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An Eco-friendly Approach to Control Storage Fungi

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ABSTRACT

In present study oilseeds were collected from different parts of Marathwada region and screened on different media for the incidence of different fungal species. Thirty fungal species were found to be associated with oilseeds. A study was also conducted to determine the antifungal activity of essential oils and gums of some medicinal plants against storage fungi. *Eucalyptus* oil and gum of *Terminalia arjuna*, *Acacia Arabica* and *Butea monosperma* inhibited the growth of storage fungi.

Introduction

Seed health plays an important role for successful cultivation and yield exploitation of a crop species and it plays an important role not only for successful cultivation but also for increasing yield of crop (Rajput et al., 2005). In Maharashtra state, oil seeds are cultivated in both Kharif and Rabbi seasons. Out of which groundnut (*Arachis hypogaea* L.), sunflower (*Helianthus annus* L.), safflower (*Carthamus tinctorius* L.), sesame (*Sesame indicum* L.) and soybean (*Glycin max* L.) are major oil seed crops. After harvesting seeds are stored in different storage conditions and if these storage conditions are not proper various microbes like viruses, bacteria, fungi and nematode are interacted with these seeds. Among these microbes fungi play a dominant role in decreasing quality and longetivity of the seeds. Fungi cause various abnormalities to the seeds like discolored seeds, damaged seeds, shrunken seeds, undersized seeds, rotted seeds and reduced in germinability. Fungal organisms plays significant role in infection, altering quality and longevity of seeds during the storage (Christensen and Kaufman, 1969). Such seeds are not fit for human consumption and rejected at industrial level, which ultimately affect on the yield and economy of the country. Therefore, in first part, associated storage fungi were isolated from oilseeds by using different media.

Storage fungi are commonly controlled by synthetic chemicals; however, most of the fungicides of this group create several side effects in the forms of carcinogenicity, teratogenicity, and residual toxicity (Bajaj, 1975 and Edward, 1973). Therefore, some alternative biodegradable chemical control measures should be discovered to replace synthetic pesticides for pest management without creating pesticidal pollution. Natural fungicides are free from environmental toxicity as compared to synthetic compound (Hooda and Srivastava, 1998). Natural products are less phytotoxic, easily biodegradable and more systematic (Saxena et. al., 2005). Therefore, in second part emphasis has been given on eco-friendly management of these storage fungi by essential oils and gums of some medicinal plants.

Materials and methods

Isolation of storage fungi

In this research work oil seed samples were collected from different store houses, market places, godowns, fields from different districts of Marathwada region. For detection of seed mycoflora associated with seed samples, the method recommended by ISTA (1966) was adopted. Seeds were further categorized according to their abnormalities to know the fungi responsible for their abnormal nature. Autoclaved Potato Dextrose Agar (PDA), Glucose Nitrate agar (GNA), Rose Bengal agar (RBA), Czapeck Dox Agar (CZA) media were used for isolation.

Eco-friendly management of storage fungi

Out of thirty isolated fungi ten dominant storage fungi were selected for further study. Fungitoxic property of essential oils and gum of some medicinal plants was screened against test fungi (Nene and Thapliyal, 1993). Glocose nitrate medium was prepared in flasks and sterilized. To this medium, in separate set the requisite quantity of the essential oil and in another set gums were added. The medium was then autoclaved at 15 lbs pressure for 20 minutes. After cooling the medium, fungi were inoculated in asceptic condition and incubated for 7 days at room temperature, suitable checks were kept, where the fungi were grown under the same condition on glucose nitrate without essential oils and gums. Mycelial growth and sporulation of the test fungi was measured after harvesting. The mycelial growth of the fungi compared with check, was taken as a measure of the fungal toxicity.

Results and Discussion

Incidence of fungi on different media

In order to study the effect of agar media on incidence of mycoflora, four media viz., Potato dextrose agar (PDA), Rose bengal agar (RBA), Glucose nitrate agar (GNA) and Czapek dox agar (CZA) were used and results are summerized in the table 1. It is clear from the table that, on PDA total twenty five fungi were isolated. Among these fungi, four fungi from Alternaria genus viz., *Alternaria alternata*, *A. carthami*, *A. dianthicola* and

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A. tenuissima; five fungi from genus *Aspergillus* viz., *Aspergillus flavus, A. fumigatus, A. niger, A. terreus* and *A. ustus*; seven fungi from genus *Fusarium* viz., *Fusarium chlamydosporum, F. culmorum, F. equiseti, F. moniliformi, F. oxysporum, F. solani* and *F. verticillioides* were occurred on PDA. RBA showed incidence of thirteen fungi. RBA showed qualitative and quantitative dominance of *Aspergillus* genera. *Alternaria dianthicola, A. tenuissima, Colletotrichum* sp., *Curvularia pellescens, Fusarium oxysporum, Macrophomina phaseolina, Rhizopus stolonifer, Penicillium digitatum* and *Trichoderma viride* were also detected on RBA. GNA media yielded total of twenty three fungi. Among these, *Fusarium* genera, showed quantitative dominance. *Aspergillus flavus, Curvularia lunata* and *Rhizopus stolonifer* showed their quantitive dominance. Whereas, *Macrophomina phaseolina, Cercospora kikuchii* and *Curvularia lunata* showed qualitative dominance. Only fourteen fungi were found to be associated with CZA media. Four fungi from genus *Fusarium* viz., *Fusarium chlamydosporum, F. equiseti, F. moniliformi* and *F. oxysporum*; two fungi from genus *Aspergillus* viz., *Aspergillus flavus* and *Aspergillus niger* were occurred on CZA media. Similar types of variations in mycoflora in different oilseed crops have also been reported by various workers as in case of groundnut (Reddy et al., 1991), soybean (Murthy and Raveesha, 1996), sunflower (Agarwal and Singh, 1974), safflower (Singh et al., 1987) and in sesame (Vyas et al., 1984).

Antifungal activity of essential oils

Essential oils were screened against the storage fungi for their antifungal activity. Among six essential oils, eucalyptus oil was found to be fungi toxic for the growth of *Alternaria dianthicola, Curvularia lunata, Curvularia pellescens* and *Penicillium digitatum*. Essential oils of caster, tulsi and sesame hampered the growth of *Rhizopus stolonifer*. Clove oil was found to be inhibitory for the growth of *Curvularia lunata, Curvularia pellescens, Fusarium oxysporum, Alternaria dianthicola* and *Penicillium digitatum*. Locke (1995) reported that in field *Alternaria alternata, Aspergillus niger* and *Fusarium oxysporum* has been completely controlled by using 2-10% neem oil. Somda et al., (2007) tested essential oils of *Azadirechta indica* and *Eucalyptus camaldulensis* against *Fusarium moniliforme, Phoma sorghina* and *Colletotrichum graminicola* and reported that extent of inhibition depends on the concentration of essential oils. Similarly, Oxenham et al. (2005) found that tulsi oil was inhibitory for the growth of phytopathogenic fungi as well as storage fungi.

Antifungal activity of gum of some medicinal plants

Gum of *Azadirachta indica* showed antifungal properties against *Macrophomina phaseolina, Penicillium chrysogenum* and *Rhizopus stolonifer*. Gum of *Terminalia arjuna* found to be fungitoxic for the growth of *Fusarium equiseti, Penicillium chrysogenum* and *Macrophomina phaseolina*. Gum of *Acacia arabica* inhibited the growth of *Rhizopus stolonifer, Penicillium chrysogenum* and *Curvularia pellescens. Fusarium equiseti, Rhizopus stolonifer* and *Penicillium chrysogenum* showed low growth in presence of gum of *Stercularia urens. Casina albens* found to be fungitoxic for the growth of *Alternaria dianthicola, Rhizopus stolonifer* and *Penicillium digitatum*. Marques et al. (1992) observed that cashew tree gum inhibited the growth of 10 out of 25 fungal samples, including *Aspergillus flavus, Penicillium implicatum, Colleotrichum musae* and *Verticillium* sp. Similarly, Torquato et al. (2004) tested cashew tree gum for their antimicrobial activity against bacteria, yeast and fungi. They found the only antimicrobial effect of cashew gum was against *S. cerevisiae*. On the other hand, Shailendra et al. (2008) found the antimicrobial activity of *Butea monosperma* against *Staphylococcus aureus, Bacillus subtilis, Bacillus cereus, Salmonella typhimurium, Pseudomonas aeuriogenosa, Escherichia coli, Candida albicans* and *Saccharomyces cerevisiae*.

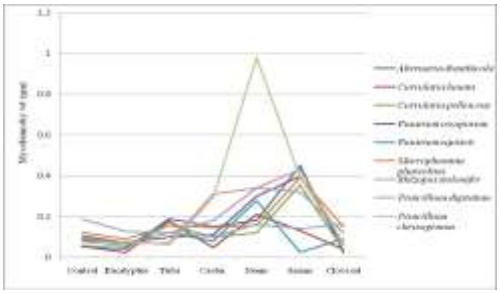

Fig 1: Antifungal properties of essential oils

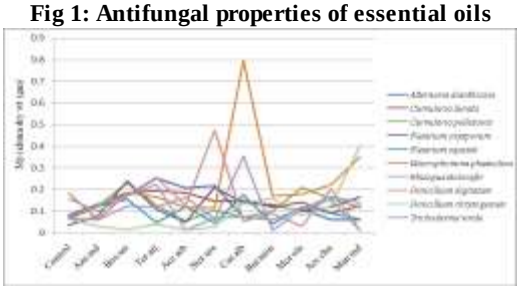

Fig 2: Antifungal properties of gums

References

[1]Agarwal, V. K. and Singh, O. V. (1974). Fungi associated with sunflower seeds. Indian Phytopath. 27(2): 240-241.

[2]Bajaj, B. S. and Ghosh, A. K. (1975). Antifungal antibiotics in perspective, p. 297-311. In S. P. Raychaudhuri, A. Verma, K. S. Bhargava, and B. S. Mehrotra (ed.), Advances in Mycology and Plant Pathology. Sagar Printers, New Delhi, India.

[3]Christensen, C. M. and Kaufman, H. H. (1969). Grain storage. The role of fungi in quality losses. Univ. Minnesota, Press Minneapolis.

[4]Edwards, C. A. (1973). Environmental pollution by pesticides, p. 1-9. Plenum Press, London.

[5]Hooda, K.S. and Srivastava, M. P. (1998). Biochemical response of scented rice as influenced by fungitoxicant and neem products in relation to rice blast. Indian J. Pl. Pathol. 16: 64-66.

[6]ISTA (1996). Seed Science and Technology 21(Suppl.): 1B288.

[7]Locke, J. E. (1995). Fungi. In: The Neem Tree, source of Unique National Products for Integrated pest Management, Medicine, Industry and Other proposes. (Ed.): H. Schmutterer. V.C.H, Weinheim, Germany. p 118-125.

[8]Marques, M. R., Albuquerque, L. M. B. and Xavier-Filho, J. (1992). Antimicrobial and insecticidal activities of cashew tree gum exudate. Annals of Appl. Biol. 121: 371–377.

[9]Murthy, Y. L. K. and Raveesha, K. A. (1996). Seed mycoflora of soybean from Karnataka. Pl. Dis. Res. 11(1): 78-82.

[10]Nene, Y. L. and Thapliyal, P. N. (1993). Fungicides in plant diseases control. Oxford and IPH. Publishing Co. Pvt. Ltd., New Delhi pp.531.

[11]Oxenham, S. K., Svoboda, K. P. and Walters, D. R. (2005). Antifungal activity of the essential oil of basil (*Ocimum basilicum*). J. Phytopath. 153: 174–180.

[12]Rajput, M. Aslam, Mumtaz. A. Pathan, A. Mubeen Lodhi, G. Sarwar Shah and Khalil A. Khanzada (2005). Studies on seed-borne fungi of wheat in Sindh Province and their effect on seed germination. Pak. J. Bot., 37(1): 181-185.

[13]Reddy, G. R., Reddy, A. G. R. and Rao, K. C. (1991). Seed mycoflora of groundnut varieties collected from different sources. J. Res. APAU. 19(2): 82-83.

[14]Saxena, A. R., Sahni, R. K.,Yadav, H. L., Upadhyay, S. K. and Saxena, M. (2005). Antifungal activity of some higher plants against *Fusarium oxysporum* f.sp. pisi. J. liv. World. 12: 32-39.

[15]Gurav, S. S., Gulkari, V. D., Duragkar, N. J. and Patil, A. T. (2008). Antimicrobial activity of Butea monosperma Lam. Iranian Journal of Pharmacology and Therapeutics 7(1)-21-24.

[16]Singh, S.N., Agarwal, S.C. and Khare, M.N. (1987). Seed-borne mycoflora of safflower, their significance and control. Seed Res. 15(2): 190-194.

[17]Somda, I., Leth, V. and Sereme, P. (2007). Antifungal effect of *Cymbopogon citrates, Eucalyptus camaldulensis* and *Azadirechta indica*. Asian J. Plant Sci. 6(8): 1182-1189.

[18]Torquato, D.S., Ferreira, M.L., Sa´, G.C., Brito, E.S., Pinto, G.A.S. and Azevedo, E.H.F. (2004). Evaluation of antimicrobial activity of cashew tree gum. World J. Microbiol. and Biotech. 20: 505–507.

[19]Vyas, S.C., Kotpal, I., Prasad, K.V.V. and Jain, A.C. (1984). Note on seed-borne fungi of sesame and their control. Seed Res. 12(1): 93-94.

Table 1: Percent incidence of fungi on different media

Fungi	PDA					RBA					GNA					CZA				
	Gn	So	Se	Sf	Sn	Gn	So	Se	Sf	Sn	Gn	So	Se	Sf	Sn	Gn	So	Se	Sf	Sn
Alt alt	30	10	--	--	--	--	--	--	--	--	10	--	--	--	--	--	--	--	--	--
Alt dia	--	20	10	--	--	--	10	--	--	--	--	10	40	--	--	--	20	30	--	--
Alt car	--	--	--	10	--	--	--	--	--	--	--	--	--	20	--	--	--	--	--	--
Alt ten	--	--	--	--	--	--	10	--	--	--	--	--	--	--	--	--	--	--	--	--
Asp fla	40	--	30	20	10	50	--	70	--	--	30	40	30	--	--	--	20	--	--	--
Asp fum	--	20	--	--	--	--	10	--	--	--	30	--	--	10	--	--	--	--	--	--
Asp nig	30	10	--	--	--	20	20	--	---	30	40	--	--	--	10	30	40	20	10	--
Asp ter	20	--	--	10	30	--	--	20	--	--	--	--	--	--	--	--	--	--	--	--
Asp ust	--	--	--	10	--	--	--	--	--	--	--	--	10	--	--	--	--	--	--	--
Cer kek	--	30	--	--	--	--	--	--	--	--	--	40	--	--	--	--	--	--	--	--
Cur lun	30	--	20	--	--	--	--	--	--	--	--	40	--	10	20	--	30	--	--	--
Cur pel	--	20	--	10	--	--	--	--	30	--	--	--	20	--	--	--	--	--	10	--
Cole glo	--	10	---	--	--	--	20	--	--	--	--	--	--	--	--	--	--	--	--	--
Fus chl	--	--	--	30	--	--	--	--	--	--	--	--	--	--	--	--	20	30	--	--
Fus cul	--	20	--	--	--	--	--	--	--	--	--	10	--	--	--	--	--	--	--	--
Fus equ	20	--	10	10	--	--	--	--	--	--	--	--	--	--	--	--	--	30	--	--
Fus mon	--	--	--	--	--	--	--	--	--	--	--	--	20	10	--	--	--	--	--	--
Fus oxy	--	50	10	30	--	--	20	10	--	--	--	10	--	--	50	--	--	30	--	50
Fus sol	--	--	--	--	--	--	--	--	--	--	--	30	--	--	--	--	--	--	--	--
Fus ver	--	10	--	--	--	--	--	--	--	--	--	10	--	--	--	--	--	--	--	--
Hel pop	--	--	--	--	--	--	--	--	--	--	--	20	--	--	--	--	--	--	--	--
Mac pha	30	50	--	20	--	10	30	--	--	--	10	60	--	--	--	--	30	--	--	--
Muc ind	10	--	--	--	30	--	--	--	--	--	20	--	--	--	--	--	--	--	20	20
Rhi sol	--	20	--	--	--	--	--	--	--	--	30	40	--	--	--	--	--	--	--	--
Rhi sto	50	--	--	30	--	30	--	--	20	--	40	--	10	--	50	60	--	--	--	30
Pen chr	30	--	20	--	--	--	--	--	--	--	10	30	--	--	--	--	--	--	--	60
Pen dig	20	--	--	20	--	10	--	--	--	--	30	--	--	--	--	10	--	30	--	--
Tri vir	20	--	10	--	--	--	--	30	--	--	--	--	20	--	--	30	--	--	20	--
Tri har	--	--	--	--	20	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
Ver ten	--	--	--	--	--	--	--	--	--	--	--	--	20	--	--	--	--	--	--	--

Gn- groundnut; So- soybean; Se- seAsfe; Sf- safflower; Sn- sunflower

Alt alt- Alternaria alternata; Alt car - Alternaria carthami; Alt dia- Alternaria dianthicola; Alt ten - Alternaria tenuissima; Asp fla - Aspergillus flavus; Asp fum - Aspergillus fumigatus; Asp nig- Aspergillus niger; Asp ter - Aspergillus terreus; Asp ust - Aspergillus ustus; Cer kek - Cercospora kikuchii; Cur lun - Curvularia lunata; Cur pel - Curvularia pellescens; Cole glo- Colletotrichum gloeosporioides; Fus chl- Fusarium chlamydosporum; Fus cul- F. culmorum; Fus equ - Fusarium equiseti; Fus mon - Fusarium moniliformi;Fus oxy - Fusarium oxysporum; Fus sol - Fusarium solani; Fus ver - Fusarium verticillioides; Hel pop - Helminthosporium papulosum; Mac pha - Macrophomina phaseolina; Muc ind- Mucor indicus; Rhi sol- Rhizoctonia solani; Rhi sto - Rhizopus stolonifer; Pen dig - Penicillium digitatum; Pen chr - Penicillium chrysogenum; Tri vir - Trichoderma viride; Tri har- Trichoderma harzianum; Ver ten - Verticillium tenerum

Table 2: Antifungal properties of essential oils

Fungi	Essential oils						
	Control	Eucalyptus	Tulsi	Caster	Neem	Sesame	Clove oil
Alternaria dianthicola	0.053	0.044	0.189	0.150	0.181	0.450	0.023
Curvularia lunata	0.055	0.024	0.178	0.048	0.211	0.131	0.038
Curvularia pellescens	0.083	0.056	0.158	0.100	0.121	0.354	0.038
Fusarium oxysporum	0.107	0.073	0.100	0.111	0.300	0.398	0.064
Fusarium equiseti	0.086	0.071	0.121	0.080	0.279	.0256	0.092
Macrophomina phaseolina	0.123	0.089	0.156	0.146	0.156	0.390	0.150
Rhizopus stolonifer	0.187	0.126	0.120	0.101	0.162	0.142	0.162
Penicillium digitatum	0.092	0.067	0.070	0.311	0.340	0.432	0.068
Penicillium chrysogenum	0.065	0.078	0.060	0.290	0.980	0.392	0.07
Trichoderma viride	0.099	0.067	0.121	0.181	0.341	0.322	0.121

Table 3: Antifungal properties of gums

Fungi	Plant gums										
	Control	*Aza ind*	*Bos ser*	*Ter arj*	*Acc arb*	*Ster ure*	*Cas alb*	*But mon*	*Mor ole*	*Acc chu*	*Man ind*
Alternaria dianthicola	0.083	0.133	0.170	0.253	0.207	0.217	0.070	0.060	0.110	0.060	0.060
Curvularia lunata	0.071	0.128	0.185	0.193	0.185	0.144	0.150	0.120	0.140	0.090	0.124
Curvularia pellescens	0.063	0.125	0.232	0.128	0.055	0.105	0.072	0.100	0.210	0.160	0.119
Fusarium oxysporum	0.037	0.079	0.239	0.109	0.050	0.210	0.136	0.130	0.100	0.120	0.165
Fusarium equiseti	0.076	0.124	0.154	0.046	0.122	0.044	0.176	0.040	0.120	0.091	0.063
Macrophomina phaseolina	0.183	0.061	0.186	0.167	0.118	0.116	0.796	0.170	0.180	0.222	0.353
Rhizopus stolonifer	0.157	0.101	0.170	0.224	0.014	0.069	0.073	0.060	0.110	0.153	0.011
Penicillium digitatum	0.072	0.128	0.175	0.258	0.127	0.474	0.054	0.090	0.030	0.204	0.011
Penicillium chrysogenum	0.065	0.030	0.016	0.041	0.013	0.029	0.139	0.160	0.080	0.125	0.403
Trichoderma viride	0.066	0.060	0.124	0.119	0.165	0.063	0.353	0.010	0.110	0.167	0.138

Aza ind- Azadirachta indica; Bos ser- Boswellia sierata; Ter arj- Terminalia arjuna; Acc arb- Acacia arabica; Ster ure- Stercularia urens; Cas alb- Casina albens; But mon- Butea monosperma; Mor ole- Moringa oleifera; Acc chu- Acacia chundra; Man ind- Magnifera indica