

DETERMINATION OF ANTIFUNGAL ACTIVITIES OF LEMON GRASS OIL ON MUCOSAL MICROORGANISM *CRYPTOCOCCUS NEOFORMANS*

Mishra R.P., Rathore Jaideo* and Rajput D.S.

Department of Post-graduate Studies and Research in Biological Sciences, Rani Durgavati University, Jabalpur, Madhya Pradesh, India.
Email: jaideo.rathore@rediffmail.com

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Abstract: The aim of the study to know the antifungal effect of lemongrass oil on mucosal microorganism *Cryptococcus neoformans*. The disc diffusion technique used for the *Cryptococcus neoformans* was carried out at different concentration of lemongrass oil as neat, 25%, 50%, 75%. The disc diffusion test for the *Cryptococcus neoformans* showed the inhibition zone of 22mm>20mm>15mm>12mm at the concentration of neat, 25%, 50%, 75%. The study has demonstrated that the essential oil of Lemongrass oil has significant antimicrobial potential against mucosal microorganism *Cryptococcus neoformans*.

Keywords: Essential oil, Antimicrobial activity, Mucosal microorganism

INTRODUCTION

Essential oils have been shown to possess antibacterial, antifungal, antiviral, insecticidal and antioxidant properties^{1, 2}. Some oils have been used in cancer treatment³. Some other oils have been used in food preservation⁴, aromatherapy⁵ and fragrance industries⁶. Essential oils are a rich source of biologically active compounds.

Lemon grass plant with scientific name *Cymbopogon citratus* belongs to the Graminae (Poaceae) family contains about 660 genus and 9000 species, widely distributed in tropical and subtropical world. Lemon grass is native to India and Sri Lanka^{7, 8}. *Cymbopogon citratus* is usually used in folk medicine for the treatment of neurological and gastrointestinal antimicrobial disorders and as an antispasmodic, analgesic, antibacterial, anti-pyretic, diuretic and sedative^{9, 10}. It showed that lemon grass plant due to having alkaloids and phenols contains antibacterial properties.

Among all the fungi only few species are pathogenic to humans. The most frequently diagnosed fungal infections are caused by pathogens from the genera *Candida*, *Cryptococcus* and *Aspergillus*¹¹. *Cryptococcus neoformans* is a saprophytic, basidiomycetous, dimorphic organism found worldwide, because its natural habitats are pigeon droppings and contaminated soil. Small-sized basidiospores (1.8 to 3.0 µm). Basidiospores or yeast cells may be inhaled by humans, then through the respiratory tract the pathogen can disseminate within the organism causing pulmonary infections, and subsequently, due to the *C. neoformans* predilection for the central nervous system, the life-threatening meningoencephalitis both in immunocompromised and immunocompetent patients. The pathogenic fungi from the genus *Cryptococcus* can invade human organism and may lead to mucosal and skin infections or to deep-seated mycoses of almost all inner organs, especially in immunocompromised

patients. The ability to grow at 37°C, capsule synthesis and melanin formation are important virulence factors of *C. neoformans*. If the infection of the central nervous system is not cured, it is fatal in 100% of cases^{12, 13}. It can turn into yeast cells, the form preferred at 37°C, or can form dikaryotic hyphae which are favored at 24°C¹⁴.

MATERIALS AND METHODS

The pure Lemongrass oil was obtained from J.N.K.V.V. Jabalpur was maintained at 4°C in brown bottle. The test microorganism *Cryptococcus neoformans* was obtained from clinical isolates. *Cryptococcus neoformans* isolated from patient cerebrospinal fluid was collected aseptically in a sterile bottle by puncturing subarachnoid space between third and fifth lumbar vertebrae. The fungal pathogens used were procured from Institute of Microbial Type Culture Collection, Chandigarh (IMTECH) viz., *Trichophyton rubrum* MTCC No.8477, *Candida albicans* MTCC No.1637 were maintained in Potato Dextrose Agar and Sabouraud dextrose agar.

Disc diffusion test

For antimicrobial activity testing the essential oil were diluted by adding equal volume of solvent. Under aseptic conditions, the test media was poured into sterile petriplates. The sterilized discs were impregnated with 15 µL of different concentrations (neat, 75%, 50%, 25%) of the essential oil and placed on the inoculated agar surface by sterile forceps. All treated and untreated plates were incubated for 18-24 h at 37 °C and after the end of incubation period antimicrobial activity was evaluated by measuring the diameters of inhibition zones.

RESULTS

The lemongrass oil was observed to possess the highest antifungal activity against all three microorganisms.

*Corresponding Author

The disc diffusion technique was used. The aqueous extract was found to be less effective. The antifungal activity was found progressively increasing with the increase in concentration of oil. The maximum effect was found at neat concentration and minimum effect was observed at 25% concentration of oil.

The zone of inhibition representing the antimicrobial activity of lemongrass essential oil on *Cryptococcus neoformans*, *Trichophyton rubrum*, *Candida*

albicans are presented on the table and figure. The zone of inhibition for *Cryptococcus neoformans* were 22mm>20mm>15mm>12mm. The zone of inhibition for *Trichophyton rubrum* were 30mm>25mm>20mm>11mm. The zone of inhibition for *Candida albicans* were 16mm>12mm>10mm>08mm. at the concentration of neat, 75%, 50%, 25%.

Inhibition zone diameter (mm)			
Concentrations of oil	<i>Trichophyton rubrum</i>	<i>Candida albicans</i>	<i>Cryptococcus neoformans</i>
25 %	11	08	12
50 %	20	10	15
75 %	25	12	20
100 %	30	16	22
Ketoconazole	35	28	30



Fig.1 *Candida albicans*



Fig.2 *Cryptococcus neoformans*



Fig.3 *Trichophyton rubrum*

DISCUSSION

Finding healing power in plants is an ancient idea. Bioactive compounds are always toxic at higher dose. The public is becoming increasingly aware of problem with the over prescription and misuse of traditional antibiotics. These substances serve as plant defense mechanism against predation by microorganisms, insects and herbivores. Essential oils are potential sources of novel drugs especially against fungal pathogens. An important characteristic of Essential oil and their components is their hydrophobicity which enables them to disturbance of microbial membranes, disrupting the proton motive force, electron flow, active transport and resulting in the coagulation of intracellular contents¹⁵. Lemongrass oil shown good anti-fungal activity against *Aspergillus* species: *A. niger*, *A. flavus*, *A. fumigatus*, *A. nidulans* and *Fusarium oxysporum* at a concentration of 2.5 ml of oil in 1000 ml of medium¹⁶. Steam distillate from *Cymbopogon citratus* completely inhibits the growth of *Ustilago maydis*, *Ustilago violacea*, *Curvularia lunata* and *Rhizopus* species¹⁷. Application of lemongrass oil as preservative to control Blue Mould Decay by *Penicillium italicum*, in oranges is shown to be effective and dosage comparable with those chemicals used in United States¹⁸. Lemon grass essential oil has the ability to control bacterial

growth and fungal pollutants in food such as *Staphylococcus aureus* and *Escherichia coli*, also the antioxidant activity of plant essential oil has proven that lemon grass essential oil in comparison with alpha-tocopherol has stronger antioxidant activity and acts as BHT equivalent (Butylated Hydroxy Toluene)¹⁹. Lemongrass repels the tsetse fly. Creams containing the oil are used as repellents against houseflies and mosquitoes. Contact toxicity of the oil in acetone solution on insects is very high. 7.5 µg/per insect was effective in killing houseflies and 30 µg/per insect killed mosquitoes²⁰.

CONCLUSION

The present study indicates that the Essential oil from Lemongrass exhibits good to excellent antifungal activity against mucosal microorganism *Cryptococcus neoformans*. So the Lemongrass may be considered as good source for antifungal activity for medicinal use in various infections. Therefore, further investigation should be necessary for the development of novel lead compound and more data will be necessary either to confirm in vitro efficacy or to explain the mechanism of essential oil.

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