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Antifungal activity of some essential oils against storage fungi

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Three essential oils viz. Cinnamon, Citronella and Rose of plant origin were tested *in vitro* against three predominant storage fungi viz., *Aspergillus flavus, A. niger* and *A. candidus.* All the essential oils showed antifungal activity of graded efficiency. Cinnamon oil showed maximum antifungal efficacy at the minimum level among tested oils.

Key words : Essential oils, Antifungal activity, Storage fungi

INTRODUCTION

Volatile fungitoxic compounds was reported to possess tremendous possibility as seed preservative by inhibiting growth of storage fungi *in vitro* (Nandi, 1990). As many of the natural volatile compounds exhibited either stimulatory or inhibitory effects on fungi no generalised behaviour of any particular group of compound could be observed. Nandi (1977) tested vapours of 16 aromatic esters and reported strong antifungal property of most of them. Several essential oils of plant origin were also reported to possess antimicrobial properties (Nigam and Rao, 1977 ; Gulati and Suri, 1982 ; Deshmukh *et al.*, 1986 ; Misra and Batra, 1987 ; Misra *et al.*, 1988 ; Onawunmi, 1989 ; Chaumont and Bardey, 1989 ; Farag *et al* , 1989).

MATERIALS AND METHODS

The present investigation was undertaken to study *in vitro* fungicidal efficacy of three essential oils viz., Cinnamon, Citronella and Rose of high purity procured

from Synoflor Co., Calcutta (India) against three storage fungi frequently associated with different seeds in storage viz., *Aspergillus flavus*, *A. niger*, *A. candidus*. These essential oils showed stronger antifungal property among a number of others tested against a number of fungi in an earlier screening experiment (Unpubl.).

The antifungal activity of the essential oils were evaluated in culture tubes following Frankelfeld *et al.* (1975). Fungicidal efficacy of the essential oils were also assayed by spore germination techique on glass slides following Pan and Sen (1976).

RESULTS

All the three essential oils showed antifungal activity (Table 1) in comparison to the control. The "minimum inhibitory concentration" (MIC) were recorded within the tested concentrations of the oils. Cinnamon proved to be most effective with a very low (0.1 μl/ml) MIC and showed only a little fungal growth (1.5 mg) of *A. flavus*. At higher tested concentrations it showed no fungal growth at all. The other two essential oils, Citronella and Rose oils were much less effective at the lowest tested concentration (0.5 μl/ml) and

Table 1. Minimum inhibitory concentrations (MIC) of different essential oils *in vitro* against predominant storage fungi after 96 hrs

Essential oil	Concentration (μl/ml)	Fungal growth*		
		A. flavus	*A. niger*	*A. candidus*
Control		4+ (5)	4+ (4)	4+ (3.5)
Cinnamon	0.1	2+ (1.5)	—	—
	0.25	—	—	—
	0.4	—	—	—
	0.5	—	—	—
Citronella	0.5	4+ (4.5)	—	—
	1.0	2+ (2)	—	—
	1.5	—	—	—
	2.0	—	—	—
Rose	0.5	4+ (4)	—	—
	1.0	+ (1.5)	—	—
	1.5	—	—	—
	2.0		—	—

*Mean of three replicates ; 4+ =Thick mycelial mat ; 2+ =Moderate mycelial mat
+ =Thin mycelial mat ; — =No mycelial mat ;
Number within parenthesis indicate mycelial dry weight in mg

showed higher fungal growth of *A. flavus* (4.5 & 4 mg in Citronella & Rose respectively). Citronella and Rose oils showed much higher (1 µl/ml) MIC to inhibit growth of *A. flavus*. *A. niger* and *A. candidus* showed no growth even at the lowest concentrations.

Table 2. *In vitro* fungicidal effect of essential oils against predominant storage fungi

Eessential oil	Concentration (µl ml)	Conidial germination (%)*											
		A. flavus				*A. niger*				*A. candidus*			
	Hours	24	48	72	96	24	48	72	96	24	48	72	96
Control		72	100	100	100	56	89	100	100	44	78	100	100
Cinnamon	0.1	36	50	74	92	—	—	21	40	—	—	—	—
	0.25	—	11	22	34	—	—	—	—	—	—	—	—
	0.4	—	—	—	—	—	—	—	—	—	—	—	—
	0.5	—	—	—	—	—	—	—	—	—	—	—	—
Citrenella	0.5	51	67	79	100	—	28	39	56	—	—	11	24
	1.0	32	46	68	88	—	—	—	—	—	—	—	—
	1.5	—	—	—	—	—	—	—	—	—	—	—	—
	2.0	—	—	—	—	—	—	—	—	—	—	—	—
Rose	0.5	59	76	89	100	20	36	49	62	—	—	—	19
	1.0	41	54	69	92	—	—	—	—	—	—	—	—
	1.5	—	—	—	—	—	—	—	—	—	—	—	—
	2.0	—	—	—	—	—	—	—	—	—	—	—	—

*Mean of three replicates ; '—' Indicate inhibition of germination

All the tested essential oils showed inhibition of conidial germination as well as complete inhibition of growth in comparison to control set. The essential oil showed antifungal activity by inhibiting spore germination of the test fungi from 24 hours onwards. Minimum amount of oil necessary for complete inhibition depended on the fungal species as well as on the type of oil.

Among the test fungi, *A. flavus* was found to be capable of overcoming the preservative efficacy of the essential oils tested at the lower concentrations, whereas of *A. niger* and *A. candidus* were completely checked at the lowest concentration of all the essential oils.

DISCUSSION

Inhibition of growth of the test fungi by the essential oils clearly indicated antifungal activity of the essential oils *in vitro* (cf. Misra and Batra, 1987).

However, the fungi showed certain differences in their degree of tolerance to the oils.

Cinnamon oil proved to be the most effective even at the lowest concentration whereas Citronella and Rose oils required higher concetrations to become effective against the test fungi. Lower fungitoxicity of Citronella and Rose oils might be due to binding of the test compounds with the substrates resulting in their limited penetration and inactivation as pointed out by Moleyar and Narasimhan (1989) in case of citral and camphor.

The reason for inhibition of the conidial germination by the essential oils is still obscure. Majority of the antifungal compounds were reported to act within the cell by inhibiting the vital processes (Byrde, 1965 ; Fries, 1977). Toxicity may also depend on the ability of the compound to accumulate in fungal cells to the effective levels.

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