

STUDY ON THE IMPACT OF SELECTED CONTACT AND SYSTEMIC FUNGICIDES ON MYCELIAL GROWTH OF *Alternaria alternata* *in vitro*

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ABSTRACT

Alternaria alternata isolated from the infected fruit of *Lagenaria*, (bottle gourd) was used as the test pathogen to determine the impact of selected contact and systemic fungicides on its mycelial growth *in vitro*. The contact fungicides used were Blitox-50, Thiram, Captan 50 WP, while the systemic fungicides were chlorothalonil, carbendazim 50% (Bavistin) and Mancozeb. Maximum inhibition of mycelial growth of the aforesaid phytopathogenic fungus was observed in culture which was treated with 0.25% of Mancozeb, followed by carbendazim 50 WV. Similarly, Captan 50 WP gave better response in comparison to the Thiram and Blitox-50 at the similar concentrations. It was further observed that Blitox-50 was the least effective among the contact fungicides used here. It was further observed that sporulation of the fungus was equally affected by the same fungicides which were effective on mycelial growth. Here also the efficacy of Mancozeb was better than that of the other fungicides while the impact of Blitox-50 was the lowest.

KEYWORDS: *In vitro*, *Lagenaria*, Fungicides, Phytopathogen, Systemic Fungicide, Sporulation

Alternaria alternata was isolated from infected fruit of bottle gourd, (*Lagenaria siceraria*) an important vegetable. It is one of the vegetable that was first domesticated by Human kinds. The fruit has different shape and size. Immature fruits are being used as vegetable and for other preparations. Bottle gourd is one of the lowest calorie vegetables carrying just 14 calories per 100 g. it is one of the vegetable recommended by the dieticians in weight control programs. The fruit contains folate, 6 µg/100 g that helps to reduce the incidence of neural tube defects in the new borne, when taken by the mother during their early months of pregnancy. Its fruits also provide Vitamin C, that is one of the powerful natural antioxidants that helps the human body scavenge harmful free radicals, which labeled as one of the reasons for cancer development. This vegetable is also a modest source of vitamin B₃, Vitamin B₅, and Vitamin B₆, Vitamin A and minerals such as, calcium, iron, zinc, potassium, manganese and magnesium, phosphorus, selenium etc. However, fruits are attacked by different pathogens at the different stage of development. Such infections are more prevalent during summer, when the vegetable is cultivated in the field and the plants are allowed to grow in contact with the soil. The infected fruits are deformed in shape and colour in such a way that their market value becomes zero. Such incidence is considered to be an important limiting factor in profitable cultivation of this vegetable crop at commercial scale in the rural areas of North Bihar in general and Muzaffarpur and Vaishali districts in particular. These fungal pathogens do damage the fruits starting from the beginning to maturity and thus

causes heavy loss to the farmers. The farmers harvest such fruits and dump them in one of the corner of the field; therefore, the infected fruits serve as the source of secondary infection. Keeping in view the importance of the disease, the present study was conducted and *Alternaria alternata* was isolated from the infected fruits.

The fungus was identified and maintained in the laboratory for further studies. In this way the efficacy of selected fungicides was determined *in vitro*, so that it may be applied *in vivo* by the growers.

Impact of different fungicides and natural plant extracts have been studied by Rao and Rajgopalan (1982); Vishwakarma and Pandeya, (1995); Singh and Majumdar (2007); Khan *et al*; (2003); Prasad and Naik (2003); Surviliene and Dambrus Kiene (2006); Thaware *et al*; (2010); Chethana *et al*; (2011); Chethana *et al*; (2012); Gondal *et al*; (2012); Mishra and Gupta (2012); Biljana (2013); Roopa *et al*; (2014); Theja Kumar (2016) and Hariprasad *et al*; (2017).

MATERIALS AND METHODS

Already isolated and cultured *Alternaria alternata* was used as the source of inoculum. The fungicides such as – Blitox-50, Thiram, Captan-50, Chlorothalonil, Carbendazim-50 (Bavistin) and Mancozeb were purchased from the local suppliers of Agrochemicals. These fungicides were diluted separately with the help of glass distilled water in the range of 0.10, 0.15, 0.20 and 0.25%. They were mixed in the liquid culture medium (Czapek-Dox) in culture

flasks. Three flasks were used for each treatment. 7 days old culture of *Alternaria alternata*, maintained on Potato Dextrose Agar was used as the source of inoculum. From this culture 6 mm disc of mycelial bit was taken from the periphery of the culture plate, with the help of pre-sterilized blade and was inoculated in the aseptic conditions of Laminar flow air chamber. The inoculated Petri-plates were incubated in BOD incubator maintained at $25\pm 1^{\circ}\text{C}$. Culture flasks containing Czapek-Dox broth, without fungicide was also inoculated that served as control.

Dry mycelial weight and spore density were calculated from 10 days old culture. The culture was filtered through, pre-weighed and pre-sterilized Whatman's filter paper No.-42.

The mycelial mat was washed with hot water to remove even the trace of the culture medium. The filter paper along with the mycelial mat was weighed which gave the fresh mycelial weight. This was placed in hot air oven at 60°C . Weight was taken regularly till a constant weight was obtained. This was considered as the dry mycelial weight. This techniques was used for all treated and control flasks. Similarly, spore suspension was taken and diluted through serial dilution. Now number of spores in one ml was determined which indicated the sporulation potential. The percentage of inhibition of mycelial growth was determined by applying the standard formula such as:

$$\text{Percentage of growth inhibition} = \frac{C-T}{C} \times 100$$

Where, C = Dry mycelial weight in control condition.

T = Dry mycelial weight in treated condition.

The means of the value have been used as data for above calculation.

RESULTS AND DISCUSSION

Six fungicides were evaluated at four different concentrations (0.10, 0.15, 0.20 and 0.25%) to evaluate their efficacy against *Alternaria alternata* isolated from rotten bottle gourd fruits by poisoned food technique as mentioned in "Materials and Methods". All experiments were performed in triplicate and each time 15 cultures were used. The mean of the results was used as data for discussion, and are represented by graph. From the graph it is evident that Mancozeb at 0.25% concentration could inhibit hundred percent mycelial growth of *Alternaria alternata in vitro*.

This was followed by Carbendazim 50% at the same concentration where the percentage of inhibition was noted as 86.28%.

It may further be noted from the table that Blitox-50 was less effective at this concentration and the percentage of inhibition of mycelial growth was only 61.50. This was followed by Thiram where the percentage of inhibition was 67.86%. The data represented by the graph, clearly reveal that all the fungicides used here had inhibitory impact on the mycelial growth of the pathogen *in vitro* at their different concentrations, however the quantum of inhibition was different for different fungicide even at the same concentration. As was noted from the results that systemic fungicides viz., Chlorothalonil, Carbendazim 50% and Mancozeb at their different concentrations could inhibit the mycelial growth of the pathogen more efficiently than that of the contact fungicides such as Blitox-50, Thiram and Captan 50 WP. Further it was noted that among the systemic fungicides Mancozeb was more efficacious with respect to the other two systemic fungicides at all the four concentrations used here.

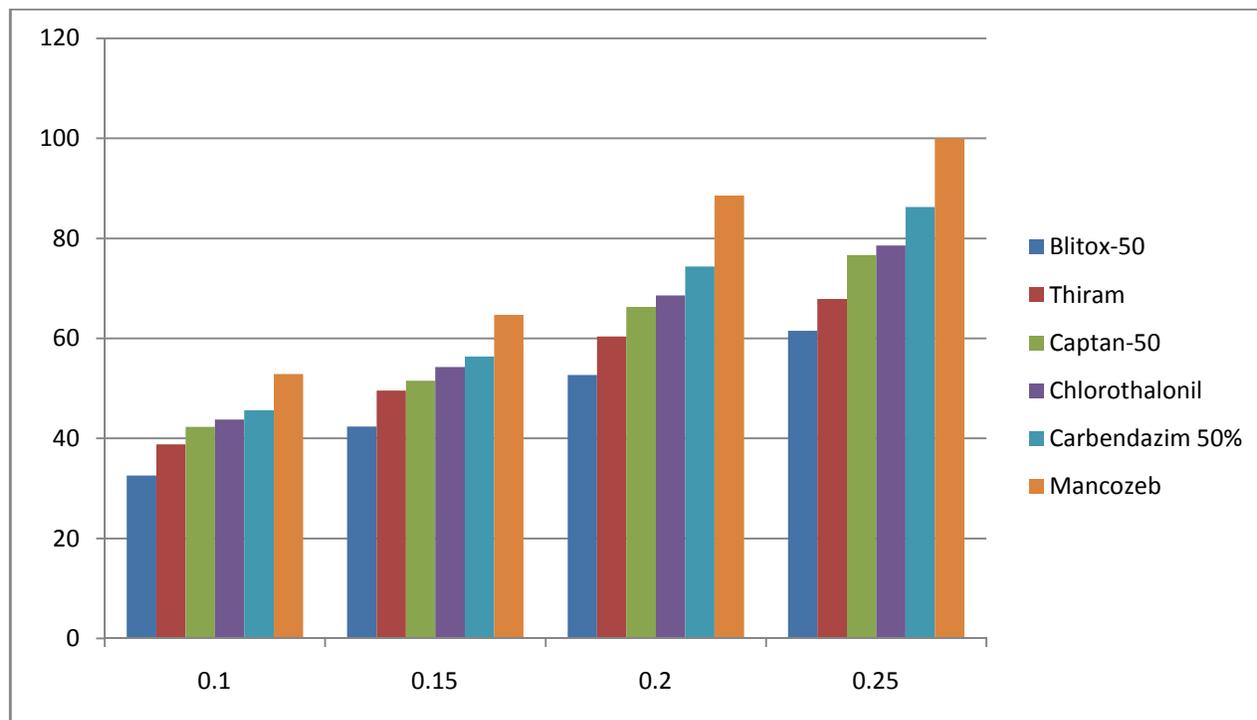
Present finding is in agreement with the findings of Kautwa *et al*; (2014) and Theja Kumar and Devappa (2016) who also reported that Mancozeb was superior in comparison to the other fungicides with respect to the inhibition of the mycelial growth of *Alternaria alternata in vitro*. Blitox-50, was found to be less effective at its various concentrations, is corroborated by the findings of Khan *et al*; (2003); Surviliene and Dambrauskiene (2006); Thaware *et al*; (2010); Mishra and Gupta (2012); Roopa *et al*; (2014); Hariprasad *et al*; (2017).

CONCLUSION

Experiments were performed for the *in vitro* evaluation of efficacy of different fungicide on the mycelial growth of *Alternaria alternata* isolated from the infected fruits of bottle gourd. Mancozeb at its various concentrations was found to be more efficacious, with respect to inhibition of the mycelial growth *in vitro*. This fungicide at this concentration may be used *in vivo* that will be more economic and eco-friendly.

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Graph 1: Showing percentage of inhibition of mycelial growth in presence in presence of different fungicides at four different concentrations.

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