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## Evaluation of different botanicals and chemical fungicides against *Alternaria lini* in *In-vitro* condition

#### Arun Kumar, Dushyant Kumar, Joni Kumar, SK Singh, Krishan Pal, Yogandar Sain and Maneesh Kumar

#### Abstract

The present investigation was carried on effect of different botanicals and other fungicides on *Alternaria lini*. The plant extracts of different botanicals were evaluated under in-vitro conditions. The poisoned food technique was used to evaluate the efficacy of botanicals at concentrations of 50, 100 and 150 ppm with three replications. Potato dextrose agar was used as medium and required quantity of each botanical extract was added separately to get a required concentration. The plant extracts inhibited the growth of the pathogen in the medium. The results of *in-vitro* studies revealed that Garlic bulb extract @ 150ppm significantly reduced the mycelial growth of *Alternaria lini* as 68.6 percent inhibition followed by Neem leaf extract @ 150 ppm and Ginger rhizome extract @ 150 ppm as 64.3% and 57.2% inhibition respectively. Among the six non-systemic fungicides evaluated at 1000 ppm, Mancozeb completely inhibited of the fungus growth (100%) and significantly superior over other treatments.

Keywords: Alternaria lini, botanical, fungicides, in-vitro conditions

#### Introduction

Linseed (Linum usitatissimum L.) belongs to the family "Linaceae" and the genus "Linum" is widely cultivated in Northern India during Rabi season and is the only species in the family, which is of economic importance. Linseed is an important oilseed and fiber crop grown both for its seed as well as fibre which is used for the manufacture of linen. The seed contains a good percentage of oil varying from 33 to 47 percent in different varieties. The oil is edible and also due to its quick drying property is used for the preparation of paints, varnishes, printing ink, oilcloth, soap, patent leather, and water proof fabrics. It is the best herbal source of Omega-3 and Omega-6 fatty acid which improve human nervous system and diabetic nephropetic disease and also useful in reducing sexual problem in women (Hurteau, 2004) <sup>[6]</sup>. Alternaria blight or black bud disease caused an annual loss of 28-60% in yield (Chauhan and Srivastava, 1975) <sup>[4]</sup>. In addition to target organism, pesticides also kill various beneficial organisms. Their toxic forms persist in soil and contaminate the whole environment. A prospect of biological control of soil-borne plant pathogens using most promising botanicals, the botanicals has been described. Successful reductions of Alternaria lini in many crops with application of different species of botanicals have been found. However, it is also reported that all the different botanicals are not equally effective in control of pathogen in vitro conditions to control diseases. Therefore, specific isolates are needed for successful control of a particular pathogen.

#### **Materials and Method**

The present investigation was carried out to evaluate the plant extracts collected from different plant species to know their toxicant properties against Alternaria blight pathogen (Table-1). The plant extracts were evaluated *in vitro* through poisoned food technique (Carpenter, 1942; Nene and Thapiyal, 1993) <sup>[3, 8]</sup>. The poisoned food technique was followed to evaluate the efficacy of botanicals in laboratory against *A. lini* at concentrations of 50, 100 and 150 ppm with three replications. For this, fresh healthy plant parts of 100g (leaves/rhizome/bulb) were collected from field, then they were washed with distilled water, air dried and crushed in 100 ml of sterile water. The crushed product was filtered by using muslin cloth. The filtrate (100%) was further diluted to required concentrations of 50 ppm, 100 ppm and 150 ppm. Potato dextrose agar was used as medium and required quantity of each botanical extract was added separately to get a required concentration. The botanical extract was thoroughly mixed with

PDA medium and sterilized. About 20 ml of poisoned medium was poured to each of the 90 mm Petri dishes and allowed for solidification. The actively growing twelve-day old culture of *A. lini* was carefully cut from the periphery using a cork borer and transferred aseptically to the centre of each Petri dish containing the poisoned solid medium. Suitable control was maintained by growing the cultures on PDA without the plant extract. The plates were incubated at  $25\pm1$  °C for twelve days and the colony diameter was recorded.

Percent inhibition	_	$\frac{\text{Radial growth in control - Radial growth in treatment}}{100} \times 100$
Percent innibition	-	Radial growth in control

**Table 1:** List of plant extracts used in the experiment

No.	Botanical name	Common name	Plant part used
1.	Azadirachta indica Juss	Neem	Leaf
2.	Zingiber officinale	Ginger	Rhizome
3.	Allium sativum L.	Garlic	Bulb
4.	Bougainvillea sp. L.	Bougainvilla	Leaf
5.	Eucalyptus globules Labill	Eucalyptus	Leaf

#### In vitro efficacy of chemical fungicides

The efficacy of three systemic and six non-systemic fungicides were evaluated against *A. lini* at three concentrations on potato dextrose agar medium using poisoned food technique. Non-systemic fungicides were tested in laboratory at concentration of 500 ppm, 750 ppm and 1000 ppm and systemic fungicides at 100 ppm, 250 ppm and 500 ppm with three replications in each treatment. The radial

growth of the fungus on poisoned food medium was recorded at 12 days after inoculation. The different fungicides tested are mentioned below.

Table 2: List of fungicides used in the ex
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Sl. No	Common name	Trade name				
Non-systemic fungicides						
1.	Copper oxy chloride	Blitox 50WP				
2.	Carbendazim 12% + Mancozeb 63%	Saaf 75 WP				
3.	Mancozeb	Indofil M-45 75WP				
4.	Carbendazim 25% + Iprodione 25%	Quintal 50 WP				
5.	Metalaxyl 4% + Mancozeb 64%	Ridomil 68WP				
6.	Hexaconazole 4% + Zineb 68%	Avtar 72 WP				
Systemic fungicides						
1.	Tricyclazole	Beam 75WP				
2.	Hexaconazole	Contaf 5EC				
3.	Propiconazole	Tilt 25 EC				

#### Result

Six plant extracts were evaluated at three concentrations (50 ppm, 100 ppm and 150 ppm) in the laboratory for their efficacy against *A. lini* through poisoned food technique as detailed in Materials and Methods. The plant extracts inhibited the growth of the pathogen in the medium. The results of *in-vitro* studies revealed that Garlic bulb extract @ 150 ppm significantly reduced the mycelial growth of *Alternaria lini* as 68.6 percent inhibition followed by Neem leaf extract @ 150 ppm and Ginger rhizome extract@ 150 ppm as 64.3% and 57.2% inhibition respectively (Table 2 and Fig. 1).

Table 3: In vitro evaluation of botanicals against A. lini causing Alternaria blight disease on linseed

C No		Percent inhibition				
S. No.	Fungicides/Concentration	50 ppm	100 ppm	150 ppm	Mean	
1.	Azadirachta indica	38.7	55.8	63.6	54.37	
2.	Allium sativum L.	43.5	58.8	64.3	55.53	
3.	Zingiber officinale	32.2	48.5	57.2	45.97	
4.	Allium cepa	27.4	44.1	55.7	42.40	
5.	Bougainvillea sp. L.	22.6	33.8	57.1	37.83	
6.	Eucalyptus globules Labill	16.7	29.4	38.6	28.23	
7.	Control	0.0	0.0	0.0	0.0	
	S.Em ±	0.32				
	CD @ 5%	0.98				

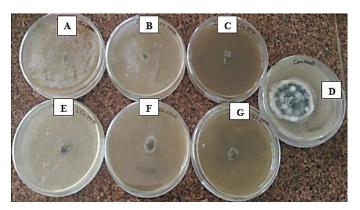


Plate 1: (A) Neem (B) Garlic (C) Ginger (D) Control (E) Onion (F) Bougainvillea (G) Eucalyptus

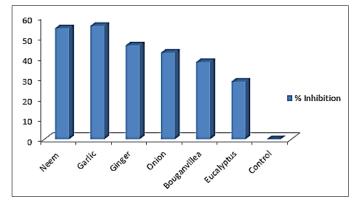


Fig 1: In-vitro evaluation of botanicals against Alternaria lini

### *In vitro* evaluation of chemical fungicides against *Alternaria lini*

The efficacy of three systemic and six non-systemic fungicides were evaluated against *A. lini*at three concentrations as described in "Material and Methods". The results indicated that there was difference among non-systemic and systemic fungicides in inhibiting the growth of *A. lini*. Among the six non-systemic fungicides evaluated at 1000 ppm, Mancozeb completely inhibited of the fungus growth (100%) and significantly superior over other

treatments, followed by Quintal 50 WP (88.89%), Ridomil 68WP (86.30%), Saaf 75 WP (82.96%). Least growth inhibition was observed in Blitox 50 WP (77.04%) followed by Avtar M-45 72 WP (77.79%). Higher concentrations of the fungicides were found more effective against *A. lini* compare to lower concentrations. Mancozeb was found completely in inhibiting the mycelial growth of *A. lini* at 1000 ppm and found significantly superior over the other lower concentrations.

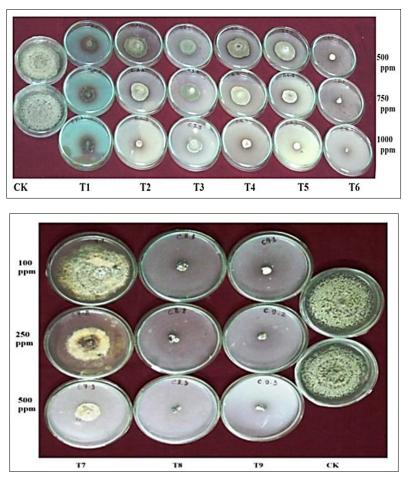


Plate 2: Carbendazim was found less effective with 85.19 per cent reduction in growth of *A. lini* at 100 ppm than at 250 ppm (90.37%) and 500 ppm (100%)

Among the different systemic fungicides, per cent inhibition of Tilt Carbendazim at 500 ppm (100%) gave complete growth inhibition of the fungus, followed by Contaf 5EC (93.70%). Least inhibition of mycelial growth was observed in Beam 75 WP (64.07%) at 500 ppm and it was not effective in reduction of growth of the fungus at 100 ppm (17.04%). Carbendazim was found less effective with 85.19 per cent reduction in growth of *A. lini* at 100 ppm than at 250 ppm (90.37%) and 500 ppm (100%).

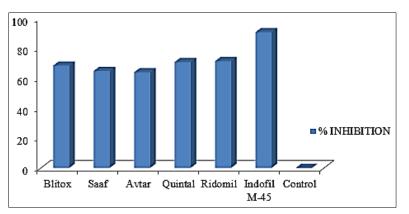


Fig 2: In vitro evaluation of non-systemic fungicides against A. Lini

#### Discussion

#### In vitro evaluation of botanicals against Alternaria lini

Keeping this in view, investigation was undertaken for screening of six plant extracts. They were evaluated at three concentrations (50 ppm, 100 ppm, 150 ppm) in the laboratory for their efficacy against *A. lini* through poisoned food technique. The plant extracts showed growth inhibition of the pathogen by increasing their concentration in the medium. The result of *in vitro* studies revealed that Neem leaf extract @ 150 ppm significantly reduced the mycelia growth of *Alternaria lini* as 68.6percent inhibition followed by Garlic bulb extract @ 150 ppm and Ginger rhizome extract@ 150 ppm as 64.3% and 57.2% inhibition respectively. The result are in conformity with the work of Rashmi and Yadav (1999) <sup>[10]</sup>, they reported that all the plant extracts were less effective at lower concentration and growth inhibition percentage.

The present investigation of various botanicals inhibiting the growth of *A. lini* is in line with the earlier findings (Patni *et al.*, 2005, Dalpati *et al.*, 2010, Anamika and Sobita, 2011; Arun Kumar, 2008 and Kota, 2003) <sup>[1, 2, 5, 7, 9]</sup>. Plant extracts that moderately inhibit development of *A. lini* are promising and need to be exploited in management of *Alternaria* leaf blight of linseed.

#### In vitro evaluation of fungicides against Alternaria lini

The efficacy of three systemic and six non-systemic fungicides were evaluated against A. lini at three concentrations as described in "Material and Methods". The results indicated that there was difference among nonsystemic and systemic fungicides in inhibiting the growth of A. lini. Among the six non-systemic fungicides evaluated at 1000 ppm, Avtar 72 WP completely inhibited of the fungus growth (100%) and significantly superior over other treatments, followed by Quintal 50 WP (88.89%), Ridomil 68 WP (86.30%), Saaf 75 WP (82.96%). Least growth inhibition was observed in Blitox 50WP (77.04%) followed by Indofil M-45 75WP (77.79%). Higher concentrations of the fungicides were found more effective against A. lini compare to lower concentrations. Avtar 72 WP was found completely in inhibiting the mycelial growth of A. lini at 1000 ppm and significantly superior over the other lower found concentrations.

Among the different systemic fungicides, percent inhibition of carbendazim at 500 ppm (100%) gave complete growth inhibition of the fungus, followed by Contaf 5EC (93.70%). Least inhibition of mycelial growth was observed in Beam 75 WP (64.07%) at 500 ppm and it was not effective in reduction of growth of the fungus at 100 ppm (17.04%). Carendazimb was found less effective with 85.19 per cent reduction in growth of *A. lini* at 100 ppm than at 250 ppm (90.37%) and 500 ppm (100%).

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