ANTIFUNGAL ACTIVITY OF EUCALYPTUS OIL AGAINST CLINICAL ISOLATES OF CANDIDA SPECIES

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INTRODUCTION

The majority of mycotic infections are due to Candida species, out of which Candida albicans is being the most frequently isolated species [1]. Various virulence factors play a crucial role in the pathogenicity of several microorganisms including Candida albicans [2,3,4].

Eucalyptus is one of the very important and most widely planted genera across the world. It is a tall, evergreen tree, native to Australia and Tasmania, successfully introduced worldwide, now extensively planted in many other countries. Thus, the aim of the present study was to determine the antifungal activity of eucalyptus oil against clinical isolates of Candida species. We have observed that, clinical isolates of Candida species were not inhibited in any of these dilutions of this oil. The eucalyptus oil does not have antifungal activity against Candida species. However, it is important to include more number of isolates and different dilutions to validate the results.

MATERIALS AND METHODS

Candida isolates

A total of 20 non repetitive clinical isolates of Candida species were collected from different samples of immunocompromised individuals attending Saveetha Medical College, Thandalam. They were characterized by carbohydrate fermentation and assimilation tests and confirmed [7]. Isolates were preserved in semisolid Sabouraud chloramphenicol semi solid stock and stored at 4°C until further use.

Characterization of Candida species

Candida species were further characterized by using Hichrom agar (HiMedia, Mumbai).

Preparation of Hichrom agar

CHRO Magar Candida (HiMedia, Mumbai) was prepared following manufacturer’s instructions. About 21.02 gram of Hi Chrome Candida differentiation agar base (modified) was suspended in 500 ml of distilled water. It was heated to boiling...
gently to dissolve the medium completely. Then it was allowed
to cool to 50°C and rehydrated (one vial) contents of
Hichrome Candida selective supplement was added under
aseptic precautions. It was mixed well and poured into
petridishes. Isolates were identified on Hichrome agar based
upon the characteristic color of the colony by subculturing from
Sabouraud's chloramphenicol agar plates and the Candida
Hichrome plates were incubated at 37°C for 24- 48 hours[9].
Based on colour produced by the isolates speciation have
been made.

| Candida species     | colour     |
|---------------------|------------|
| C. albicans         | Green      |
| C. tropicalis       | Blue       |
| C. krusei           | Pale       |
| C. kefyr            | Pale       |
| C. parapsilosis     | Pale       |

Detection of antibacterial activity of eucalyptus oil against clinical isolates of Candida spp

Antifungal activity of eucalyptus oil was tested against Candida
spp isolates by minimum inhibitory concentration method.
Mueller Hinton broth was supplemented with 0.002% (V/V)
tween 80 (HiMedia, Mumbai) to enhance the dispersion of the
essential oil. Agar dilution method was performed to attain
the different concentrations of essential oils such as
0.03%, 0.06%, 0.125%, 0.25%, 0.5%, 1% and 2% in Mueller
Hinton Agar (MHA).

Media containing various concentrations of essential oils were
poured over the sterile petridishes and allowed to dry. Media
without essential oil was served as control plate. Spot
inoculation of 0.5

McFarland standard turbidity adjusted isolates were made on
the plates and incubated at 37°C for overnight. The lowest
concentration of the essential oils that completely inhibited the
growth of isolates was considered as MIC. [10]

RESULTS

Characterization of Candida species by Hichrom Candida
agar

A total of 20 clinical isolates of Candida spp were seeded on to
Hichrom Candida agar and results were tabulated based on
pigment production.

Sample wise distribution of Candida species

Of the 20 clinical isolates of Candida spp, 6/20 (30%) were
from oral thrush, 5/20 (25%) from urine, 3/20 (15%) from
sputum, 3/20 (15%) from vaginal swab, 2/20 (10%) from ear
swab and one (5%) from wound swab.

Result of antifungal activity of eucalyptus oil against clinical
isolates of Candida species

We have observed that, clinical isolates of Candida species
were not inhibited in any of these dilutions of this oil.

DISCUSSION

Study conducted by Prakasam et al from Chennai in 2014
demonstrated that, Acinetobacter strains were inhibited from
0.06 to 0.25%, 0.25-1% and 0.125-1% for clove, peppermint
and eucalyptus oils respectively. In clove oil, 14/50 (28%)
isolates were inhibited at 0.06%, 25/50 (50%) at 0.125%
and 11/50 (22%) at 0.25% of clove oil. In peppermint oil, 34/50
(68%) isolates were inhibited at 0.25%, 12/50 (24%) and 4/50
(8%) were at 0.5% and 1% concentrations of peppermint oil
respectively. In eucalyptus oils, 10/50 (20%) isolates were
inhibited at 0.125%, 18/50 (36%) at 0.25%, 16/50 (32%) and
6/50 (12%) were at 0.5% and 1% respectively. Thus, the MIC
of clove oil was found to be 0.06%, 0.25% for peppermint oil
and 0.125% for eucalyptus oil. [10]. In contrast, our isolates did
not show any inhibitory activity against eucalyptus oil with
these dilutions.

CONCLUSION

The eucalyptus oil does not have antifungal activity against
Candida species. However, it is important to include more
number of isolates and different dilutions to validate the results.
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