4	100.0	100.0	95.4
A64	0.0	90.7	100.0	100.0	100.0	97.7
A65	0.0	89.8	100.0	100.0	100.0	97.4
Mean	0.0	84.5	92.2	98.8	100.0	
	Isolate		**Concentration**		**Interaction (I x C)**	
SEM	0.2		0.1		0.3	
CD (5%)	0.4		0.2		0.9	

Table 2. Effect of different concentrations of Chlorothalonil on the growth of *A. alternata* isolates

Isolate no.	Fungicidal concentration (ppm)/Inhibition (%)					Mean
	Control	100	200	300	400	
A7	0.0	46.2	50.5	57.6	65.5	54.9
A12	0.0	35.7	39.8	57.1	64.5	49.3
A13	0.0	32.6	43.1	60.5	66.7	50.7
A15	0.0	38.3	57.1	76.9	100.0	68.1
A16	0.0	50.0	61.2	64.3	70.7	61.5
A24	0.0	38.8	50.5	76.9	100.0	66.5
A25	0.0	28.6	36.4	46.7	74.5	46.5
A32	0.0	36.0	50.0	78.3	90.0	63.6
A40	0.0	27.6	49.8	78.1	88.6	61.0
A41	0.0	43.0	57.1	71.4	100.0	67.9
A47	0.0	20.7	65.5	75.2	100.0	65.4
A51	0.0	17.8	31.9	82.4	90.0	55.5
A52	0.0	50.0	57.1	68.6	75.0	62.7
A64	0.0	47.4	57.1	77.6	81.4	65.9
A65	0.0	14.3	28.6	50.0	64.3	39.3
Mean	0.0	35.1	49.0	68.1	82.1	
	Isolate		**Concentration**		**Interaction (I x C)**	
SEM	0.1		0.1		0.3	
CD (5%)	0.4		0.2		0.8	

Diseases are one of the major limiting factors in cultivation of poplars (Singh and Singh, 1986). Use of single genotype (clone) over a large area entails an enormous risk. Monocultures are widely believed to attract diseases and insects. The most common method of managing plant diseases is use of chemicals that are toxic to the pathogens. Such chemicals are fungistasis or fungicidal in their mode of action. *In vitro* efficacy of two fungicides namely Propiconazole (systemic) and Chlorothalonil (non-systemic) was tested against *A. alternata* isolates. It was observed that Propiconazole was more effective in suppressing the growth of the fungus than Chlorothalonil as cent percent inhibition was achieved for all the isolates at 40ppm. The results are in agreement with Anon. (2002), Verma and Verma (2010), Mesta *et al.* (2011), Sharma *et al.* (2013). Growth suppression of five isolates, no. A13, A32, A51, A64 and A65 were 100 percent from 20ppm. On the other hand, cent percent inhibition of growth was achieved by only three isolates A15, A24, A41 and A47 at highest concentration of 400ppm of chlorothalonil.

Similar observations were made by Gorawar *et al.* (2006). Among systemic fungicides, penconazole, propiconazole and hexaconazole showed 100 per cent inhibition of the *A. alternata* isolates at all three concentrations (0.1, 0.2 and 0.3%) tested while, the non-systemic fungicide mancozeb at all three concentrations and zineb at 0.3 per cent were completely inhibiting the growth of *A. alternata*. *In vitro* evaluation of fungicides by Gupta *et al.* (2014) revealed that mancozeb proved to be more effective than chlorothalonil to control the fungal growth at 0.10, 0.15 & 0.20 per cent. Chethana *et al* . (2012) reported that Chlorothalonil was less effective against *A. porri* by recording mean inhibition of 17.27 per cent for mycelia growth. Similar results were obtained by Sharma *et al.* (2013), Wagh (2015) and Hegde *et al.* (2015) against different Alternaria species tested. However, observations made by Dillard and Cobb (2008) suggested that chlorothalonil proved to be better fungicides besides other against *A. alternata*.

IV.CONCLUSION

The present investigation indicated that propiconazole proved to be effective even at very low concentration of 0.004 per cent against *A. alternata* under *in vitro* conditions. Thus, it can be further tested in field condition for management of the disease.

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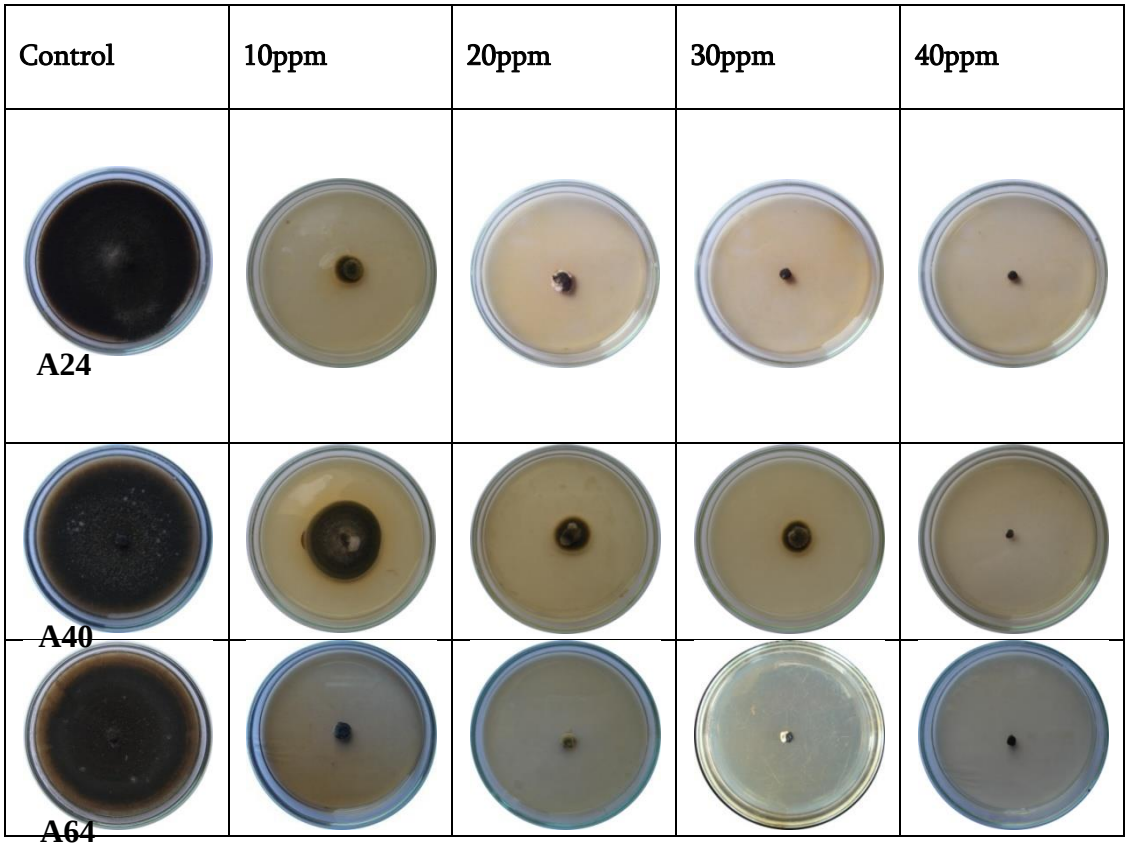

Fig1. Effect of propiconazole on *A. alternata* isolates

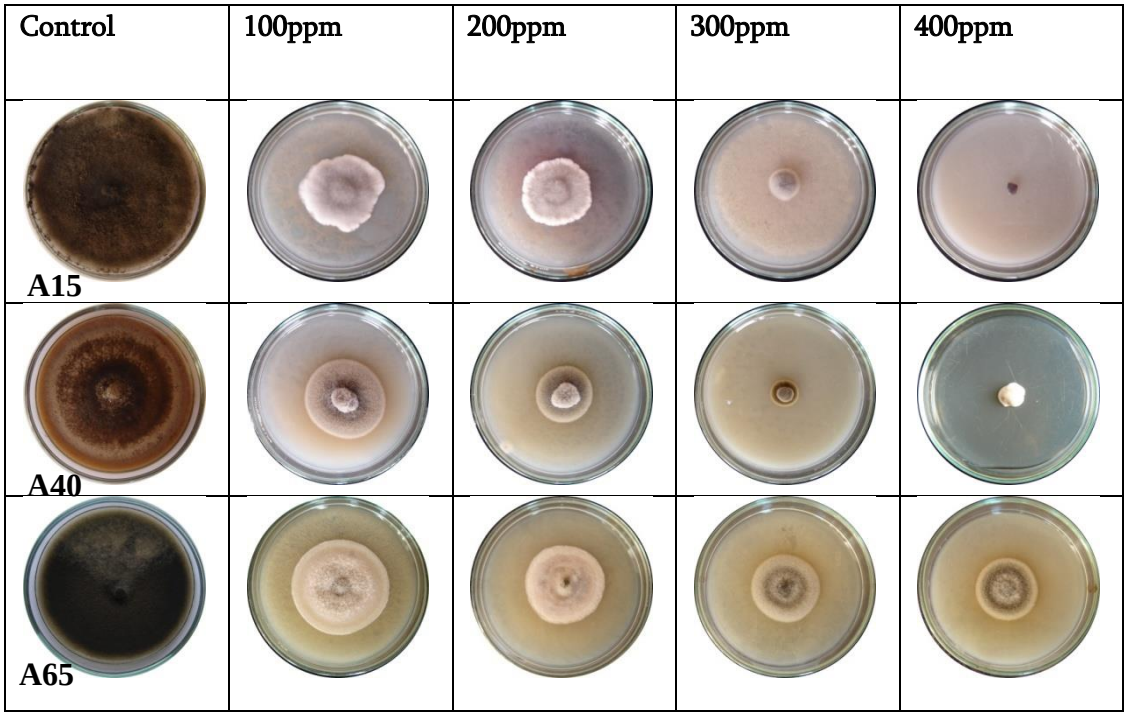

Fig2. Effect of chlorothalonil on *A. alternata* isolates