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Antibacterial Potential of Essential Oil Extracted from *Eucalyptus* Species

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Abstract: ***The antibacterial activity of essential oil of Eucalyptus species against pathogenic bacteria was evaluated following agar disc diffusion method. This aromatic medicinal plant is the richest bioresources of drugs for traditional system of medicine, modern medicine, folk medicine, nutraceutical, pharmaceutical and chemical entities for synthetic drugs. Aromatic plants are the sources of fragrance, flavours, cosmeceuticals, health beverages and chemical terpens. TLC profiling of pigments and oil of Eucalyptus sp. were done and their R_f values were recorded. Alcoholic extract of 3 sp. of Eucalyptus leaves viz.- E. robusta, E. tereticornis, E. globulus were analyzed for its antibacterial potential against 6 bacterial sp. selected. The highest antibacterial activity was observed against bacteria- Agrobacterium tumifasciens followed by A. rhizogene, Xanthomonas sp. and Salmonella typhii while the minimum activity was observed against Pseudomonas aeruginosa and Staphylococcus aureus. In the present study leaves of three selected Eucalyptus sp. were screened for antibacterial potential against 6 clinically significant bacterial sp. by disc diffusion method.***

Keywords: antibacterial activity, Eucalyptus sp., essential oils etc.

1. Introduction

The *Myrtaceae* family includes 140 genera and about 3800 species distributed in tropical and subtropical regions of the world. *Eucalyptus* is evergreen 300 feet tall tree. Leaves of the tree are opposite, sessile, lanceolate, 6-12 inches long and 1-2 inches broad. It is native of Australia, now extensively planted in many countries. It is also known as Nilgiri, Tailpatra, Sugandhpatra etc. *Eucalyptus* species is well known as medicinal plant because of their medicinal and pharmacological properties *E. globulus*is main furnisher of essential oils. These essential oils are in great demand in the market since they find applications as anesthetic, antiseptic, astringent, deodorant, disinfectant, fumigant, hemostat, inhalant, insect repellent, for a folk remedy for abscess, inflammation, fracture healing, obesity, arthritis, asthma, boils, bronchitis, burns, cancer, diabetes, laryngitis, flu, worms and wound etc. Sometimes their demand is also in the cosmetic industries. Its pulpwood is used in paper industry, making plywood, doors and windows. Medicinal plants also show dyspeptic, antioxidant, and antimicrobial activity [1].

Eucalyptus has an important role in farm industry [2]. There are 3 ways in which plant have been found useful in medicine

1) They may be used directly as tea or in other extracted forms.
2) They may be used as agent in the synthesis of drugs.
3) Organic molecules found in plants may be used as models for synthetic drugs.

"Essential oils are the volatile materials derived by a physical process from odorous plant material. An essential oil is concentrated hydrophobic, volatile ethereal oil or ethereal [3] and used in aromatherapy, gynecomatia, ingestion and flammability. Photosynthesis drives life on earth by producing both oxygen and organic matter [4].

The main objective of the study is to examine the antibacterial activity of 3 *Eucalyptus* sp. on clinically significant microorganisms by means of the disc diffusion test.

2. Materials and Methods

Plant sample

Leaves of all the 3 selected sp. *Eucalyptus robusta, Eucalyptus tereticorni, Eucalyptus globulus* were collected from different areas of Jabalpur (M.P.) and screening were done.

Extraction of oil

By hydrodistillation of air dried *Eucalyptus* leaves oil was extracted by a Clevenger- type apparatus [5].

Pigment extraction from Eucalyptus leaves

Sample preparation (Mc Laughlin and Masters, 2004) 0.5 gm Eucalyptus leaves, 0.5 gm anhydrous sulphate and 1 gm sand, this mixture was grinded and transferred into a test tube and added 2 ml acetone, was stirred and allowed to settle for 20 minutes. Centrifugation was done at 5000 rpm for 30 min and the supernatant green layer was pipette out.

For profiling of pigments, TLC plates [6, 7] was prepared and chromatography done. R_f values were determined by measuring the movement of solvent molecule and movement of solute molecule with a ruler and calculated by formula-

$$R_f = \frac{\text{Distance travel by the solute}}{\text{Distance travel by the solvent}}$$

Antimicrobial activity of *Eucalyptus* sp

The essential oil extracted from *Eucalyptus robusta , Eucalyptus tereticorni , Eucalyptus globulus* and were tested against 6 bacteria:

1) Agrobacterium tumifasciens
2) Agrobacterium rhizogene
3) Xanthomonas sp.
4) Staphylococcus aureus

5) Pseudomonas aeruginosa
6) Salmonella typhii

The antibacterial activities of all the 3 *Eucalyptus* sp. were done by filter paper disc diffusion method. A suspension of each test organisms diluted was spread over the solid agar plates. Filter paper disc (6 mm in diameter) were soaked in 13 uL of essential oil and placed on the incubated plates and allowed to dry for 15 min. Now plates were inoculated at 37ºC for a day.

3. Result and Discussion

The study showed that the solvent extracts investigated were active against various Gram positive and Gram negative bacteria. All the 3 sp of Eucalyptus leaves exhibited stronger antimicrobial activity in alcoholic extracts. Extract inhibited the growth of *A. tumifaciens* to the maximum followed by inhibition zones of A. *rhizogene* and *Xanthomonas* sp. intermidiate zone was observed for *Salmonella typhii* while minimum zone of inhibition was for *Pseudomonas-aeruginosa* and *Staphylococcus aureus* against all the 3 sp. of *Eucalyptus* (fig. 1).

The antimicrobial activity and toxicity can't be attributed to the same component of the culture. Since the chloroform fraction contained the bacteriostatic compound, the aqueous fraction had some toxic effect but did not affect antibacterial growth. Result of TLC study showed that *E. globulus* was the richest source of secondary metabolite because 7 different bands were observed, other two sp. also showed 5 bands each and these bands were represent that all the sp. possesses different active groups which were responsible for antibacterial activity of the plants (table 1).

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Table 1: R_f values of different fractions from leaf extracts of Eucalyptus sp.

S. No.	Name of pigments	R_f Velues			Colors
		E. robusta	*E. tereticorni*	*E. globulus*	
1	Lutein	0.29	0.09	0.07	Light Yellow bands
2	Chlorophyll a	0.42	0.54	0.50	Dark Green bands
3	Pheophytin a	0.60	-	0.81	Light yellow bands
4	Xanthophylls	-	0.15	0.11	Dark Yellow bands
5	Oil	0.73	0.81	0.87	Grayish spot
6	Chlorophyll b	-	-	0.38	Green band
7	Beta Carotene	0.78	0.91	0.93	Dark Yellow band

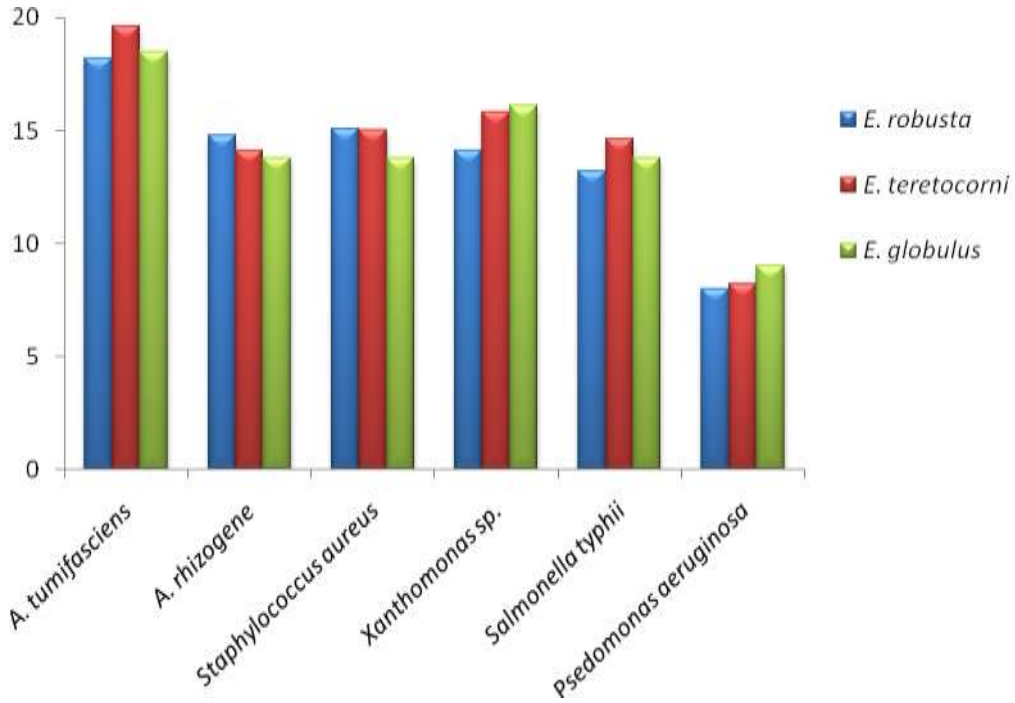

Figure 1: Graphical representation of antibacterial activity of *Eucalyptus* sp.