

INVESTIGATION OF ANTIMICROBIAL ACTIVITY OF *CORDIA MACLEODII* AND *LEUCAS CILIATA* LEAVES

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ABSTRACT: This study was carried out with an objective to investigate the antibacterial and antifungal potential of leaves of *Cordia macleodii* and *Leucas ciliata*. Antibacterial activity of ethanolic extracts of the leaves was carried out against three Gram negative bacteria – *Escherichia coli*, *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* and two Gram positive bacteria – *Bacillus Subtilis* and *Staphylococcus aureus*. The antifungal activity of the extracts was evaluated on two common pathogenic fungi – *Aspergillus niger* and *Candida albicans*. The testing was done by the agar plate method. Zones of inhibition of extracts were compared with that of standard Chloramphenicol for antibacterial activity and Nystatin for antifungal activity. The extracts showed antibacterial and antifungal activities comparable with that of standard against the organisms tested. The results showed that the inhibition of the bacterial growth was more pronounced on *E. coli* and *S. aureus* as compared to the other tested organisms. Both the extract showed the antifungal activity against *C. albicans*.

KEYWORDS: Antibacterial, antifungal, *Cordia macleodii*, *Leucas ciliata*

INTRODUCTION

Infectious diseases are the second leading cause of death world wide.¹ In industrialized nations, despite the progress made in the understanding of microbiology and their control, incidents of epidemics due to drug resistant microorganisms and the emergence of hitherto unknown disease-causing microbes, pose enormous public health concerns². The emergence of multidrug-resistant bacteria has created a situation in which there are few or no treatment options for infections with certain microorganisms³. Along with bacterial infections, the fungal infections also are a significant cause of morbidity and mortality despite advances in medicine and the emergence of new antifungal agents⁴.

Although the need for new antimicrobials is increasing, development of such agents faces

significant obstacles⁵. A number of factors make antimicrobial agents less economically attractive targets for development than other drug classes⁶. Pharmaceutical research and development costs, which are estimated to be \$400–\$800 million per approved agent⁷, pose a considerable barrier to new drug development in general.

Historically, plants have provided a good source of anti-infective agents; emetine, quinine, and berberine remain highly effective instruments in the fight against microbial infections. Phytomedicines derived from plants have shown great promise in the treatment of intractable infectious diseases including opportunistic AIDS infections. Plants containing protoberberines and related alkaloids, picralima-type indole alkaloids and garcinia biflavonones used in traditional African system of medicine, have been found to be active

against a wide variety of micro-organisms⁸. Many plants have been reported to have antifungal activity^{9,10}.

Cordia macleodii (Griff) Hook. F. & Thomas, is commonly known as Dahiphalas or Dahivan in Hindi and Bhoti in Marathi¹¹. It is an 8-10 m high tree with a corky grey bark. The leaves are broad ovate, 5 – 10 cm as long as broad, scabrous, base cordate and crenate-serrate margins. They are arranged alternate to subopposite. The flowers are white in colour and polygamous, in short terminal axillary corymbs. The calyx is densely tomentose the corolla lobes are oblong in shape and 0.6 to 0.8cm long. The drupes are 1.2 to 1.9 cm long, ovoid, acuminate at apex, seated at persistent calyx. The flowers and fruits appear in February – August¹².

Leucas ciliata (Benth) belongs to the family Lamiaceae (Labiatae). It is commonly known as Burumbi. It occurs as herbs or under shrubs 30-100 cm high with stems and branches obtusely four angled and has brownish hairs. The leaves are ovate or lanceolate in shape, about 3 – 9cm long and 2.5 - 4 cm wide. The lamina is membranous, sparsely hairy on both sides with an acute apex, a cuneate base and a serrate margin. The flowers are white in colour and have dense globose whorls. They have slender spinulose bracts equalling calyx. The calyx is tubular, hairy outside, with a ring of hairs at mouth and measures 1.2 – 1.8 cm in length. The corolla is long tube annulate inside and measures about 1.8-2.0 cm in length. The nutlets are smooth oblong-obvoid in shape and brown in colour. The flowers and fruits appear in May – August¹².

MATERIALS AND METHODS

Fresh Leaves of *C. macleodii* and *L. ciliata* were collected from same tree from Nandurbar district, Maharashtra and were authenticated by Botanical Survey of India, Pune. The leaves were shade dried and crushed to make coarse powder. The powder (250g) was extracted with three liters of ethanol (95%v/v) by continuous extraction method for 48 hours. Solvent was distilled off and the extract was concentrated and dried under reduced pressure, which yielded a brownish green mass. The extract was preserved at 2 to 4°C.

The antibacterial activity was evaluated on five common pathogenic bacteria viz *Escherichia coli* 2109 (NCIM, Pune), *Klebsiella pneumoniae* 2719 (NCIM, Pune), *Pseudomonas aeruginosa* 2036 (NCIM, Pune), *Bacillus Subtilis* 6633 (ATCC, Chandigarh), *Staphylococcus aureus* 2079 (NCIM, Pune). While the antifungal activity of the extracts was evaluated on two common pathogenic fungi viz. *Aspergillus niger* 545

(NCIM, Pune) and *Candida albicans* 3471(NCIM, Pune).

For evaluation of antibacterial and antifungal activities of alcohol extract of *C. macleodii* and *L. ciliata* leaves, Agar diffusion assay method was used¹³. For investigation of antibacterial activity, Sterile Muller Hinton agar media (Hi-media) were prepared in Petri dishes. The bacteria (1×10^8 bacteria/ ml) were inoculated separately in the media. In each Petri dish four wells (diameter 6mm) were prepared under aseptic conditions. In these wells, DMSO (1ml/well, as control), alcohol extract of *C. macleodii* (1ml of 500µg/ml solution in DMSO), alcohol extract of *L. ciliata* (1ml of 500µg/ml solution in DMSO) and Chloramphenicol (1ml of 10µg/ml solution in DMSO) were added. All the dishes were incubated at 35°C for 24 Hrs.

For investigation of antifungal activity, Sterile Potato dextrose agar media (Hi-media) were prepared in Petri dishes. The fungal spores (1×10^6 spores/ ml) were inoculated separately in the media. In each Petri dish four wells (diameter 6mm) were prepared under aseptic conditions. In these wells, DMSO (1ml/well, as control), alcohol extract of *C. macleodii* (1ml of 500µg/ml solution in DMSO), alcohol extract of *L. ciliata* (1ml of 500µg/ml solution in DMSO) and Nystatin (1ml of 100U/ml solution in DMSO) were added. All the dishes were incubated at 35°C for seven days.

At the end of the incubation period, the media were observed for zone of inhibition. The zones of inhibition were measured in millimeter using Vernier Calipers.

RESULT & DISCUSSION

The results of investigation of antibacterial and antifungal activities of *C. macleodii* and *L. ciliata* are summarized in tables 1 and 2 respectively. Ethanolic extract of *C. macleodii* and *L. ciliata* shows antibacterial activity against the *E. coli*, *K. pneumoniae*, *P. aeruginosa*, *B. subtilis* and *S. aureus*. *C. macleodii* extract shows maximum antibacterial effect on *E. coli* and *S. aureus* (Table 1). Ethanolic extract of *C. macleodii* and *L. ciliata* shows antifungal activity against the *Candida albicans* (Table 2).

Preliminary phytochemical analysis showed that the leaf extracts of *C. macleodii* and *L. ciliata* possess phenolic compounds, saponins and glycosides. Phytoconstituents such as saponins, phenolic compounds and glycosides have been reported to inhibit bacterial growth and to be protective to plants against bacterial and fungal infections^{14, 15}. Further studies need to be carried out to isolate the various classes of phytoconstituents and determine their antimicrobial potential.

Table 1: Antibacterial activity of alcohol extracts of *C. macleodii* and *L. ciliata* leaves.

Microorganism	Zone of inhibition of different bacteria (mm)			
	Control (DMSO)	<i>C. macleodii</i> extract	<i>L. ciliata</i> extract	Chloramphenicol
<i>E. coli</i>	@	13.25	10.52	30.18
<i>K. pneumoniae</i>	@	12.00	11.20	30.59
<i>P. aeruginosa</i>	@	09.60	11.20	28.90
<i>B. subtilis</i>	@	08.20	12.60	25.22
<i>S. aureus</i>	@	13.50	11.40	31.21

@ – NO zone of inhibition

Table 2: Antifungal activity of alcohol extracts of *C. macleodii* and *L. ciliata* leaves.

Microorganism	Zone of inhibition of different bacteria (mm)			
	Control (DMSO)	<i>C. macleodii</i> extract	<i>L. ciliata</i> extract	Nystatin
<i>Aspergillus niger</i>	@	-	-	20.50
<i>Candida albicans</i>	@	06.00	07.20	22.13

@ – NO zone of inhibition

REFERENCES

- World Health Organization (WHO). Deaths by cause, sex and mortality stratum in WHO Regions, estimates for 2001. World Health Report—2002. Geneva: WHO, 2002.
- Iwu M.M., Duncan A.R. and Okunji C.O., New antimicrobials of plant origin. In: Janick J ed. Perspectives on new crops and new uses. ASHS Press, Alexandria, VA. 1999, 457–62.
- Wenzel R.P. and Edmond M.B., Managing antibiotic resistance, New Engl. J. Med. 2000, 343, 1961–1963.
- McNeil M.M., Nash S.L., Hajjeh R.A., Phelan M.A., Conn L.A., Plikaytis B.D. and Warnock D.W., Trends in mortality due to invasive mycotic diseases in United States, 1980-1997, Clin. Infect. Dis., 2001, 33, 641-647.
- Gilbert D.N. and Edwards J.E. Jr., Is there hope for the prevention of future antimicrobial shortages?, Clin. Infect. Dis., 2002, 35, 215–16.
- Spellberg B., Powers J.H., Brass E.P., Miller L.G. and Edwards Jr J.E., Trends in antimicrobial drug developments: Implications for the future, Clin. Infect. Dis., 2004, 38, 1279-1286.
- DiMasi J.A., Hansen R.W. and Grabowski H.G., The price of innovation: new estimates of drug development costs, J. Health Econ., 2003, 22, 151–185.
- Iwu M.M., Jackson J.E. and Schuster B.G., Medicinal plants in the fight against leishmaniasis, Parasitol. Today, 1994, 10, 65 – 68.
- Parekh J. and Chanda S., *In vitro* antifungal activity of methanol extracts of some Indian medicinal plants against pathogenic yeast and moulds, African J. Biotech., 2008, 7, 4349-4353.
- Ertürk O., Antibacterial and antifungal activity of ethanolic extracts from eleven spice plants, Biologia, 2006, 61, 275-278.
- Prajapati N.D. and Kumar U., Agro's Dictionary of Medicinal Plants, 2003, Eastern Book Corporation, Delhi, pg. 92.
- Patil D.A., Flora of Dhule and Nandurbar Districts (Maharashtra), Botanical Survey of India, New Delhi, 2003, 418-420, 727-728.
- Pelczar Jr. M.J., Reid R.D. and Chan E.C.S., Cultivation of bacteria. In: Microbiology 4th Ed., Tata McGraw Hill Publishing Co. Ltd., New Delhi 1982, 103.
- Mather S.B. and Gonzale L., Identification of terpenoids from leaves of *Piptocarpha peritoria* and their biological activities, J. Nat. Prod., 1982, 45, 495-496.
- Okwute S.K., Plant derived pesticidal and antimicrobial agents for use in agriculture. A review of phytochemical and biological studies on some Nigerian plants, J. Agric. Sci. Technol., 1992, 2, 62-70.
