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In vitro Control of *Alternaria solani*, the Cause of Early Blight of Tomato

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Abstract: *In vitro* control of *Alternaria solani* was studied with different fungicides. Using Poison Food Technique (PFT), seven fungicides, Antracol, Benlate, Copper oxychloride, Dithane M-45, Ridomil, Topas, and Topsin were evaluated at four different concentrations (250, 500, 750, and 1000 ppm) to control colony growth of fungi . The lowest colony growth was recorded in Dithane M-45 treatment at 1000 ppm, and the highest in treatment where no fungicide was used, indicating the significance of using fungicides in controlling early blight disease in tomato.

Key words: Tomato, early blight, *Alternaria solani*, fungicides, *in vitro* control, Pakistan

Introduction

In Pakistan per hectare yield of tomato (10.84 t) is very low due to several production constraints including diseases. One such disease is the early blight caused by the fungus *Alternaria solani*. (Ell), Martin. Common names used for the disease at various stages of plant and fruit development include seedling blight and damping off on seedlings, foot rot and collar rot on young plant stem, stem blight and canker on stems and branches, early blight and leaf spot on leaves, blossom blight on the calyx, black rot and hard rot on fruit and fruit drop on fruit and petioles. Leaf spot symptoms are characteristic for early blight. They are circular up to ½ inch in diameter, brown and contain dark concentric ring. The spots occur singly or in large number on each leaf, yellowish areas may develop on effected leaves and eventually they turn brown and usually drop from the plant.

In the absence of fungicide treatment, maximum fruit infection for susceptible varieties was about 30%. Potential yield in processing tomato and fruit size are reduced on an average of 30% and 10%, respectively (Sherf & Macnab, 1986). Choulwar and Datar (1989) assessed the efficacy of eight fungicides (copper oxychloride, zineb, mancozeb, carbendazim, dithianon, iprodione, thiophanate-methyl and captafol) to reduce mycelial growth of *A. solani in vitro*. Mancozeb (1000 ppm) was the most effective (77% growth inhibition) followed by captafol. Carbendazim and thiophanate-methyl were not effective. Increasing concentration of fungicide generally decreased mycelial growth. Fadl *et al.* (1985) in laboratory tests with 3 fungicides observed that Ridomil MZ (metalaxyl) was most inhibitory to linear growth at low concentration followed by Trimiltox Fort and Bravo 500 (chlorothalonil) whereas at higher concentrations Trimiltox Fort performed best. Sinha and Prasad (1991) tested seven fungicides in the field over 3 seasons against *A. solani*. Dithane M-45 (Mancozeb) @ 0.2% was the best and cost effective treatment with the highest yield. Khade and Joi (1980) reported that all nine fungicides reduced incidence, but the highest yield increases were obtained with Dithane M-45 (Mancozeb), blue Cu 50, Cuman L, Dithane Z-78 (Zineb) and Difolatan (Captafol). Choulwar and Datar (1988) reported that in tests, 12 treatments of 1-6 sprays of 0.2% mancozeb applied at different times after transplanting significantly reduced the intensity of early blight caused by *A. solani*. The lowest disease intensity and highest yield was obtained with 6 early sprays followed by 6 late and 5 early sprays. Early sprays were generally more effective than equal numbers of late sprays. The yields were negatively correlated with disease intensity. Vidhyasekaran (1983) reported that both mancozeb and Captafol effectively controlled *A. solani* and *Septoria lycopersici*. These treatments reduced defoliation and increased fruit production. Fruits from sprayed plots had significantly more sugars and vitamin and less phenolics.

The attack of early blight has been observed on tomato crop in several parts of the NWFP. It is feared that losses from this disease may increase year after year if no protective measures are adopted well ahead of time. However, very little work has been done in this province on control of this disease. As a first step towards achieving this goal, this project was conducted to study its in-vitro control.

Materials and Methods

Diseased specimens of early blight were collected from tomato growing areas of the NWFP. The infected leaves and stems were cut into small pieces, surface sterilized with mercuric chloride (0.1%) for 30 seconds, rinsed thrice with sterile distilled water, blotted dry and incubated at 25°C on Potato Dextrose Agar (PDA) medium for 7 days. Pure culture of the fungus was maintained at 4 C for further studies.

Poison Food Technique (PFT) was used to test different concentrations (250, 500, 750, 1000 ppm) of the fungicides Antracol, Benlate, Copper oxychloride, Dithane M-45, Ridomil, Topas, Topsin, Dithane M-45 + Ridomil, Dithane M-45 + Topas, and Dithane M-45 + Benlate against *A. solani*. Different quantities of the fungicides were mixed with the PDA medium at 50°C before pouring. Each treatment was replicated five times. One treatment was maintained as control where no fungicide was added to the PDA medium. After mixing the fungicides and solidification of the medium, the fungus *A. solani* was seeded in the centre of each petri plate using 5mm agar disc having active mycelial growth of the fungus. The plates were incubated at 25°C for seven days. At the end of incubation period, radial colony growth was measured (cm) in each treatment. The data were analyzed statistically by analysis of variance and the means were compared by Duncan's Multiple Range Test (Steel & Torrie, 1980).

Results and Discussion

The different treatments showed significant differences ($P < 0.05$) among themselves (Table 1). At 250 ppm concentration, the lowest growth (2.02 cm) was observed in the treatment Dithane M-45 and the highest (7.4 cm) in control. At this concentration, treatment Dithane M-45 was significantly different from all other treatments except treatment Dithane M-45 + Benlate. The control was only non-significantly different from treatment Antracol. Combination of Dithane M-45 with other fungicides such as Ridomil and Topas was not as good as with Benlate (Dithane M-45 + Benlate) which caused the second lowest growth. At 500 ppm, the lowest growth was observed again Dithane M-45, followed by that in treatment Topas. Both these treatments were non-

Table 1: Effect of different concentrations of fungicides on mycelial growth of *A. solani in vitro*

Fungicides	Mycelial growth (cm) Fungicide concentrations				Mean
	250 ppm	500 ppm	750 ppm	1000 ppm	
Antracol	6.49 abc	4.59 f-l	2.60 p-t	3.75 l-p	4.35 cd
Benlate	5.05 d-j	5.63 c-f	3.65 l-p	5.28 c-l	4.90 bc
Copper Oxychloride	5.8 c-f	5.55 c-g	1.43 tuv	3.45 l-q	4.06 de
Dithane M-45	2.02 rst	1.81 stu	0.67 uv	0.22 v	1.18 g
Dithane M-45 + Benlate	2.21 q-t	4.27 h-n	1.55 tu	3.12 m-r	2.78 f
Dithane M-45 + Ridomil	4.12 l-n	4.57 f-l	3.57 l-p	3.77 k-p	4.01 de
Dithane M-45 + Topas	4.08 l-n	6.30 a-d	3.42 l-q	3.91 j-o	4.43 bcd
Ridomil	4.36 g-m	4.92 e-k	2.20 q-t	3.35 l-q	3.78 e
Topas	5.18 d-l	3.02 n-s	2.68 o-t	3.56 l-p	3.61 e
Topsin	4.93 e-k	5.39 c-h	3.65 l-p	6.06 b-e	5.00 b
Control	7.4 a	7.20 ab	5.68 c-f	7.30 ab	6.90 a
Mean	4.70 a	4.84 a	2.83 c	3.98 b	--------

Figures in the same column followed by different letters are significantly different from one another at 0.05 level of significance.

significantly different from one another. The highest growth was recorded in the control treatment. The latter differed significantly from other treatments with the exception of Dithane M-45 + Topas. In control treatment, the growth was higher by 7.20% and 6.30% than treatments Dithane M-45 and Topas, respectively. It was also higher by 7.40% than the overall mean of the fungicides at this concentration, indicating that fungicide application has after all decreased the growth of the fungus.

Fungicide treatments at 750 ppm differed significantly from one another ($P < 0.05$). The lowest growth was in treatment Dithane M-45 (0.67 cm), followed by Copper oxychloride and then Dithane M-45 + Benlate. The three treatments did not differ significantly from one another. The highest growth was recorded in control. The higher growth in this than the other treatments indicated that fungicide treatment depressed the mycelial growth significantly. Growth of *A. solani* varied significantly at 1000 ppm level of different fungicides. The lowest and the highest growth were recorded in treatment Dithane M-45 and in treatment where no fungicide was applied, respectively. These two treatments showed significant differences from one another and from other treatments. None of the combinations including Dithane M-45 and the systemic fungicides were better than treatment Dithane M-45 alone in reducing colony growth of fungi.

The overall mean calculated for each fungicide showed that the lowest value (1.18 cm) was in case of fungicide Dithane M-45. This was followed by treatment Dithane M-45 + Benlate (2.78 cm) and then Topas (3.61 cm). The highest value was for control. Among the different concentrations being used, the overall lowest growth was calculated for 750 ppm (2.83 cm) and the highest for 500 ppm (4.84 cm). In case of the best fungicide (Dithane M-45), the growth of the fungus was the lowest at 1000 ppm and the highest at 250 ppm. This was the reverse in other treatments where the lowest growth was observed at 750 ppm. The latter concentration can also be recommended for Dithane M-45 as the two treatments (1000 pmm and 750 ppm) of the fungicide were non-significantly different from one another.

In treatment where fungicides were used, the mycelial growth of *A. solani* was depressed substantially as compared to the treatment where no fungicide was used. This indicated the importance of fungicide used in controlling disease. However, these fungicides were variable in their effect which depended on their type and concentration being used. Dithane M-45 caused maximum reduction in mycelial growth at higher concentrations. This broad-spectrum fungicide is used for the control of a number of fungal diseases. The availability of this fungicide at reasonable price in the market is an additional advantage of Dithane M-45 over other fungicides for the effective control of early blight in tomato.

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