

Antifungal activity of *Commiphora wightii*, an important medicinal plant

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ABSTRACT

Commiphora wightii (Arnott.) Bhandari is an endangered, slow growing medicinal tree. The present investigations are being carried out to evaluate the antifungal medicinal properties of *Carica papaya*. The effects of different concentrations of alcoholic extract of *Commiphora wightii* (root, shoot and seed) on the radial growth of plant against the pathogenic fungi viz. *Aspergillus niger*, *Aspergillus flavus*, *Candida albicans* and *Microsporum fulvum*. That with the increase in concentrations the rate of growth inhibition also increases. Observation further shows that like root extract growth is also inhibited in the presence of shoot and seed alcoholic extract under culture medium. Further shows that the growth of these fungi inhibits more in presence of higher concentrations as compared to lower concentrations of extract.

Keywords: *Commiphora wightii*, *Aspergillus flavus*, *Candida Albicans*, *Calatropis procera*, *Microsporum fulvum*

INTRODUCTION

Commiphora wightii (Arnott.) Bhandari is an important medicinal plant of herbal heritage of India. *Commiphora wightii* belongs to family Burseraceae. Unfortunately the plant *Commiphora wightii* has become endangered because of its slow growing nature, poor seed setting, [1] lack of cultivation, poor seed germination rate [2] and excessive and unscientific tapping for its gum resin by the pharmaceutical industries and religious prophets. This plant is incorporated in Data Deficient category of IUCN [3] Red Data list. *C. wightii* occurs in Rajasthan, Gujarat, Maharashtra, Madhya Pradesh and Karnataka states of India. Many of the species produce resins of commercial importance. About five species occur in India of which *C. wightii* (Arnott) Bhandari and *C. roxburghii* yield guggul, an oleoresin gum [4]. It yields guggul, an important oleogumresin used as incense, fixative in perfumery and in Ayurvedic medicine. Its antiarthritic, hypocholesterolaemic and hypolipidaemic properties have been established [5]. It is mainly used to treat diseases like atherosclerosis, leprosy, pneumonia, rheumatism etc. The gum resin also has aphrodisiac, diuretic and immunostimulant properties.

MATERIALS AND METHODS

Commiphora wightii were collected from herbal garden of Singhania University, Pacheri Bari, Jhunjhunu, Rajasthan. The method was followed based on work of [6] with some modifications.

For the preparation of *Commiphora wightii* plant extracts 5 gm of fresh plant part was washed 2-3 times with distilled water & then surface sterilized with 90% alcohol. Subsequently the plant material was grounded in 50 ml of distilled water & alcohol separately for aqueous and alcoholic extracts, respectively. The alcoholic extracts were kept for 24 hrs. at room temperature to evaporate the alcohol. In the remaining residue, 50 ml of distilled water added. The macerates were squeezed through double layered Muslin cloth & filtered through filter paper. After filtration, the aliquot was centrifuged at 5000 rpm for 30 minute. The supernatants were filtered through whatmann no. 1 filter paper & then sterilized by passing through 0.2 micron disposable filters. The various concentrations of extract made & thus obtained were used in studies. initially treated with 0.1% HgCl₂ solution for sterilization and

subsequently washed thoroughly with sterile distilled water and grounded in mortar & pestle with 50% methanol. The homogenized liquid was filtered and centrifuged at 3000 to 5000 rpm. The supernatant was used as test extract & made up into 20 ml using 50% methanol. Further, the extract was diluted into different concentrations, i.e. 10%, 25%, 50%, 75%, 20 ml of SDA (Sabouraud Dextro Agar) culture medium with 5 ml of the above concentration of the extract was poured in sterile petriplates and allowed to solidify. Then the test fungus was inoculated at the centre of the medium and incubated at room temperature $25^{\circ}\text{C} \pm 2^{\circ}\text{C}$. Replicates and controls were maintained throughout the study. The diameter of the fungal growth was measured on 5th and 7th day.

To study the effects of antifungal alcoholic extract and aqueous extract of above selected plants two sets of culture media were prepared separately for control and treatment. In the test sets of neutral pH 7, requisite amount of the experimental material were mixed and then added into the sterilized Sabouraud dextrose agar (SDA) medium of respective pH level. In the control set of each experimental set, the same volume of distilled water (in place of experimental material) was mixed in appropriate amounts whenever found necessary.

Mycelial discs of 5 mm diameter, were cut from the periphery of 7 day old culture of the test organisms were aseptically inoculated upside down on the surface of the SDA medium in petri plates. Inoculated petri plates were incubated at $25^{\circ}\text{C} \pm 2^{\circ}\text{C}$ and observation were recorded at 5th and 7th day. Fungal growth taken as measurement parameter. The absence of Fungi denoted antifungal property of fungicidal nature.

Percentage of mycelial growth inhibition on different pH levels were calculated using following equation:

$$\% \text{ growth inhibition} = \frac{\text{Colony diameter in control} - \text{colony diameter in treated sets} \times 100}{\text{Colony diameter in control}}$$

$$I = (C - t) \times 100$$

Test fungi isolated and were used for in vitro studies. The culture was purified by hyphal tip technique. The stock culture of the test fungus was maintained on SDA medium at $25^{\circ}\text{C} \pm 1^{\circ}\text{C}$.

Effect of different concentration of aqueous and alcoholic extract against the fungus was studied employing techniques.

Poisoned food technique. (Nene and Thapliyal, 1979) Poisoned food technique used to assess the antifungal activity of selected plant extracts. A series of double strength of test plant extracts viz. 10%, 25%, 50% and 75% were prepared using sterile distilled water. 30 ml of test extract was poured into 100 ml conical flask containing 30 ml sterilized melted SDA of double concentration. 30 ml of this mixed medium was then poured in each petriplate aseptically. The petriplates were inoculated with previously maintained 7 days old culture. 5 ml mycelial disc was cut with sterilized cork borer and transferred aseptically in the centre in inverted position. All petriplate including control and experimental were incubated at $25^{\circ}\text{C} \pm 1^{\circ}\text{C}$ for 7 days. After 7 days of incubation, observation were recorded and percent inhibition of radial growth was calculated using following formula:

$$I = \frac{C - T}{C} \times 100$$

where, I = percent inhibition

C = radial growth in check in mm/cm

T = radial growth in treated set in mm/cm

RESULTS

Aspergillus flavus.

Table 1 shows the effects of different concentrations of alcoholic extract of *Commiphora wightii* on the radial growth of *Aspergillus flavus*. Table shows that the alcoholic extract of root, shoot and seed is inhibitory to the growth of these fungi. Table further shows that with the increase in concentrations the rate of growth inhibition also increases. Thus, 10%, 25%, 50% and 75% root extract of *Commiphora wightii* retards radial growth of this fungi in culture medium by 10.1%, 18.2%, 26.4% and 48.9% of the control respectively at 7th day of growth. Observation further shows that like root extract growth is also inhibited in the presence of shoot and seed alcoholic extract under culture medium. Result further shows that the growth of these fungi inhibits more in presence of higher concentrations as compared to lower concentrations of extract.

Candida Albicans

Table 1 the effect of various alcoholic extract concentrations of the plant parts on the radial growth of *Candida albicans* in culture medium. Result shows that the growth is inhibited by alcoholic extract concentration and this inhibition rate increases with the increase in doses of plant part extract. Thus, radial growth of these fungi in 10%, 25%, 50% and 75% root extract concentration is 93.1%, 89.6%, 82.7% and 62.0% of the control respectively. Result further shows that the growth is inhibited more in 75% shoot and seed extract concentration as compared to 10% alcoholic extract concentration. Thus, in 10% shoot and seed extract concentration the radial growth of this fungi was 80.2% and 23.5% of control respectively, at 7th day, while, these values in 75% shoot and seed concentrations are 30.4% and 48.9% of the control respectively on 7th day.

Calatropis procera

Table 1 the effect of different concentrations of alcoholic extracts of various plant parts of *Calatropis procera*. Result shows that the radial growth of *Candida albicans* is affected by various concentration of alcoholic extract of plant parts. Observation further shows that with the increase in concentration of this medicinal plant part the rate of inhibition of fungal growth also increases. Thus, in 10%, 25%, 50% and 75% alcoholic concentration of root the radial growth is 92.2%, 72.5%, 48.3% and 42.1% of the control respectively at 7th day of growth. Result further shows that like root extract, shoot and seed extract also inhibits radial growth of fungi, however, this inhibition is more in higher concentration as compared to lower concentration of various plant parts of *Calatropis procera*.

Microsporum fulvum

Table 1 shows the effect of alcoholic extract concentrations of various plant parts of a plant *Mentha piperita* on the radial growth of *Candida aebicans*. Result shows that various concentrations of alcoholic extract of this plant also inhibit the growth of this fungi. Observation shows that with the increase in the concentration of alcoholic plant part extracts like root extract, shoot extract and rhizome extract the rate of inhibition increases. Thus, in 10% root extract the growth is 90% of the control whereas in 75% root extract concentration the growth is 35.8% of the control. Result further shows that shoot and rhizome extract also causes increase in inhibition rate like root extract.

TABLE – 1: Effect of different concentration of alcoholic extracts of plant parts of *Commiphora wightii* on growth performance of *A. Niger*, *A. Flavus*, *C. Albicans* and *M. Fulvum*

Days of Study	Diameter of Growth (Cm.) <i>A. Niger</i>			Diameter of Growth (Cm.) <i>A. Flavus</i>			Diameter of Growth (Cm.) <i>C. Albicans</i>			Diameter of Growth (Cm.) <i>M. Fulvum</i>		
	Root	Shoot	Seed	Root	Shoot	Seed	Root	Shoot	Seed	Root	Shoot	Seed
Growth in Control 0% extract												
5 th	2.1	2.9	2.2	1.2	1.4	1.7	1.3	1.2	1.1	1.8	1.1	1.8
7 th	2.5	1.8	3.2	2.3	2.6	2.7	2.5	2.3	2.4	2.6	2.4	2.2
Growth in 10% alcoholic extract												
5 th	1.6	2.5	1.2	1.4	1.5	1.3	1.2	1.1	1.3	1.4	1.5	1.7
7 th	2.2	3.2	3.6	2.8	2.2	2.3	2.4	2.7	2.4	2.2	2.3	1.7
Growth in 25% alcoholic extract												
5 th	1.1	1.3	1.9	1.8	1.5	1.7	1.5	1.4	1.5	1.8	1.2	1.4
7 th	1.8	2.0	2.6	1.7	2.2	1.4	2.3	2.9	2.4	1.6	1.2	1.3
Growth in 50% alcoholic extract												
5 th	1.2	0.3	1.2	1.0	1.3	1.2	0.5	0.8	1.2	1.8	0.4	1.3
7 th	2.2	1.2	2.1	1.2	1.4	1.9	1.8	1.6	2.4	1.6	1.1	1.2
Growth in 75% alcoholic extract												
5 th	0.8	0.4	0.7	0.9	0.7	0.3	0.5	0.3	0.8	0.2	0.7	0.6
7 th	1.2	1.1	1.3	1.6	1.7	1.2	1.8	1.3	1.7	1.4	1.3	1.1

DISCUSSION

The present investigations are being carried out to evaluate the antifungal medicinal properties of *Commiphora wightii* plant against the pathogenic fungi viz. *Aspergillus niger*, *A.flavus*, *Candida albicans* and *Microsporum fulvum*. Fungal infections comprise an important faction of diseases occurring not only in plants and animals but also in human beings. Moulds and yeasts are so widely distributed in human environment that human beings are instantly exposed to them. Fortunately, because of the relative resistance of human beings and comparatively non pathogenic nature of fungi, most of these exposures do not lead to over infection. However, fungi are gaining importance with respect to increased incidence of chronic, often fatal, mycoses in immune compromised patients [7]. The fungi present in soil, water and air constitute exogenous fungal opportunists. The roster of opportunistic fungal species continues to increase. However, some of the common ones include *Aspergillus fumigates*, *A.niger*, *A.terreus*, *A.flavus*, *Absida*, *Candida albicans*, *Cryptococcus neoformis*, *Microsporum fulvum*, *Mucor*, *Rhizomucor*, *Rhizopus*, and *Torulopsis globrata* [8]. To find suitable drug for the management of fungal diseases is difficult because fungi, like human beings, are eukaryotes. Many of the cellular and molecular processes are similar, and still

a number of chemicals are reported to have antifungal activity [9]. These include the derivatives of quinazolinone [10], coumarin [11], thiazolidinone [12], thiadiazole [13], Thiazole [14], Pyridine [15] and Sydnone [16].

REFERENCES

- [1] Soni V. (2010). *Conservation Evidence*, 7:27-31.
- [2] Kumar S, Shankar V (1982). *J Arid Environ* 5:1–11.
- [3] IUCN Red List of Threatened Species. Version. www.iucnredlist.org 17 June 2010.
- [4] Anonymous (1950). Council of Scientific and Industrial Research, New Delhi. Vol. 11: 313.
- [5] G.V. Satyavati (1990). *Economic and medicinal plant research*, vol 4. London: Academic Press, 39:56.
- [6] Natarajan D.J. Britto, S. Selvaraj and D.I. Arochiaswanay (2001), *Geobios* 28: 223-224.
- [7] De Hoog, G.S., Guarro, J., Gene, J. and M.J. Figueras (2000). Utrecht, the Netherlands and Universitat Rovirai Vigili, Reus, Spain
- [8] Singh, L. (1976), *Acta Botanica India* 4: 71-73.
- [9] Nene, Y.L. and B.W. Thapliyal (1979), Oxford & IBH Publisher house New Delhi. 425.
- [10] Farghaly, A.O. and A.M. Moharram (2000), *Chem. Abstr.* 132: 677.
- [11] Hankare, P.P., Jagtap., Battase, P.S. and S.R. Naravane (2002), *J. Indian Chem. Soc.* 79: 440-444.
- [12] Datta, N.J. Khunt, R.C. and A.R. Parikh (2002), *Indian J. Chem.* 41B: 433-435.
- [13] Yu, D.T., Macina, O.T., Sircar, I., Sircar, J.C. and C.M. Riviello (2001), *Chem. Abstr.* 134(3): 551.
- [14] Jag, M. 2000, *Chem. Abstr.* 132: 685.
- [15] Bhatt D.C., M.A. Nurani, K.D. Mitaliya & U.S. Baxi (2001), *Adv. Plant Sci.* 14(1): 427-431.
- [16] Bekhit, A.A., Habib, N.S. and A. El-Din Bekhit (2002), *Chem. Abstr.* 136: 805.