

Efficacy of chemical fungicides and bio-agents against major cotton fungal foliar diseases *in vitro*

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SUMMARY

An investigation studies on major fungal foliar diseases of cotton was carried out *in vitro* in the Department of Plant Pathology, MPKV, Rahuri since isolation of leaf spot pathogens, pathogenicity to efficacy of chemical fungicides and bioagent in *in-vitro*. The isolated pathogens associated with fungal foliar diseases were *Alternaria macrospora*, *Myrothecium roridum* and *Helminthosporium spiciferum*. The pathogenicity of isolated pathogens was proved on susceptible cotton var. LRA-5166. These pathogens produced symptoms within 8-13 days. The pathogenicity test proved the pathogenic nature of isolated pathogens. Efficacy of six fungicides and two bioagents was tested in *in-vitro*. Mancozeb (0.3%), propiconazole (0.1%), propineb (0.3%) were found more effective against *Alternaria* leaf blight, propiconazole (0.1%) and copper oxychloride (0.25%) against *Myrothecium* leaf spot and mancozeb (0.3%) and propiconazole (0.1%) were more effective against *Helminthosporium* leaf spot in full and ½ concentration. *Aspergillus niger* was found more effective than *Trichoderma viride* in *in-vitro*.

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Key words : Chemical fungicides, Bio-agents, Cotton, Fungal diseases

Cotton is the most important commercial crop which plays a vital role in the national economy. The area under Maharashtra state during 2007-08 was 31.91 lakh ha with the production of 60.00 l bales and productivity of 320 kg lint / ha (Anonymous, 2008). Cotton plant subjects to infection by various fungi, bacteria and viruses which leads to reduction in gross yield and deterioration in quality causing depreciation of market value. Amongst the diseases *Alternaria* leaf blight, *Myrothecium* leaf spot and *Helminthosporium* leaf spot poses an alarming situation in Maharashtra but very less information is available on these aspects. Hence, systemic studies on isolation, pathogenicity of isolated organisms and testing the efficacy of chemical / bioagents in *in vitro* condition was carried out.

MATERIALS AND METHODS

Fresh specimen of cotton leaf spot were collected from the experimental field of All India Coordinated Cotton Improvement Project, Mahatma Phule Krishi Vidyapeeth,

Rahuri. Six chemical fungicides namely chlorothalonil, carbendazim, copper oxychloride, mancozeb, propineb and propiconazole and two bioagents *Trichoderma viride* and *Aspergillus niger* in powder form were evaluated against the pathogens.

The common laboratory medium potato dextrose agar was used for the isolation of fungus associated with leaf blight and leaf spot of cotton. Potato dextrose broth (PDB) was used for growing pathogens to study pathogenicity. The well developed fungal growth free from any contamination was transferred to agar slants by hyphal tip method to obtain pure culture. Thirty sterilized earthen pots were filled with sterilized soil. Three seeds of susceptible var. LRA-5166 were sown in each pot in glasshouse under controlled conditions. Forty five days old plants were used to test the pathogenicity. The control plants were sprayed with sterilized water only. Koch's postulates were proved and three isolates were identified as *Alternaria macrospora*, *Myrothecium roridum* and *Helminthosporium spiciferum*.

Poisoned food technique was followed to evaluate the fungicide against pathogens responsible for leaf spots of cotton in *in vitro*. Observations on growth of colony diameter were recorded on 8th day after inoculation. Radial growth was measured and results were expressed as per cent inhibition of mycelial growth over control by using the following formula (Padule and Shinde, 1989).

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

where,

I = per cent inhibition of fungal growth.

C = Growth in mm on 8th day after inoculation in control

T = Growth in mm on the 8th day after inoculation

The bioagents *viz.*, *Trichoderma viride* and *Aspergillus niger* were tested for their antagonistic properties against leaf spots causing pathogens by direct bit placement method. After seven days of incubation the growth of the fungus was measured in each plates and the per cent growth inhibition of intersecting colonies was calculated by the formula outlined by Arora and Upadhyay (1978).

$$\text{Growth inhibition (\%)} = \frac{\text{Colony growth in control plate} - \text{Colony growth in interacting plates}}{\text{Colony growth in control plate}} \times 100$$

RESULTS AND DISCUSSION

The results obtained from the present investigation have been discussed in the following sub heads :

Isolation, pathogenicity and reisolation:

Isolation :

The microscopic examination of the diseased cotton leaves collected from cotton field of AICCIP, MPKV, Rahuri, revealed the presence of spores of *Alternaria*, *Myrothecium* and *Helminthosporium* spp. The pathogens responsible for the leaf spot and blight were

isolated on PDA in the laboratory. On the basis of morphological characters they were taxonomically identified as *Alternaria macrospora*, *Myrothecium roridum* and *Helminthosporium spiciferum*.

The pathogens responsible for blight and leaf spot were also isolated and reported by Kurundkar and Mayee (1986).

Pathogenicity :

The pure cultures of *Alternaria macrospora* Zimm., *Myrothecium roridum* Tode Fr. and *Helminthosporium spiciferum* (Bain) Nicot were inoculated separately by leaf injury method on the healthy seedling of cotton var. LRA-5166 under controlled conditions.

The leaf spots and leaf blights symptoms developed within 8 to 13 days on susceptible cotton var. LRA-5166, were identical with symptoms of disease as previously observed in the field. Sharma and Chauhan (1987) isolated *Myrothecium* proved its pathogenicity and Lambhate *et al.* (2002) isolated and proved the pathogenicity of *Alternaria macrospora*.

Reisolation :

The reisolations were made from the leaf spot developed on inoculated plants and isolates were compared with original cultures and identified as *A. macrospora* Zimm., *Myrothecium roridum* Tode Fr. and *Helminthosporium spiciferum* (Bain) Nicot. The similar type of work was also performed by Sharma and Chauhan (1987) and Lambhate *et al.* (2002).

Chemical and biological control in *in vitro*:

Out of six chemicals mancozeb, propineb and

Table 1 : Effect of fungicides and bioagents on the growth of pathogens in *in-vitro* at full concentration

Sr. No.	Treatments	Concentration (%)	<i>Alternaria macrospora</i>		<i>Myrothecium roridum</i>		<i>Helminthosporium spiciferum</i>	
			Mean colony diameter (mm)	Inhibition of growth (%)	Mean colony diameter (mm)	Inhibition of growth (%)	Mean colony diameter (mm)	Inhibition of growth (%)
1.	Chlorothalonil	0.2	23.16*	74.26	29.00	63.75	38.00	57.41
2.	Carbendazim	0.1	66.83	25.74	56.83	28.96	65.00	27.77
3.	Copper oxychloride	0.25	23.56	73.88	0.000	100	24.83	72.41
4.	Mancozeb	0.3	0.000	100	22.33	72.08	0.000	100
5.	Propineb	0.3	0.000	100	24.16	69.80	21.83	75.74
6.	Propiconazole	0.1	0.000	100	0.000	100	15.00	83.33
7.	<i>Trichoderma viride</i>	5mm bit	29.33	67.41	20.00	75.00	34.33	61.85
8.	<i>Aspergillus niger</i>	5mm bit	22.16	75.37	19.00	76.25	30.5	66.11
9.	Control	--	90	--	80.00	--	90	--
	S.E. ±		0.646		0.430		0.491	
	C.D. (P=0.05)		1.380		1.27		1.45	

* Average of four replications

Table 2 : Effect of fungicides and bioagents on the growth of major fungal foliar disease of cotton in *in-vitro* at ½ concentration

Sr. No.	Treatments	Concentration (%)	<i>Alternaria macrospora</i>		<i>Myrothecium roridum</i>		<i>Helminthosporium spiciferum</i>	
			Mean colony diameter (mm)	Inhibition of growth (%)	Mean colony diameter (mm)	Inhibition of growth (%)	Mean colony diameter (mm)	Inhibition of growth (%)
1.	Chlorothalonil	0.1	25.66	71.48	38.00	62.50	39.66	55.93
2.	Carbendazim	0.05	68.50	23.88	61.75	22.81	68.00	24.44
3.	Copper oxychloride	0.125	27.66	69.26	13.33	83.33	26.41	70.65
4.	Mancozeb	0.150	0.000	100	26.50	66.87	19.33	78.52
5.	Propineb	0.150	0.000	100	32.41	59.48	24.5	72.77
6.	Propiconazole	0.05	0.000	100	0.00	100	21.00	76.66
7.	<i>Trichoderma viride</i>	5mm bit	32.33	64.06	22.41	71.98	36.66	59.27
8.	<i>Aspergillus niger</i>	5mm bit	23.66	73.71	20.83	73.96	32.33	64.08
9.	Control	--	90	--	80	--	90	--
	S.E. ±		0.305		0.364		0.461	
	C.D. (P=0.05)		0.915		1.08		1.382	

* Average of four replications

propiconazole were found significantly superior to rest of the fungicides. The bioagents *Aspergillus niger* and *Trichoderma viride* were also found effective in arresting the growth of *Alternaria macrospora* (Table 1 and 2).

The growth of *Myrothecium roridum* was completely inhibited by the fungicidal treatments of propiconazole and copper oxychloride. The next best fungicide was mancozeb. The bioagents *A. niger* and *T. viride* were also found effective in restricting the growth of *Myrothecium roridum* by 76.25 per cent and 75.00 per cent, respectively.

Mancozeb and propiconazole were found the most superior in checking the growth of *H. spiciferum*. The bioagents were less effective against *H. spiciferum* as compared with *A. macrospora* and *M. roridum*.

From these results it is seen that propiconazole (0.1%) was the best fungicide for control of all the three pathogens causing leaf spot diseases of cotton. This was followed by mancozeb (0.3%) as it has completely inhibited the growth of *Alternaria macrospora* and *Helminthosporium spiciferum* and also 72.08 per cent

inhibition of growth of *Myrothecium roridum*.

The bioagent *Aspergillus niger* was found more effective than *Trichoderma viride* in restricting the growth of all the three pathogens. These two bioagents were more effective against *A. macrospora* and *M. roridum* as compared to *H. spiciferum*.

These results were more or less similar to the results reported by Jain and Gupta (1973), Lokesh and Hiremath (1988), Padule and Shinde (1989), Ghosh *et al.* (2002) and Silva *et al.* (2006).

Conclusion:

The chemical fungicides mancozeb (0.3%), propiconazole (0.1%), propineb (0.3%) were found more effective against *Alternaria macrospora*, propiconazole (0.1%) and copper oxychloride (0.25%) against *Myrothecium roridum* and mancozeb (0.3%) and propinconazole (0.1%) were found effective against *Helminthosporium spiciferum* whereas in bioagents *Aspergillus niger* was more effective than *Trichoderma viride* in *in vitro* control of all the three pathogens isolated.

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