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Comparative Evaluation of Ethno-Medicinal Use of two Species of *Eucalyptus* Plant as an Antimicrobial Agent

Gero Mohammed, Abe Ayotunde S, Inuwa Bashir,
Babakura M. Aji, Sule Aliyu M,.Mohammed Hauwa
Environmental Technology Division,
National Research Institute for Chemical Technology- Zaria, Nigeria.

ABSTRACT

In the search for alternative ways of plant diseases control, essential oil from two species *Eucalyptus* plant (*Eucalyptus citriodora and Eucalyptus camaldulensis)* were studied for their antimicrobial properties against six microorganisms comprising of five bacteria(*Bacillus subtilis Pseudomonas aeruginosa, Escherichia coli, Staphylococcus aureus and Klebsiella pneumoniae*) and one fungus (*Candida albicans*). The disk diffusion method was used for the assessment of the inhibitory effects of these essential oils. It was discovered that the antibacterial activity of *Eucalyptus citriodora* oil was significantly higher than *Eucalyptus camaldulensis* against the tested microorganisms. *Bacillus subtilis, Staphylococcus aureus, Klebsiella spp, Pseudomonas aeruginosa* were all resistant against *Eucalyptus camaldlensis* while high antibacterial activities of 16mm, 18mm, 18mm, 8mm zone of inhibition was recorded when the same bacteria was tested against *Eucalyptus citriodora* respectively. The highest zone of inhibition o antibacterial activity was recorded when *Eucalyptus citriodora* was used against *Candida albicans(22mm)*.With the result of this findings it can be observed that *Eucalyptus* plant most especially *Eucalyptus citriodora* provide a promising solution in ethnomedicine practice of diseases control providing the need to further investigate and evaluate other essential oil from other plants to complement and possible replace the synthetic antibiotic used in the treatment of infection since it is generally agreed that natural medicinal practice if used in the proper manner is more safe than the use of synthetic drugs in the treatment of infection due to side effect associated with the use synthetic drugs

Keywords: *Ethno-medicinal, antimicrobial agent, pathogenic microorganisms, essential oil.*

I. INTRODUCTION

"Essential oils" are the therapeutic, volatile oils that come from plants. In aromatherapy, the word "volatile" is not meant as "explosive" or "inconsistent." Rather, this refers to the meaning: "evaporating readily at normal temperatures and pressures…[an oil that] changes readily from solid or liquid to a vapor [as in] 'it was heated to evaporatethevolatiles'"(http://dictionary.reference.com/search?q=volatile). Essential oils may be found in leaves, rinds of fruit, seeds, bark, heartwood of trees, flowers, and any other part of a plant, so long as the extracted oil has medicinal or otherwise therapeutic use. *Eucalyptus camaldulensis* is an important ethnomedicinal plant belonging to the family, Myrtaceae. It is used as a remedy for sore throat and other bacterial infection of the respiratory and urinary tracts. Essential oils of the leaves are used in the treatment of lung diseases while the volatile oils are used as expectorants (. Adeniyi etal 2006). Topical ointments containing eucalyptus oil have also been used in traditional Aboriginal medicines to heal wounds and fungal infections. Eucalyptus oil obtained by steam distillation and rectification of the fresh leaves has Eucalyptol (1,8-cineole) as its active ingredient and this is responsible for its various pharmacological actions (Trivedi etal 2004). The antimicrobial activities of the methanolic extracts of E. camaldulensis have also been reported (Akin-Osanaiye etal 2007, . Mehraban etal 2005). The emergence of bacterial resistance to the currently available antimicrobial drugs necessitates further research in the discovery of new safe and effective antibacterial agents(Lokhande etal 2007). The investigation of certain indigenous plants for their antimicrobial activity is therefore of utmost importance. This study is aimed at investigating evaluating the antimicrobial activities of *Eucalyptus camaldulensis* and *Eucalyptus citriodora* against some clinical isolated microorganisms thereby establishing it as a potential antimicrobial agent.

II. METHODOLOGY

Plants Materials

The two plants *Eucalyptus citriodora, Eucalyptus camaldulensis* used in this study was obtained from National Research Institute for Chemical Technology, Zaria Nigeria.

Isolation of the Essential Oils

The isolation of the essential oil was carried out as described by Mazari1 etal 2010.The two species of *Eucalyptus* plant (*Eucalyptus citriodora* and *Eucalyptus camaldulensis*) was dried and samples (leaves) were subjected to steam distillation as described by Mbata etal. Samples oils were dried over anhydrous sodium sulphate and stored at low temperature before analysis and bioassay.

Tested Microorganisms

Six microorganisms, *Bacillus subtilis Pseudomonas aeruginosa, Escherichia coli, Candida albicans Staphylococcus*

aureus and Klebsiella pneumoniae was used for the research. These microorganisms were isolated from Ahmadu Bello Teaching Hospital Microbiology laboratory.

Inocculum Preparation

A loopful of isolated colonies was inoculated into 4 ml peptone water and incubated at 37^0C for 4 hours. The turbidity of actively growing bacterial suspension was adjusted to match the turbidity standard of 0.5 McFarland units prepared by mixing 0.5 ml of 1.75% (w/v) barium chloride dehydrate with 99.5 ml 1% (v/v) sulphuric acid. This turbidity was equivalent to approximately 1.2×10^8 colony-forming units per milliliter (cfu/ml). The grown suspension was used for further testing.

Antimicrobial Bioassay

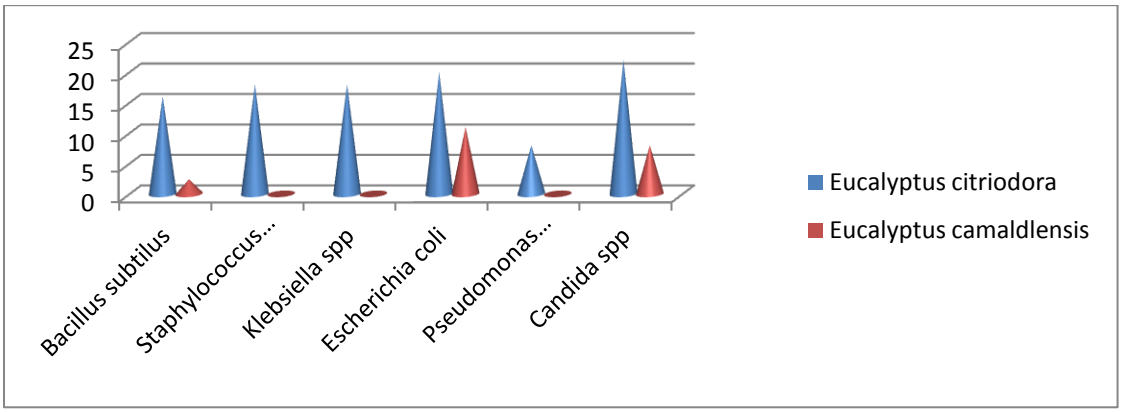
The activities of the essential oil were determined by the Kirby-Bauer agar diffusion method according to NCCLS standards (Bauer *etal* 1966, NCCLS, 2000). Nutrient agar was used for the antimicrobial activity test. Under aseptic conditions in the Biosafety chamber, 15ml of Nutrient agar medium was dispensed into pre-sterilized Petri dishes to yield a uniform depth of 4 mm and inoculated by the bacterial culture. Sterile disks (Diameter 6mm) were impregnated with the two different essential oil dried for 10-15 minutes. The dried discs were placed on Nutrient agar surface with flamed forceps and gently pressed down to ensure contact with the agar surface. The discs were spaced far enough to avoid reflections wave from the edges of the Petri dishes and overlapping rings of inhibition. Finally, the Petri dishes were incubated for 18 to 24 hours at 37^0C . The diameter of zone of inhibition as indicated by clear area which was devoid of growth of microbes was measured and recorded.

III. RESULTS

The antibacterial activities of two essential oils of different species of *Eucalyptus (Eucalyptus citriodora* and *Eucalyptus camaldlensis*) was investigated against six clinical pathogenic isolates comprising of five bacteria (*Bacillus subtilis Pseudomonas aeruginosa, Escherichia coli,Staphylococcus aureus and Klebsiella pneumoniae*) and one fungus (*Candida albicans*) were assessed for the presence or absence of zone of inhibition and zone diameter was measured and recorded as showed in the table and graph below.

Table Showing the Antimicrobial Activities of *Eucalyptus Citriodora* and *Eucalyptus Camaldlensis* against Clinical Isolates

ISOLATES	*Eucalyptus citriodora*	*Eucalyptus camaldlensis*
Bacillus subtilus	16	R
Staphylococcus aureus	18	R
Klebsiella pneumoniae	18	R
Escherichia coli	20	11
Pseudomonas aeruginosa	8	R
Candida albicans	22	8



Graphical Representation of the Antimicrobial Activities of *Eucalyptus Citriodora* and *Eucalyptus Camaldulensis* against Clinical Isolates

IV. DISCUSSION

The results of susceptibility test of two essential oils of *Eucalyptus citriodora* and *Eucalyptus camaldlensis* shows that the zone of inhibition recorded with *Eucalyptus citriodora* against the tested microorganisms was higher than *Eucalyptus camaldlensis* most especially against *Candida spp* (22mm zone diameter) against 8mm recorded with *Eucalyptus camaldlensis.* The highest zone of inhibition recorded with *Eucalyptus camaldlensis* was against *Escherichia coli* (11mm) which is less when compared with 20mm recorded against *Escherichia coli* when *Eucalyptus citriodora* was used. Other bacteria such as *Bacillus subtilis, Staphylococcus aureus, Klebsiella spp, Pseudomonas aeruginosa* were all resistant against *Eucalyptus camaldlensis* but higher zone of 16mm, 18mm, 18mm and 8mm zone diameters swas observed when *Eucalyptus citriodora* was used. The findings of this research further proof the fact that there is different in terms of genetic, Phytochemical and also antibacterial activity of different species of a particular plant and also the antibacterial activities demonstrated confirms earlier findings that volatile plants

V. CONCLUSION AND RECOMMENDATION

From the research carriedout it is obvious that *Eucalyptus citriodora* is more effective against the tested microorganisms (*Bacillus subtilis Pseudomonas aeruginosa, Escherichia coli, Candida albicans Staphylococcus aureus and Klebsiella pneumoniae*) than *Eucalyptus camaldlensis*. And this further proof the need to investigate different species of a particular plant when carrying out antimicrobial activity in other to know which species of a particular pant offer the best antimicrobial effect the tested microorganisms. Finally Phytochemical analysis of this two species of *Eucalyptus* should be carried out to ascertain the different chemical composition necessary for this difference in antimicrobial activity and also invi-vo studies should be carried out to establish its safety to be use by humans as it posses significant antimicrobialproperties.(Cowan 1999., Dorman etal 2000.,Onawuummi etal 1984.,Ibrahim etal 1999 and Onawummi etal 1987)

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