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Antifungal Activity of *Euphorbia fusiformis.* A Redlisted Medicinal Plant

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Euphorbia fusiformis **is one of the redlisted category medicinal plant, used to cure various illnesses. The present investigation was focuses the antifungal activity of various solvent extracts (Benzne, Xylene, Tolene, Isopropyl alcohol, Diethyl ether, Dimethyl formamide and Butanol) from the leaf of *Euphorbia fusiformis* were tested against 4 aflatoxin producing fungi (*Aspergillus flavus, A. niger, A. fumigatus, A. erythrocephalus*) and one other mold *i.e. Fusarium* sp. by conducting disc diffusion assay. Among the different solvents, isopropyl alcohol and xylene extracts of this plant expressed significant activity compared with other solvents. *Fusarium* sp. had found more sensitive than other organisms. The result of present investigation is suggest the plant may be used in future, to treat diseases caused by selected fungal pathogens.**

Key words: *Euphorbia fusiformis*, Antifungal activity, Solvent extracts, Aflatoxin producing fungi, *Fusarium* sp.

Euphorbiaceae is one of largest family in angiosperms and it consists of about 300 genera and 6000 or more species. Most of them are trees or shrubs and few are herbs. The biological activity of *Euphorbia* species have been reported to possess antitumor activity and used as anticancer remedies (Ahmed *et al.,* 1988; Duarte *et al.,* 2006). The biological activities of *Euphorbia* species reported as carcinogens (Jury *et al.,* 1987; Vogg *et al.,* 1999), antimicrobial (Cateni *et al.,* 2003; Natarajan *et al.,* 2005; 2007; Ramachandran *et al.,* 2008), antimalarial (Spencer *et al.,* 1947), insecticidal (Heal *et al.,* 1950), mollusicidal (Singh and Agarwal, 1988; Mendes *et al.,* 1997) anti-inflammatory and antipyretic property (Parmar *et al.,* 1989).

One such plant *Euphorbia fusiformis* is an IUCN redlisted medicinal herb (FRLHT, 2002; Britto *et al.,* 2002; Natarajan *et al.,* 2004) found in the Eastern Ghats of India *viz*. Andhra Pradesh, Orissa and Tamil Nadu. The medical point of view, rootstock and whole plant are used treatment of rheumatism, gout, arthritis, paralysis (Prakash and Singh, 2001) and against inflammation action (Singh *et al.,* 1984; Pullaiah, 2002). The fresh

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latex and dried powder of rhizome is used for increasing lactation in human beings (Raju *et al*., 2004) as well as cattles (Mitchell *et al.,* 2003). The biochemical nature of the plant showed diterpenes (Suri *et al.,* 1990), ellagic glycoside (Suri *et al.,* 1988) and euphol (Pullaiah, 2002). The antimicrobial activity of this plant was least explored *i.e.* antibacterial (Natarajan *et al.,* 2005; Ramachandran *et al.,* 2008) and anticandidial and anticrytocococcal activity (Natarajan *et al.,* 2007). Hence, this attempt was first time reports to screen the antifungal property of various solvent extracts from the leaf of *E. fusiformis* were tested against four aflatoxin producing filamentous fungi (*Aspergillus flavus, A. niger, A. fumigatus, A. erythrocephalus*) and a *Fusarium* sp. by performing disc diffusion method.

MATERIAL AND METHODS

Plant materials and Extracts Preparation

The fresh and healthy leaves of *E. fusiformis* were collected from Chitteri hills, Eastern Ghats, Tamil Nadu. The collected leaves were taken into the laboratory and shade dried for 10-15 days. Then, dried leaves were powdered by using electric mixer/grinder. The dried powdered leaves were weighed (25gms) and dissolved in 100ml of respective/selected solvents for a week. The samples were stirred by using rotary shaker for complete extraction of plant crude biomolecules. The crude extracts were vaccum-dried and stored at refrigerator for further use. The paste of the extract was dissolved in Dimethyl Sulfoxide (DMSO) solution.

Fungal cultures

The test fungal cultures were procured from Institute of Microbial Type culture Collection (MTCC, IMTECH, Chandigarh, India). All the fungal pathogens were subcultured and stored at appropriate temperature. The fungal inoculum (two-four days young culture) was sub-cultured in the Sabouraud Dextrose broth (Himedia, India) medium into sterile test tubes. Test fungi were inoculated and incubated under room temperature. The fungal broth cultures (spore suspension) are stored for screening antifungal activity.

In vitro screening for antifungal activity

The antifungal activity of different solvent extracts form the leaf of *E. fusiformis* was tested against selected fungal strains by performing disc diffusion method. The Sabouraud Dextrose Agar (SDA) medium was prepared and sterilized along with several covered petriplates (autoclaved at 5000rpm/15 minutes). The medium was poured aseptically into each sterile petridish. The fungal broth cultures were seeded over the medium using sterile glass L-rod. The plant extract (0.5ml) was aseptically added into the sterile discs.The discs were placed on the top of each petridish and incubated at room temperature (27^0C). Then, the diameter of inhibition zone of each disc was measured and noted. The standard antifungal agent (Clotrimazole) and respective solvents are used as positive and negative control respectively. Each extract was tested in triplicate to calculate standard deviation (Gupta, 1977).

RESULTS AND DISCUSSION

The results of organic solvent extracts from the leaf of *E. fusiformis* were tested against four important aflatoxin producing fungi namely A*spergillus flavus, A. niger, A. fumigatus, A. erythrocephalus* and *Fusarium* sp. (Table 1). The benzene, toluene, diethyl ether extracts have found inactive against tested pathogens except *Fusarium* sp. The xylene extracts was expressed broad spectrum of antifungal activity. The maximum growth inhibitory effect was observed in *Fusarium* sp. followed by *A. fumigatus, A. erythrocephalus* and least activity was noted in *A. niger* and *A. flavus*. The isopropyl alcoholic extracts from the leaf showed least to moderate activity against selected fungi. On the other hand, the dimethyl formamide and butanol extracts contribute moderate activity against most of the pathogens except *A. niger*. The present research was correlated with other findings in related to the genera *Euphorbia*, such as the crude ethanol and water extract of leaves, flowers and bark from *Euphorbia hirta* were tested against three fungi including *Aspergillus fumigatus* (Somchit *et al.,* 2001). Similarly, Murugan *et al.* (2001) reported different concentration of crude drug extracts (5, 10, 15, 20, 25, 30 and 35 mg/ml) from the dry flower powder of *Euphorbia milli* and *Euphorbia pulcherrima* and *Euphoria splendons* (Abubacker & Ramanathan, 2003) were tested against the growth of two aflatoxin producing toxigenic

Table 1. Antifungal activity of different solvents extracts from the leaf of *Euphorbia fusiformis* - A redlisted medicinal plant

S. No.	Solvents Used	Diameter of growth inhibition zone (in mm)				
		A. niger	*A. flavus*	*A. fumigatus*	*A. erythrocephalus*	*Fusarium* sp.
1.	Benzene	-	-	-	-	9.00±0.00
2.	Xylene	9.00±0.00	7.33±0.47	17.66±0.46	10.00±0.00	20.00±0.81
3.	Toluene	-	-	-		9.33±0.47
4.	Isopropyl alcohol	9.66±0.46	10.00±0.00	10.33±0.47	8.00±0.00	10.66±0.46
5.	Diethyl ether	-	-	-	-	10.00±0.00
6.	Dimethyl formamide	-	9.66±0.00	7.00±0.00	10.33±0.47	9.00±0.00
7.	Butanol	-	10.66±0.46	10.33±0.47	8.00±0.00	8.00±0.00
8.	Clotrimazole (antifungal agent)	10.00±0.00	12.66±0.46	15.00±0.00	13.33±0.47	20.00±0.00

- = No Activity

fungal strains namely *Aspergillus flavus* and *Aspergillus parasiticus.* These study highlights different solvent extractions of herbals have found to be promising antimicrobial activity and this is agreed in worldwide (Faizi *et al.,* 2002; Wanjala *et al.,* 2003; Vukovic *et al.,* 2007; Vukovic *et al.,* 2008). Further more attention of this plant, will be needed for the isolation, identification of antimicrobial/antifungal compounds, molecular level, clinical studies (animal and human) are necessary to elucidate antimicrobial/biological activity of the rare taxon in future.

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REFERENCES

1. References and further reading may be available for this article. To view references and further reading you must purchase this article.
2. Abubacker, M.N., Ramanathan, R. Efficacy of *Euphoria splendons* and *Leonotis nepetaefolia* on aflatoxin producing fungi *Aspergillus flavus* and *Aspergillus parasiticus*. *Indian J. Experimental Biol.,* 2003; **41**: 1473-1475.
3. Ahmed W., Nazir M., Khan, S.A. The chemical composition of various *Euphorbia* species for industrial applications. II: Neutral lipids of *Euphorbia cauducifolia*. *J. Scientific and Indus. Res.,* 1988; **31**: 85-89.
4. Bindra, R.S., Satti N.K,, Suri, O.P. Isolation and structures of ellagic acid derivatives from *Euphorbia aqualis*. *Phytochem.,* 1988; **27**: 2313-2315.
5. Biradar, Y.S., Jagatap, S., Khandewal, K.R., Shinghania, S.S. Exploring of antimicrobial activity of Triphala mashi - an Ayurvedic formulation. *Evid. Based Altern. Med.*, 2008; **5**:107-113.
6. Britto, S.J, Soosairaj, S., Natarajan, D., Nagamurugan N., Ravipaul, S. *Euphorbia fusiformis* Buch.Ham. Ex. D. Don (Euphorbiaceae): a new record for Tamil Nadu. *Journal Econ. and Taxon. Bot.,* 2002; **26**: 467-471.
7. Cateni, F.,. Zilic, J., Falsone, G., Scialino G., Banfi, E. New cerebrosides from *Euphorbia peplis* L.: antimicrobial activity evaluation. *Bioorganic and Med Chem. Let.,* 2003; **13**: 4345-4350.
8. Duarte, N., Gyémánt, N., Abreu, P.M., Molnár J., Ferreira, M.J.U. New macrocyclic lathyrane diterpenes, from *Euphorbia lagascae*, as inhibitors of multidrug resistance of tumour cells. *Planta Med*., 2006; **72**: 162-168.
9. Faizi, S., Khan, R.A., Azher, S., Khan, S.A., Tauseef S., Ahmad, A. New antimicrobial alkaloids from the roots of *Polyalthia longifolia* var. *pendula*. *Planta Med.,* 2003; **69**: 350-355.
10. Foundation for Revitalisation of Local and Health Traditions (FRLHT)., *Medicinal plant species of Andhrapradesh community, Forestry Project Area. Including a tabulation of redlisted*

species (with presence in the three agro-climatic zones), Andhrapradesh forest department, Andhrapradesh, India 2002.

11. Gupta., *Statistical Methods.* S. Chand and Co, New Delhi 1977.
12. Heal, R.E., Rogers, E.F., Wallace, R.I., Starnes, O. A survey of plants for insecticidal activity. *Lloyd*., 1950; **13**: 89-162.
13. Jury, S.L., Reynolds, T., Culter, D.F., Evans, F.J., *The Euphorbials-chemistry, taxonomy and economic botany*. Academic Press, Orlando, Florida 1987.
14. Mendes, N.M., Vasconcello, S.M.C., Baptista, D.F., Rocha, R.S., Schall, V.T. Evaluation of the molluscicidal properties of *Euphorbia splendens* var. *hislopii* (N.E.B.) latex: Experimental test in an endemic area in the State of Minas Gerais. *Brazil. Mem. Inst. Oswaldo Cruz.,* 1997. **92**: 719-724.
15. Mitchell, C.P., Corbridge, S.E., Jewitt, S.L., Mahapatra, A.K., Kumar, S., In; *NTFP (Non-Timber Forest Products); Availability, Production, Consumption, Management & Marketing in Eastern India*. Project Report, Funded by International Development of the United Kingdom (UK), London 2003.
16. Murugan, S., Anand, R., Uma Devi, P., Vidhya, N., Rajesh, K.A. Efficacy of *Euphorbia milli* and *Euphorbia pulcherrima* on aflatoxin producing fungi (*Aspergillus flavus* and *Aspergillus parasiticus*). *African J Biotechnol.,* 2007; **6**: 718-719.
17. Natarajan, D., Britto, S.J., Balaguru, B., Nagamurugan, N., Soosairaj, S., Arockiasamy, D.I. Identification of conservation priority sites using remote sensing and GIS – A case study from Chitteri Hills, Eastern Ghats, Tamil Nadu. *Curr. Sci.,* 2004; **86**: 1316-1323.
18. Natarajan, D., Britto, S.J., Srinivasan, K., Nagamurugan, N., Mohanasundari, C., Perumal, G. Anti-bacterial activity of *Euphorbia fusiformis* - A rare medicinal herb. *J. Ethnopharmacol.,* 2005; **102**: 123-126.
19. Natarajan, D., Nagamurugan, N., Ramachandran, A., Mohanasundari, C., Srinivasan, K. Anticandidial and anticryptococcal activity of *Euphorbia fusiformis*, a rare medicinal plant. *World J. Microbiol. and Biotechnol*., 2007; **23**: 719-721.
20. Parmar, N.S., Mossa, J.S., Al-Yahya, M.A., Al-Said, M.S., Tariq, M., Ageel, M.A. Anti-Inflammatory and antipyretic activity of *Euphorbia Peplis* L. *International J. Crude Drug Res.,* 1989; **27**: 9-13.
21. Prakash, A, Singh, K.K.Use of medicinal plants by certain tribal people in North India. *J. Trop. Med. Plants.,* 2001; **2**: 225-229.
22. Pullaiah, T., *Medicinal plants in India*. Vol. 1. Regency publications, New Delhi, India 2002.
23. Raju, K., Natarajan, S., Karuppusamy, S., Manian, S. New ethnobotanical report on *Euphorbia fusiformis* form Chitteri hills of Tamil Nadu, India. *J Swamy Bot. Club.*, 2004; 21:79-80.
24. Ramachandran, A., Natarajan, D., Mohanasundari, C., Srinivasan, K., Soosairaj, S., Natarajan, E. Screening for antibacterial activity of combined extracts of *Euphorbia fusiformis* from Tamil Nadu, India: a new application. *J. Biological Res*., 2008; **9**: 97- 102.
25. Singh, A., Agarwal, R.A. Possibility of using latex of euphorbiales for snail control. *The Science of the Total Environment*., 1988; **77**: 231-236.
26. Singh, G.B., Kaur, S., Satti, N.K., Atal, C.K., Maheswari, J.K. Anti-inflammatory activity of *Euphorbia aqualis* Roxb. *J. Ethnopharmacol.,* 1984; **10**: 225-233.
27. Somchit, M.N., Mutalib, A.R., Hasmawie, R., Murni, M. A *In Vitro* antifungal and antibacterial properties of *Euphorbia hirta. J. Trop. Med. Plants*., 2001; **2**: 179-182 .
28. Spencer, G.F., Koniuszy, F.R., Rogers, E.F. Survey of plants for antimalarial activity. *Lloyd.* 1947;**10**: 145-174.
29. Suri, O.P., Satti, N.K., Suri, K.A., Dhar, K.L. Ent-Seco-atisane skeleton-diterpene from a natural resource rhizome of *Euphorbia aqualis*. *J. Natural Prod*., 1990; **53**: 470-473.
30. Vogg, G., Mattes, E., Rothenburger, J., Hertkorn, N., Achatz Jr, S., Sandermann H. Tumor promoting diterpenes from *Euphorbia leuconeura. Phytochem.* 1999; **51**: 289-295.
31. Vukovic, N., Milosevic, T., Sudolak, S., Solujic, S. Antimicrobial activities of essential oil and methanol extract of *Teucrium montanum*. Evid. *Based Altern.Med.*, 2007; **4**:17-20.
32. Wanjala, C.C., Juma, B.F., Bojas, G., Gashe, B.A., Majnda, R.R. Erythrinaline alkaloids and antimicrobial flavonoids from *Erythrina latissima*. *Planta Med.,* 2002; **68**: 640-642.