

Original Research Article

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Effect of Fungicides and Botanicals against Leaf Rot Disease Caused by *Fusarium oxysporum* in Aloe vera (*Aloe barbadensis* Miller)

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ABSTRACT

Keywords

Aloe vera, Leaf rot, *Fusarium oxysporum*, Fungicides and Botanicals.

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Aloe vera is an oldest medicinal plant grown worldwide. The first written record about the use of *Aloe vera* is found on 6,000 years old clay tablets found in Mesopotamia. *Aloe vera* is a stemless succulent plant growing to 80-100 cm tall, spreading by offsets and root sprouts. The result clearly shows that all chemical control the maximum per cent disease control were recorded in treatment T₃ = Propiconazole 25 EC @ 0.25% PDC (65.20, 65.71 and pooled 65.46) followed by T₂ = Mancozeb 75 WP@ 0.25% PDC (61.05, 62.28 and pooled 61.67), and T₁ = Carbendazim 50 WP@ 0.25% PDC (59.36, 60.28 and pooled 59.82) during 2014 and 2015. Among the botanicals maximum per cent disease control were recorded T₄ = Neem leaf extract @ 5% PDC (58.26, 59.16 and pooled 58.71) followed by T₅ = Garlic bulb extract @ 5% PDC 51.81, 51.79 and pooled 51.80) and T₆ = Tulsi leaf extract @ 5% PDC 44.99, 45.84 and pooled 45.41) control leaf rot of *Aloe vera* caused by *Fusarium oxysporum*.

Introduction

Aloe vera (*Aloe barbadensis*) is a perennial, drought resisting succulent plant belong to the family Liliaceae with its believed to have originated in African continent specifically in Egypt (Daodu, 2000). There are over 250 species of *Aloe vera* growing around the world however, only *Aloe barbadensis*, *Aloe ferox* and *Aloe arborescence* are used as herbal drugs. It is distributed to other tropical countries like South Arab, India, and East Asia. *Aloe vera* is grown largely in South America, Central America, Australia and Africa. *Aloe vera* is cultivated in fairly large area in many in parts of India viz. Eastern part

of Utter Pradesh Chhattisgarh, Maharashtra, Madhya Pradesh and Gujarat, Tamil Nadu and Andhra Pradesh.

Aloe vera is a stemless succulent plant growing to 80-100 cm tall, spreading by offsets and root sprouts. It is a stemless or very short-stemmed plant growing to 80-100 cm tall, spreading by offsets and root sprouts. The leaves are lanceolate, thick and fleshy, green to grey-green, with a serrated margin. The flowers are produced on a spike up to 90 cm tall, each flower pendulous, with a yellow tubular corolla 2-3 cm long. When a leaf is

cut an orange yellow sap drips from the open end. A stabilized product is incorporated in a wild variety of preparation, including juice, gel, ointments, cream, lotion and shampoos (Daodu, 2000). At the rate of 3 or 4 leaves cut from each plant. On an average, the yield per acre annually is about 6000 kg. In the present study, the leaf yield showed significant variation among the accessions and it varied from 0.599 to 2.922 kg per plant per year (Abhila and Jessyeutty, 2010). As medicinal herb *Aloe vera* has been used externally to treat various skin conditions such as cuts, burns and eczema. Furthermore, *Aloe vera* gel has been reported to be very effective for the treatment of sore and wounds, skin cancer, skin disease, cold and cough, constipation, pile and fungal infection (Daodu, 2000; Djeraba and Quere, 2000; Olusegun, 2000), while Davis and Moro (1989) reported aloe plants can be used for treatment of asthma, ulcer and diabetes. The ten main area of chemical constituents of *Aloe vera* include amino acid, anthraquinones, enzymes, minerals, vitamins, lignins, monosaccharide, polysaccharides, salicylic acid, saponins and steroids (Barcroft and Myskja, 2009). The row pulp of *Aloe vera* contains approximately 98.5% water, while the mucilage or gel consists of about 99.5% water. The remaining 0.5 to 1% solid material consist of a range of compounds including water-soluble and fat soluble vitamins, minerals, enzymes, polysaccharides, phenolic compound and organic acid (Hamman, 2008). The partial control of fungus may be achieved with the application of chemicals and botanicals by the controlling the fungus (*Fusarium oxysporum*) population but complete safer protection from the fungus through host plant resistance is more preferred and effective option, but their long term stability virtue of true resistance are need for their confirmation under field condition because the role are mineral metabolism and total soluble phenol an imparting resistance/susceptibility against

fungus diseases of plant has been also infested. A leaf rot diseases caused by *Fusarium oxysporum* were reported from the USA. In India, leaf rot is one of the most serious fungal diseases affecting the commercial cultivation of *Aloe vera* in North India. Among the fungal diseases, leaf spot of *Aloe vera* caused by *Fusarium oxysporum* is one of the most wide spread and destructive disease of *Aloe vera* under field conditions. Severity grade of the leaf rot of *Aloe vera* caused by *Fusarium oxysporum* are given in the Table 1.

Materials and Methods

The experiment was conducted during *Kharif* season 2014 and second 2015, at experimental farm of Medicinal and Aromatic Plants of N.D.U.A.T. Kumarganj, Faizabad. Randomized Blok Design (RBD) was adopted with three replications. The infected leaves collected from infected plants in sterilized poly bags. Thus, collected typical symptom showing parts were kept in rough dry envelopes and marked clearly mentioning location, infected parts, reaction types, date of collection etc. and brought to the laboratory for isolation of the pathogen. The samples were dried for 24 hours in shade in order to remove excess surface moisture. After drying the samples were kept in B.O.D. incubator in paper envelope marked with date of collection maintained at 6-8 °C for isolation and further studies. The peeled potatoes were cut into 12 mm cubes. Two hundred grams of potato cubes were rinsed in water and boiled for 20 minutes in 500 ml water. Potato broth was filtered through cheese cloth. At the same time, agar was also melted in 500 ml of water by heating. Potato broth was then poured into the melted agar and dextrose was added in it. The final volume was made up 1000 ml by adding distilled water. The pH was adjusted to 7.0; the stock solution was then poured in to sterilized 250 ml Erlenmeyer conical flasks

up to 1/3rd only. Then it was plugged with non-absorbent cotton. These poured flasks were kept for sterilization in autoclave at 15psi for 20 minutes.

$$\text{Per cent disease intensity (\%)} = \frac{\text{Sum of total numerical rating}}{\text{Total No. of leaves examined} \times \text{highest rating}} \times 100$$

$$\text{Per cent diseese control (\%)} = \frac{C - T}{C} \times 100$$

Where,

C = Per cent disease incidence in untreated plot

T = Per cent disease plot incidence in treated plot

Results and Discussion

Isolation, purification and identification

Isolation

Isolations were made from infected leaves. The pathogen was isolated on Modified Potato-Dextrose-Agar medium (Singh and Chaube, 1970) in Petri plate by transferring them after surface sterilization. After the mycelial growth, the fungus was purified through hyphal tip culture method. Subsequently, the culture was maintained on PDA slant for further studies. The pathogen under study was identified as *Fusarium oxysporum* on the basis of its cultural and morphological characters described by Booth (1971).

Purification

The pure culture of isolated pathogen was done by single spore isolation. A dilute spore suspension was poured on plain agar petriplate to form a very thin layer on it and spores allowed settling down on the agar surface. Settled spores were separated out from each other, selected under the microscope and enriched with the help of dummy culture in petriplates. They were

lifted along with agar blocks and transferred to petriplates containing sterilized 2% PDA. After proper growth of fungus obtained by single spore culture regular sub-culturing was done to check contamination till pure cultures were obtained. These cultures were sub-cultured at monthly intervals and maintained on PDA slant under refrigeration at 6 to 8°C temperature for further studies.

Identification

White fluffy colony of *Fusarium oxysporum* was seen on PDA and the microscopic studies revealed that the mycelium was septate and hyaline, microconidia were slightly curved and coma shaped and had 0-1 septa while macroconidia were strongly curved or hooked at apex, smooth hyaline and had 1-4 septa. The chlamydospores were oval to globose hyaline and found in solitary or chain form.

Pathogenicity test

Pathogenicity test was conducted on *Aloe vera* leaves by inoculation and germination technique in Petri plates. One leaf each was placed in 30 petriplates, inoculated with spore suspension of *Fusarium oxysporum* using moist sterilized filter paper, plates were incubated at 25 ± 2°C and are regularly observed for development of symptoms. The leaf spot symptoms appeared as minute dark brown coloured spots with prominent halo on inoculated leaves in 10 days' time. The re-isolation was done to confirm the test pathogen from infected portion of the leaves taken for pathogenicity test were taken and cultured in petri plates and incubated at 25 ± 2°C. The whitish velvety growth similar to that of test fungus appeared after 7 days of incubation in petriplates; slides were prepared and examined under microscope. The cultural and morphological behaviour of the pathogen i.e., isolate from naturally infected was similar to the standard pure culture proves its pathogenicity.

Table.1 Effect of fungicides and botanicals on per cent disease incidence of *Aloe vera* leaf rot

S.No.	Treatments	Per cent Disease Incidence		
		2014	2015	Pooled
1	T ₁ = Carbendazim 50 WP @ 0.25%	36.47 (37.15)	36.39 (37.10)	36.43 (37.13)
2	T ₂ = Mancozeb 75 WP @ 0.25%	33.06 (35.08)	29.26 (32.75)	31.16 (33.92)
3	T ₃ = Propiconazole 25 EC @ 0.25%	28.24 (32.10)	26.61 (31.05)	27.42 (31.57)
4	T ₄ = Neem leaf extract @ 5%	36.19 (36.98)	34.51 (35.96)	35.35 (36.47)
5	T ₅ = Garlic bulb extract @ 5%	39.38 (38.86)	38.66 (38.44)	39.02 (38.65)
6	T ₆ = Tulsi leaf extract @ 5%	40.91 (39.76)	39.34 (38.81)	40.12 (39.29)
7	Control (Untreated)	82.64 (65.38)	78.71 (62.53)	80.67 (63.95)
	SEm±	0.67	1.01	0.60
	CD at 5%	2.06	3.11	2.10
	CV	2.85	4.43	3.70

Table.2 Effect of fungicides and botanicals on Per cent disease Control of *Aloe vera* leaf rot

S.No.	Treatments	Per cent Disease Control		
		2014	2015	Pooled
1	T ₁ = Carbendazim 50 WP @ 0.25%	59.36 (50.40)	60.28 (50.93)	59.82 (50.67)
2	T ₂ = Mancozeb 75 WP @ 0.25%	61.05 (51.39)	62.28 (52.11)	61.67 (51.75)
3	T ₃ = Propiconazole 25 EC @ 0.25%	65.20 (53.88)	65.71 (54.20)	65.46 (54.04)
4	T ₄ = Neem leaf extract @ 5%	58.26 (49.76)	59.16 (50.30)	58.71 (50.03)
5	T ₅ = Garlic bulb extract @ 5%	51.81 (46.04)	51.79 (46.03)	51.80 (46.03)
6	T ₆ = Tulsi leaf extract @ 5%	44.99 (42.12)	45.84 (42.61)	45.41 (42.37)
7	Control (Untreated)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	SEm±	0.79	1.12	0.68
	CD at 5%	2.43	3.45	2.37
	CV	3.26	4.58	3.98

Per cent disease incidence

All the treatments delayed the appearance of disease symptoms with respect to per cent disease incidence. Results presented in Table 1, indicated that all treatments were found significantly superior over control during 2014 and 2015 respectively. However, minimum disease incidence were recorded in treatment T₃= Propiconazole 25 EC @ 0.25% PDI (28.24, 26.61 and pooled 27.42) in years 2014 and 2015, followed by T₂= Mancozeb 75 WP @ 0.25% PDI (33.06, 29.26 and pooled 31.16) and T₁= Carbendazim 50 WP@ 0.25% (36.47, 36.39 and pooled 36.43). While among the botanicals T₄= Neem leaf extract @ 5% (36.19, 34.51 and pooled 35.35), followed by T₅= Garlic bulb extract @ 5% PDI (39.38, 38.66 and pooled 39.02) and T₆= Tulsi leaf extract @ 5% (40.91, 39.34 and pooled 40.12). All the treatments were found significantly superior per cent disease intensity during both the years.

Per cent disease control

Data elaborated in Table 2 revealed that in case of chemical control the maximum per cent disease control were recorded in treatment T₃ = Propiconazole 25 EC @ 0.25% PDC (65.20, 65.71 and 65.46) followed by T₂ = Mancozeb 75 WP@ 0.25% PDC (61.05, 62.28 and pooled 61.67), and T₁= Carbendazim 50 WP@ 0.25% PDC (59.36, 60.28 and pooled 59.82) during 2014 and 2015. Among the botanicals maximum per cent disease control were recorded T₄= Neem leaf extract @ 5% PDC (58.26, 59.16 and pooled 58.71) followed by T₅= Garlic bulb extract @ 5% PDC 51.81, 51.79 and pooled 51.80) and T₆= Tulsi leaf extract @ 5% PDC 44.99, 45.84 and pooled 45.41) during both the years 2014 and 2015, respectively.

The presence of *Fusarium oxysporum* in *Aloe barbadensis* plant has great implications on

the health of the rural populace that use this plant in multi-purpose herbal preparations and recipes to treat various ailments. This is because the pathogen produces mycotoxin (Trichothecene) in the infected hosts which are hazardous to human health when consumed.

On the basis of results obtained it may be concluded that even though, all the tested fungicides were found slightly more effective over botanicals tried in the present investigation to inhibit the growth of pathogens but seeking the hazardous effects of fungicides, the plant extracts with antifungal activity at given concentration viz, (Neem leaf extract) can be recommended to control the leaf rot disease in *Aloe vera* at commercial level to maintain the medicinal value of the plant.

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