



## Research Article

**ANTIMICROBIAL ACTIVITY OF ESSENTIAL OIL OF FLOWERS OF *PLUMERIA ALBA* LINN (APOCYNACEAE)**ZAHID ZAHEER\*, KHAN SUBUR W, PATEL KHUMAN A, KONALE AJINKYA G, LOKRE SHEKHAR S<sup>1</sup>

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**ABSTRACT**

Antimicrobials of plant origin have enormous therapeutic potential. They are effective in the treatment of infectious diseases while simultaneously mitigating many of the side effects that are often associated with synthetic antimicrobials. The spread of drug resistance pathogens is one of the most serious threats to successful treatment of microbial diseases. *Plumeria alba* Linn (Apocynaceae) is an aromatic plant which little information has been reported so far on the composition and activity of essential oil. This research focuses on the antimicrobial activity of essential oil. The essential oil of flowers was obtained by hydrodistillation with a Clevenger apparatus. The oil content was 0.3 (%w/w), on a dry weight basis.

**Keywords:** *Plumeria alba*, Essential oil, Apocynaceae, Antimicrobial

**INTRODUCTION**

Herbs are widely exploited in the traditional medicine and their curative potentials are well documented.<sup>1</sup> The results of different studies provide evidence that some medicinal plants might indeed be potential sources of new antibacterial agents even against some antibiotic resistant strains.<sup>2</sup> *Plumeria alba* Linn (Apocynaceae) commonly called White Champa, a small laticiferous tree or shrub, native of tropical America. It is 4.5m high, cultivated in Indian gardens and popularly called 'peru' in Tamil.<sup>3</sup> The plant is mainly grown for its ornamental and fragrant flowers. Leaves lanceolate to oblanceolate, flowers white, fragrant, in corymbose fascicles.<sup>4</sup> The fruit is edible, latex is applied to ulcers, herpes and scabies and seeds possess haemostatic properties. Moreover its bark is bruised and applied as plaster over hard tumours.<sup>5</sup> Whereas the latter taxon finds use as purgative, cardiotoxic, diuretic and hypotensive.<sup>6</sup> Methanolic extract showed antimicrobial activity against *Bacillus anthracis*, *Pseudomonas aeruginosa*.<sup>7</sup> The plant is reported to contain amyriacetate, mixture of amyriins,  $\beta$ -sitosterol, scopoletin, the iridoids isoplumericin, plumieride, plumieride coumerate and plumieride coumerate glucoside.<sup>8,9</sup> The milky sap of the stem and leaf is applied to skin diseases such as herpes, scabies and ulcers.<sup>10</sup> Its bark is used as plaster over hard tumors', the seeds in hemostasis while the latex is used as purgative, cardiotoxic, diuretic and hypotensive.<sup>11</sup> An extract of it is used internally and externally for syphilitic ulcers.<sup>12</sup> The composition of volatile oil was not reported but reported that volatile oil has antioxidant activity (IC<sub>50</sub> of 1.0766 mg/ml). This is the first report on the antimicrobial activity of the flower volatile oil of *Plumeria alba*. The goal of the present work was to study the antimicrobial activity of the flower volatile oil of *Plumeria alba*.

**MATERIALS AND METHODS****Authentication of plant**

The fresh plant materials were collected from Aurangabad, Dr. Rafiq Zakaria Maulana Azad Campus, Y.B. Chavan College of Pharmacy (MS), India during the month of August 2009. The botanical identity of the plant was confirmed at the Botany Department of Dr. BAMU, Aurangabad. A voucher specimen has been deposited at the Museum of the Department of Botany, Dr. BAMU, Aurangabad.

**Essential oil isolation<sup>13, 14, 15</sup>**

The flowers of *P. alba* were harvested from healthy, well grown plants. Freshly harvested flowers (200g) were subjected to hydrodistillation using a modified Clevenger type glass apparatus for 3-4 h for isolation of oil. The oil sample was stored at 0°C in air tight container after drying them over anhydrous sodium sulphate before going to activity.

**Drug resistant test organisms used for antibacterial activity**

The following microorganisms were used as test organisms: Gram positive *Staphylococcus aureus*, *Bacillus subtilis*, Gram negative *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella typhi*,

**Preparation of Muller-Hinton agar medium**

Muller-Hinton agar (Himedia, India) is considered to be the best for routine susceptibility testing of drug resistant bacteria. It should be prepared from a commercially available dehydrated base according to the manufacturer's instruction.

**Well diffusion method<sup>16, 17</sup>**

The organisms were cultured in nutrient broth and the tests carried out on Mueller Hinton agar plates. The inocula of the microbial strains were prepared from 24 h broth cultures and suspensions were adjusted to optical density of 1.0 at 600nm turbidity. The essential oils were dissolved in methanol (0.3 ml oil/2 ml methanol) and standard antibacterial Doxycycline Hyclate was dissolved in methanol (5 mg/ml). Muller Hinton agar was poured into petridishes. After solidification 0.5 ml of test strains were inoculated in the media separately. Care was taken to ensure proper homogenization.

The experiment was performed under strict aseptic conditions. After the medium solidified, a well was made in the plates with sterile borer (5 mm). The oil samples (10 $\mu$ l, 15 $\mu$ l, 20 $\mu$ l) were introduced into the well and plates were incubated at 37°C for 72 hrs. All samples were tested in duplicate. Microbial growth was determined by measuring the zone of inhibition. A control with Doxycycline Hyclate was kept for all test strains and the control activity was deducted from the test & results were recorded. The MIC value was defined as the lowest concentration of the volatile oil required for inhibiting the growth of each microorganism.

**RESULTS AND DISCUSSION**

The essential oils obtained from flowers of *Plumeria alba* were tested against set of microorganisms in order to estimate their antimicrobial potentials. The results are presented in Table 1. Gram-negative bacteria appear to be least sensitive to the action of many other plants essential oils.<sup>18-23</sup> The volatile oils of *P. alba* flower part were more active against *S. aureus* and *B. subtilis* (Fig.1, 2), presenting an important growth inhibition at lower concentrations. The antibacterial activity of the volatile oils tested was more pronounced against Gram-positive than against Gram-negative bacteria. This generally higher resistance among Gram-negative bacteria could be ascribed to the presence of their outer membrane, surrounding the cell wall, which restricts diffusion of hydrophobic

compounds through its lipopolysaccharide covering.<sup>24</sup> The absence of this barrier in Gram-positive bacteria allows the direct contact of the essential oil's hydrophobic constituents with the phospholipids bilayer of the cell membrane, causing either an increase of ion

permeability and leakage of vital intracellular constituents, or impairment of the bacterial enzyme systems.<sup>25,26</sup> MIC of Volatile oil was carried out using gradient plate technique, minimum concentration to inhibit growth is approximately 7-8  $\mu$ l.

**Table 1: Antimicrobial activity of *Plumeria alba* volatile oil**

Bacterial species	Inhibition zone diameter (mm)				
	Essential oil			Control	
	10 $\mu$ l	15 $\mu$ l	20 $\mu$ l	Negative	Positive
<i>Gram (+)</i>					
<i>S.aureus</i>	21	25	28	0	50
<i>B. subtilis</i>	15	18	21	0	49
<i>Gram (-)</i>					
<i>E. coli</i>	0	0	0	0	35
<i>S. typhi</i>	0	2	3	0	35
<i>Pseudomonas</i>					
<i>Aeruginosa</i>	2	3	3	0	35



**Fig.1 Inhibition zone of *S. aureus***



**Fig.2 Inhibition zone of *B. subtilis***

## CONCLUSION

In conclusion, our study can be considered as the first report on the antimicrobial properties of *Plumeria alba* volatile oils. Our results are a contribution to a better valorization of this medicinal plant. Several other biological tests will be worthwhile to search for more eventual activities of this plant. Phytochemical investigations will be planned to identify and characterize active principles, and assess toxicity by laboratory assays.

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