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EFFICACY OF BOTANICALS ON THE GROWTH OF *FUSARIUM OXYSPORUM* F.SP. *CICERI*

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Crude aqueous leaf extracts of *Argemone mexicana* L., *Rauvolfia tetraphylla* L., ***Vitex negundo*** L., *Withania somnifera* (L) Dunal., extracts of fresh tea powder (before boiling) and used tea powder (after boiling) were evaluated for their antifungal efficacy on the growth of ***Fusarium oxysporum*** f. sp. *ciceri.* All the extracts inhibited the growth of test fungus. The extracts of ***R. tetraphylla,*** *V. negundo*, *W. somnifera* suppressed the mycelial growth more than that of ***A. mexicana.*** The extract of tea powder thrown as waste after use inhibited the growth totally. Higher concentration of extract showed greater inhibitory activity than the lower concentration.

Keywords : Antifungal; Extract; *Fusarium oxysporum;* Inhibition.

Introduction

Plants are rich source of wide variety of secondary metabolites such as tannins, terpenoids, alkaloids, flavonoids, phenols, essential oils, which have been found to have antimicrobial properties[1-3]. These compounds actually form the agents for defence mechanism of plants against the pathogens[3]. The presence of antifungal compounds in higher plants is an important factor to disease resistance[4]. Since the chemical fungicides used to control the fungal pathogens cause pollution in the environment and also kill beneficial organisms, attention has been diverted towards the exploration for alternate sources of ecofriendly antifungal compounds in plants. Such compounds being biodegradable and selective in their toxicity are considered valuable for controlling some plant diseases[5]. Therefore, the investigations on the antimicrobial properties in plants has attained significance to understand the therapeutic action particularly against the microbes developing resistance to the currently used antibiotics. The potential antibacterial and antifungal activities in plants were screened by several workers[6-14].

Fusarium is one of the common soil borne fungi which causes wilt disease in Chickpea, Pigeonpea, tomato, Brinjal etc. The inhibitory effect of plant extracts on the growth of pathogen *Fusarium* sp. was evaluated in recent years[15-20]. Chickpea (*Cicer arietinum*) is cultivated in large areas as an important winter pulse crop. The plants are often inflicted with wilt disease caused by *Fusarium oxysporum f. sp. ciceri* causing heavy loss in yield.

In the present work, the effect of aqueous leaf extracts of *Argemone mexicana* L., ***Rauvolfia tetraphylla*** L., *Vitex negundo* L., *Withania somnifera*(L) Dunal, extracts of fresh tea powder (Before boiling) and used tea powder (after boiling) were evaluated for their antifungal efficacy on the growth and development of *Fusarium oxysporum* f. sp. *ciceri.*, the causal organism of wilt of chickpea, isolated from the rhizosphere soil sample was investigated *in vitro.*

Material and Methods

Initially the fungus was isolated on potato dextrose agar (PDA) medium by serial dilution technique from the rhizosphere soil sample of chickpea showing the wilt symptoms. The identity of the isolated pathogen (test organism) was confirmed following characters given by Ahmed and Reddy[21]. This pathogen was then maintained as pure culture on the sterile PDA medium in culture plates as well as in slants.

Fresh and mature leaves of ***A. mexicana,*** *R. tetraphylla, V. negundo, W.somnifera* and fresh tea powder (Before use), used tea powder (thrown as waste) after making beverage were collected and dried in shade. These dried leaves were then ground in to fine powder in a blender. 100 gm leaf powder of each sample was extracted in 100 ml sterile distilled water (1:1 w / v) and filtered through muslin cloth. The filtrate was used as 100 % stock for assessing the effect on *F. oxysporum* test following food poison technique[22]. 2.5 ml and 5.0 ml from stock of each extract was added to 10 ml of the sterile PDA medium

and shaken thoroughly for uniform distribution of the extract. This amended medium was then poured in the Petriplates under aseptic conditions and kept for solidification.

A 6 mm disc of the mycelium of pathogen (inoculum) maintained as pure culture on the PDA medium in separate Petriplate was taken out with cork borer and inoculated under aseptic conditions on the amended medium in the plates. A control set without plant extract was also maintained. Each treatment was replicated four times. The inoculated plates were then incubated at 28 ± 2° C for seven days after which the diameter of the growth of colony was measured in cms and compared with that of the colony growth in the control plates. Percent inhibition of colony growth was calculated by employing Vincent's formula[23].

$$\text{Percent inhibition} = \frac{C - T}{C} \times 100$$

Results and Discussion

The observations are represented below in the table and also by photographs of the colony growth.

All the extracts tested were found to be effective in suppressing the growth of pathogen (Table 1 and Figs 1-6) which is alike to the earlier investigations[16-19, 24]. However, the percentage of inhibition was found to be more by the effect of extracts of *R. tetraphylla* (16.22 and 32.07), *V. negundo* (13.20 and 21.13) and *W. somnifera* (10.56 and 31.32) thus demonstrating the highest antifungal activity as compared to that of *A.mexicana* (3.21 and 10.56) and fresh tea powder extracts (4.90 and 10.56), which showed moderate antifungal activity. At higher concentration (5ml) all the extracts exhibited more antifungal activity than at lower (2.5 ml) concentration.

It is interesting to note that the extract of tea powder thrown as waste after preparing the beverage inhibited the growth of mycelium totally at both (2.5 ml and 5 ml) concentrations. The biochemicals in the tea powder might have acquired toxic nature after boiling. Bhale *et al.*[25] reported significant reduction in the incidence of wilt disease in *Spinach* by the plant extracts. At higher concentrations the mycelial growth was inhibited more than at lower concentrations[26].

Conclusion

The plant extracts can be safely employed as an ecofriendly and alternative biofungicides for controlling the fungal pathogens. The tea powder dumped as waste after preparing beverage can also be used as a fungicide as a process of recycling the waste. Further investigations on the isolation and characterization of antimicrobial 7.

Table 1. Effect of leaf extract on the colony growth of *Fusarium oxysporum f. sp. ciceri.*

Plant Extract	Control	Experimental concentration of extract			
		2.5 ml	% growth inhibition	5.0ml	% growth inhibition
Argemone mexicana	6.625 ± 0.150	6.412*** ± 0.063	3.21	5.925* ± 0.029	10.56
Rauvolfia tetraphylla	6.625 ± 0.150	5.550*** ± 0.264	16.22	4.500*** ± 0.216	32.07
Vitex negundo	6.625 ± 0.150	5.750*** ± 0.173	13.20	5.225** ± 0.556	21.13
Withania somnifera	6.625 ± 0.150	5.925*** ± 0.029	10.56	4.550*** ±0.081	31.32
Tea powder (Before boiling)	6.625 ± 0.150	6.300*** ± 0.182	4.90	5.925* ± 0.096	10.56
Tea powder (After boiling)	6.625 ± 0.150	00	100	00	100

All values are expressed as means of ± SD. $p < 0.05$*, $p < 0.01$** $p < 0.001$***

Fig.1. Effect of Argemone mexicana leaf extract.

Fig.2. Effect of Rawolfia tetraphylla leaf extract.

Fig.3. Effect of Vitex negundo leaf extract

Fig. 4. Effect of Withania somnifera leaf extract

Fig.5. Effect of Tea Powder (before boiling)

Fig.6. Effect of Tea Powder (After Boiling)

compounds in plant extracts will help to understand more in this regard.

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