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In vitro antifungal activity of some common botanicals against fungal wilt caused by *Fusarium oxysporum* of tomato (*Solanum lycopersicum* L.)

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ABSTRACT

Fusarium oxysporum f. sp. *lycopersici* is an important disease that causes wilt disease in tomato crop. Management through chemical fungicides cause environmental problems and are toxic to non-target organisms as well. Plant metabolites and plant extract appear to be one of the better alternatives as they are known to have minimal environmental impact. In an approach towards the development of eco-friendly management, *in vitro* antifungal assay was conducted against *Fusarium oxysporum* f. sp. *lycopersici* (FOL), using plant extracts of seven botanicals. Out of them, three extract proved to be potential in inhibiting the growth of the FOL viz., *Allium sativum* (77.43%), *Pongamia glabra* (73.06%), *Azadirachta indica* (72.382%). The poison food technique was employed for the evaluation of antifungal activity of the extracts at three different concentrations (10%, 15%and 20%) on mycelial growth of FOL. This study indicates that the botanical extracts could be a good alternative in developing a potent plant based fungicides which can be used in organic farming for the management of *Fusarium oxysporum* f. sp. *Lycopersici*.

Key words: *Allium sativum, Fusarium oxysporum f. sp. Lycopersici, Poison food technique, Plant extract, Tomato, Wilt disease.*

Introduction

Tomato (*Solanum lycopersicum* L.) is mostly affected by fungal diseases, which leads to cause a heavy crop loss and was the most limiting factor in tomato production areas. one of the most prevalent and damaging diseases of tomato Fusarium wilt disease caused by *Fusarium oxysporum* f. sp. *lycopersici* (Sacc.) Snyder and Hans causes economic crop losses, especially in susceptible varieties, underambient weather conditions because of its soil-borne, air-borne, and spread through plant residues mode of spreading in nature (Tabet *et al.,* 2018). The economic importance of *Fusarium oxysporum* sp. *lycopersici* originates from its voracious destructive nature towards the tomato which is the most important horticultural crop in India as well as Odisha. It is widely cultivated besides having a high nutritive value, antioxidant and curative properties, making this vegetable crop the most consumed. Affected plant shows severe wilting in case of seedling and recognisable in adult plants. Infected plant produces

wilt in older leaves turns yellow and afterwards droop. Yellowing of Leaves on one side of the plant appears and gradually most leaves turn to yellow and wilt (El-Aswad *et al.*, 2023). In Shivapratap order to protect the plants against pathogens and to prevent the plant diseases, now a day's farmers are adopting only chemical control methods for quick and easy management but most of the soil borne pathogens (i.e., *Fusarium* spp.) are difficult to control (Bawa, 2016) with chemical fungicides, the intensive application of synthetic fungicides has adverse effects on fruit, soil biodiversity, biotic environment as well as human health, as well as development of fungicide resistance in pathogens (Moutassem *et al*., 2019). With the view of the high cost of fungicides and their hazardous consequence, biodegradable and different eco-friendly material like plant extracts from different parts of the plants gained importance day by day (Moutassem *et al.,* 2019). The xylem of the host plant is colonized by fungi and as a result, blockage and breakdown of the xylem cause wilt disease symptoms such leaf withering, yellowing, and eventually plant death. Chemical fungicides are used to control *Fusarium* wilt of tomato. Unfortunately, these chemical fungicides are not readily bio degradable; tend to persist for years in the environment and few fungi have developed resistance to them (Tabet *et al.*, 2018). Phytochemicals can be used safely against soil borne diseases since they are biodegradable, non-toxic, and environmentally safe (Javaid and Rauf, 2015). Use of natural products like botanical amendments or botanical extracts for the management of fungal diseases in plants is considered as a substitute method to synthetic fungicides, due to their fewer negative impacts on the human and environment health hazard or implications. This may be used for formulating new, safer and eco-friendly fungicides. In the present work, *in vitro* screening for antifungal properties of some plant extracts against *Fusarium oxysporum*, were evaluated.

Materials and Methods

Collection of Plant Sample

Seven plant extracts from Garlic (*Allium sativum*), Tulsi (*Ocimum tenuiflorum*), Turmeric (*Curcuma longa*), Madhuca (*Madhuca longifolia*), Parthenium (*Parthenium hysterophorus*), Karanja (*Pongamia glabra*), Neem (*Azadirachta indica*) were investigated *in vitro* for their antifungal effectiveness against *Fusarium oxysporum*. For the extraction process, fresh plant components were employed. The following are the various plant parts that are used for extraction (Table 1).

Extraction of Plant extract

Before being distilled, fresh plant materials were collected and cleansed with tap water. These leaves were left out in the open to dry naturally. In a surface-sterilized pestle and mortar, 100 g of fresh material were cut and then crushed with 100 ml sterile water (1:1 w/v). After that, shake for 12 hours in a Rotary Shaker. The extracts were filtered through Whatman's no. 1 filter paper. The extracts were centrifuged at 1500 rpm for 10 minutes. The obtained supernatant was then utilised to makea stock solution.

Antifungal activity assay of botanical extracts by using poison food technique

Plant extract at different concentrations 10%, 15% and 20% from the each stock solution were added in 20 ml of sterilized potato dextrose agar in petri plates. A 5 mm diameter of the actively growing mycelium disc of the pathogen of 6–7 day old culture was placed in the center of the Petri dish. Plates without plant extract served as negative control. Plates were incubated at 27 °C. Triplicates were maintained for each treatment. Radial growth of mycelium was measured after seven days of incubation. The results were compared with negative control. The experiment was repeated thrice and mean of three readings was taken for calculations. The percent inhibition of the fungus in treatments was calculated using following formula;

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

Where, I = Percent inhibition of mycelium, C = Growth of mycelium in control, T = Growth of mycelium in treatment (Vincent's formula (1947).

Results and Discussion

Allium sativum suppressed mycelial growth the greatest at a 20% concentration, with 77.43 percent, followed by *Pongamia glabra* with a 73.06 percent inhibition, and *Azadirachta indica* 72.38 percent, whereas highest growth of mycelium was observed with control (no treatment). Moreover, *Parthenium hysterophorus*, with a 61.74 percent inhibition, was the least inhibited. The result at a concentration of

Table 1. Different plant extracts used with their common names, Scientific name and plant parts used

Treatment name	Common name	Scientific name	Plant Part used
1	Garlic	*Allium sativum*	Blub
2	Tulsi	*Ocimum tenuiflorum*	Leaf
3	Turmeric	*Curcuma longa*	Rhizome
4	Madhuca	*Madhuca longifolia*	Leaf
5	Parthenium	*Parthenium hysterophorus*	Leaf
6	Karanja	*Pongamia glabra*	Leaf
7	Neem	*Azadirachta indica*	Leaf

Table 2. *In vitro* bioassay of plant extracts

Sl. No.	Treatments	10% Concentration	15% Concentration	20% Concentration
T1	Control	33.57	35.09	36.27
T2	Garlic *(Allium sativum)*	72.86	76.60	77.43
T3	Haldi *(Curcuma longa)*	63.44	68.01	69.44
T4	Karanja *(Pongamia glabra)*	67.8	72.83	73.06
T5	Madhuca *(Madhuca longifolia)*	57.84	62.33	63.84
T6	Parthenium *(Parthenium hysterophorus)*	56.9	60.480	61.741
T7	Neem *(Azadirachta indica)*	67.55	71.987	72.382
T8	Tulsi *(Ocimum tenuiflorum)*	65.09	69.197	70.267
	C.D. (P= 0.05%)	2.18	1.221	0.960
	S. Em (±)	0.72	0.467	0.3207

20% followed the same pattern as the result at a concentration of 15%. The highest level of inhibition was found in *Allium sativum* (76.60 percent), followed by *Pongamia glabra* (72.83 percent), and *Azadirachta indica* (71.98 percent) (Table 2, Fig. 1 and 2). The current findings are supported by Wszelaki and Miller's (2005) findings, which showed that garlic extracts greatly reduced tomato leaf blight disease (Bawa, 2016) found that *Fusarium oxysporum* f. sp. *lycopersici* has the slowest mycelial growth in*In vitro*, garlic clove extracts were used in the medium (10 percent). Ajigbola and Babalola (2013) evaluated extracts of 15 plant components against *Fusarium oxysporum* isolates' spore germination and mycelial development. The extracts of *Allium sativum* were shown to be the most effective in controlling fungal development. El Shami *et al*. (1986) tested the bio efficacy of six plant items against Fusarium wilt of water melon.

Conclusion

At a 20 percent concentration, *Allium sativum* re-

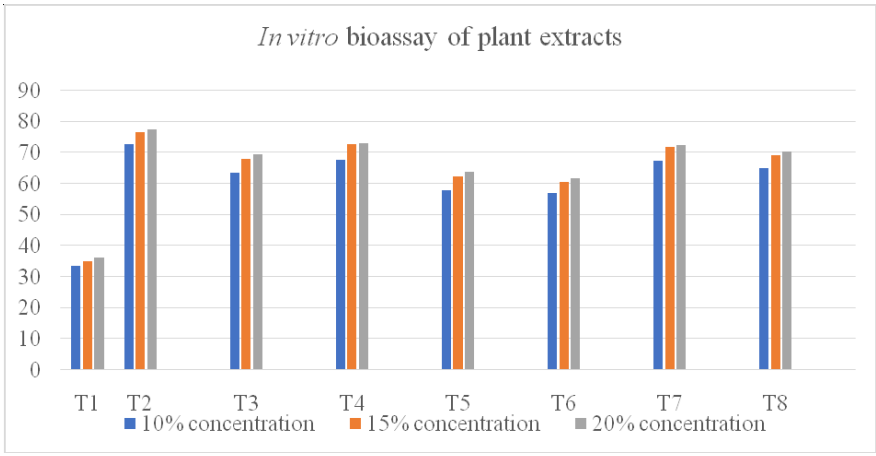

Fig. 1. *In vitro* bioassay of plant extracts

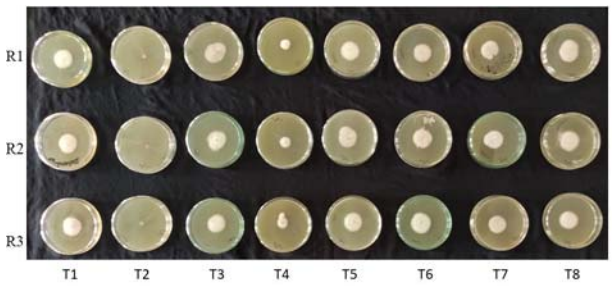

Fig. 2. Evaluation of aqueous plant extracts by poison food technique

duced mycelial development the most (77.43 percent), followed by *Pongamia glabra* (72.06 percent), and *Azadirachta indica* (72.38 percent). The least inhibited plant was *Parthenium hysterophorus*, which had a 61.74 percent inhibition. The result at a 15 percent concentration followed the same pattern as the result at a 20 percent concentration. *Allium sativum* had the highest level of inhibition (76.60 percent), followed by *Pongamia glabra* (72.83 percent), and *Azadirachta indica* (71.98 percent). *Parthenium hysterophorus* was the least inhibited, with only 60.48 percent inhibition. The presence of Allicin in *Allium sativum* may explain its effectiveness against fusarium wilt pathogen, although more research is needed. Furthermore, the effective plant products against fusarium wilt of tomato could be evaluated in the field.

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