



Research Article

Extraction, Phytochemical Investigation and Antimicrobial Activity of *Plumeria obtusa*

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ABSTRACT

Many medicinal plants are claimed to be useful in skin diseases in all traditional systems of medicine and folklore. At the same time, these herbal remedies are used orally and by topical application. Looking at the scope of the herbal drug and the increasing demand, especially for liver disease, cancer, diabetes, hypertension, kidney disease, inflammation, infectious disease, skin disease, etc. *Plumeria obtusa* is a deciduous tree with a thick, widely spread, common rather wet garden; in lawns and open plantations, the tree is unusual in appearance. The plant loses leaves for a short time during the winter. This study aims to determine phytochemical components using different solvents and to study the antibacterial effect of methanol extract to varying concentrations against these bacteria. Phytochemical analysis of *Plumeria obtusa* leaves extracts indicate the presence of various medicinally valuable components, including alkaloids, glycosides, carbohydrates, phytosterols, phenolic compounds, tannins, saponins, flavonoids, proteins, and amino acids. Current findings indicate that methanol is a good solvent for extracting antibacterial agents. Further studies are needed to investigate the individual phytochemical compounds of *Plumeria obtusa* Leaves can be considered as effective as the most effective artificial antibiotics.

INTRODUCTION

Plants contain secondary metabolites, organic compounds not directly involved in the normal growth, development, or reproduction of organisms but often play an important role in plant defenses¹. Examples include alkaloids, glycosides, terpenoids, phenols, tannins, flavonoids and

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saponins². Furthermore, there is growing interest in the chemical composition of plants towards the discovery of more effective biotherapeutic agents³. The importance of natural products in modern medicine has been discussed in recent reviews and research^{4,5}.

Medicinal plants are now getting more attention than ever because they can benefit society, especially in medicine and pharmacology. The therapeutic value of these plants lies in bioactive phytochemical constituents that produce definite physiological action on the human body⁶. Plants contain secondary metabolites, also known as phytochemical, natural bioactive compounds, or plant constituents, that are not directly involved in an organism's normal growth, development, or reproduction but often play an important role in plant defence. Some of the most important bioactive phytochemical constituents are alkaloids, essential oils, flavonoids, tannins, terpenoids, saponins, phenolic compounds, and many more⁷. They are found in plants, such as vegetables, fruits, flowers, leaves, and roots, which work with nutrients and fibre to act as a defence against disease^{8,9}.

An antibacterial is a substance that kills or inhibits the growth of bacteria, and antimicrobial drugs either kill bacteria or prevent microbial growth. Plants have evolved to synthesize chemical compounds that help defend against attacks from predators such as insects, fungi, and herbivorous mammals. By chance, some of these compounds, although toxic to plant predators, have been shown to have beneficial effects in treating human diseases. Such secondary metabolites have a very diverse structure; many are aromatics, most of which are phenols or their oxygen-substituted derivatives. Many herbs and spices humans use to flavour food yield useful medicinal values¹⁰.

❖ *Plumeria obtusa*

- **Kingdom:** Plantae
- **Order:** *Gentianales*

- **Family:** *Apocynaceae*
- **Genus:** *Plumeria L.*
- **Species:** *Plumeria obtusa Linn.*



Figure 1: *Plumeria obtusa*

Plumeria obtusa commonly known as frangipani, West Indian jasmine, Champa, and temple tree, is used as folklore medicine to treat many inflammatory and infectious diseases^{11,12}. It is a flowering plant native to Central and South America, the Caribbean, Brazil, and India¹³. It belongs to the family *Apocynaceae*. Various bioactive constituents with potent antimicrobial, anti-inflammatory, anthelmintic, antipyretic, and antirheumatic properties have been extracted from the *Plumeria obtusa* bark, leaves, and flowers. The stem and leaf of *Plumeria obtusa* are used to treat skin disorders such as herpes, scabies, and ulcers^{14,15}.

Plumeria obtusa is effective against some common fungi linked with systemic fungal infections, such as *Aspergillus niger*, *Penicillium chrysogenum*, *Microsporium gypsum*, and *Epidermophyton floccosum*^{16,17}. *Plumeria obtusa* has good antibacterial properties against the *Staphylococcus aureus*, coagulase-negative *Staphylococci*, *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus vulgaris*, *Pseudomonas aeruginosa*, *Serratia marcescens*, *Salmonella typhi*, *Staphylococcus saprophyticus*, *Salmonella Para typhi*, and *Bacillus anthracis*^{18,19}. The essential oils from the flower of *Plumeria obtusa* are effective against *S. aureus*, *Bacillus subtilis*, *A. niger*, *Candida albicans* (Ca), and *P. chrysogenum*^{20,21}.

Plumeria obtusa is commonly grown for ornamental purposes. The plant is propagated through cuttings. It sets seed rarely in India. The plants raised from the seed showed a wide variation in character, evidently being different strains. *Plumeria obtusa* is a small tree 3.5-6.0 m high, commonly grown in gardens, and leaves lanceolate to obovate –oblong. Flowers are fragrant, generally red, pink or purple, centre rich with yellow. Flowers large in terminal 2-3 chitinous cymes, bracts many broad, deciduous, calyx small, glandular within, lobes wide, obtuse. Corolla salver-shaped, naked throat lobes overlapping to the lefty, rarely to the right. Stamen near the base of the tube. Seeds oblong or lanceolate²². Its broad, usually round-headed canopy is often about as wide as the tall tree. The species and hybrids vary in tree size, compactness, branching character, leaf and flower size and colour, and deciduousness^{23,24}.

It is a medicinal plant with a wide range of bioactive constituents with therapeutic properties that can treat malaria, diabetes, inflammation, hypertension, abdominal tumours, rheumatism, leprosy, skin cancer, and fungal, viral and bacterial infections^{24,25}.

1. Pharmacodynamics and Formulations:

Table 1: Pharmacodynamics and Formulations of *Plumeria obtusa*

| | <i>Plumeria obtusa</i> |
|------------------------------------|--|
| Ayurvedic Names | Kathgolop, Gulchin, Champa, Champeya, Hemapushpa, Champaka, Golenchi, Chafa |
| Regional Names | Hindi: Golenchi Tamil: Nela Sampangi Telugu: Deva garner Marathi: Champa |
| Important Phytoconstituents | Benzyl esters, aliphatic alkanes, oxygenated monoterpenes |

| | |
|-------------------------|---|
| Therapeutic Uses | Dravya – Substance, Rasa – Taste, Guna – Qualities, Veerya – Potency, Vipaka – Post-digestion effect, Karma – Pharmacological activity, Prabhava – Therapeutics |
| Dose | Flower – 5-10 gm, leaf powder/ paste – 3-5 gm |
| Formulations | Oils, Lotions, Powders, Fertilizers |

2. Nutrients and their Constituents:

Plumeria obtusa obtains carbon, hydrogen, and oxygen from the surrounding air and water. There are two groups of nutrients: macronutrients, required in large quantities and micronutrients, needed in small amounts. Nitrogen (N), phosphorus (P), and potassium (K) are the big three, together comprising over 75% of the mineral nutrients found in *Plumeria obtusa*. Calcium (Ca), Magnesium (Mg) and Sulphur (S) are also macronutrients. The micronutrients are Iron (Fe), Manganese (Mn), Zinc (Zn), Copper (Cu), Boron (B) and Chlorine (Cl)²⁶.

3. Traditional and Claimed Applications:

Plumeria obtusa fruit is reported to be eaten in West Indies. In India, however, it has been used as an abortifacient. Root bark- Drastic, Purgative, Blenorrhagia. Latex – used in toothache and for carious teeth. Flowers- Aromatic, Bechic and used as very popular pectoral syrup.

4. Therapeutic and Medicinal Characteristics:

Plumeria obtusa is reported to have antifertility, anti-inflammatory, antioxidant, hepatoprotective and antimicrobial activities. However, it has been used in the folk medicine systems of civilizations to treat abortifacient, drastic, purgative, and blenorrhagia, and used in toothache and carious teeth. The plant's different parts are traditionally used to treat various diseases like diabetes

mellitus, wounds and skin disease, and abortion, and also used in cosmetics, aromatherapy, necklaces and as offerings.

MATERIALS AND METHODS

• Preparation of Plant Material

The freshly collected leaves of *Plumeria obtusa* were carefully cleaned and plucked from the stem. The leaves were air-dried in an aerated room for 6 weeks. The dried leaves were pulverized using mortar and pestle into smaller particles and then blended to powder using an electric blender. 50g of the powdered leaves were obtained. 500ml of methanol was measured into the 50g of the powdered leaves and then stored in an air-tight container for 4 days.

• Preparation of Extract

50g of *Plumeria obtusa* leaves powder was subjected to extraction using methanol and water by shaking the mixture for about one hour in a shaker at room temperature. Subsequently, the samples were filtered on Whatman filter paper. Then the filtrate was taken for extraction and concentration in a Soxhlet apparatus using Soxhlet extraction. The final extract was collected in sterile-labelled containers.

• Preliminary Phytochemical Screening

Different extractions were stored in a refrigerator at 4°C to screen their phytochemical constituents and detect antibacterial activity. Phytochemical screening of the leaves extract was tested for phytochemicals by standard procedures. The methanol extract of the leaves was used for the preliminary phytochemical screening procedure for the presence of bioactive ingredients such as tannins, alkaloids, flavonoids, saponins, and steroids.

PHYSICOCHEMICAL EVALUATION

The existence of ash value and extractive value was determined using physicochemical testing.

• Ash that dissolves in water

The ash was boiled for 5 minutes with 25 ml of water, and then the soluble materials were

collected in a crucible, burned, and weighed. Concerning air-dried medication, we computed the percentage of water-soluble ash as follows.

• Insoluble acid ash

The residue is formed by burning the residual insoluble materials after boiling the entire ash with weak hydrochloric acid. Using 25 ccs of dilute hydrochloric acid, simmer the ash for 5–10 minutes, collect the insoluble materials in a crucible or ashless filter paper, ignite, and weigh. Concerning the air-dried medication, we estimated the percentage production of acid-insoluble ash as follows²⁷.

• Extractable value in water

In a closed flask, stirring frequently, 1 g of coarsely ground air-dried medication was macerated with 100 ml of distilled water for 24 hours. The filtered solution was evaporated in a tarred flat bottom shallow dish, dried at 100°C, and weighed. The proportion of water-soluble extractives was calculated concerning the air-dried drugs²⁸.

• Value of alcohol-soluble extractive

In a stoppered flask, macerate 5 g of carefully weighed, coarsely powdered medication with 100 ml of alcohol (90 per cent v/v) for 24 hours, stirring frequently during the first 6 hours. Filtered quickly via filter paper to avoid excessive alcohol loss. In a covered dish, evaporate 25 mL of alcoholic extract to dryness and weigh it. We used the formula below to determine the percentage w/w of alcohol-soluble extractive concerning the air-dried medication. Extractive value in per cent alcohol-soluble=4 (Wt. of residue) Based on the comparison of Petroleum ether and methanolic extracts in the table above, the methanolic section had a higher ash value and extractive value than the Petroleum ether extract.

SOXHLET EXTRACTION





Figure 3: Assembly of Soxhlet Extraction

Principle

Extraction is performed in the laboratory using a fat extractor (Soxhlet extractor). The fat extractor employs the solvent reflux and syphon concept to continually extract solid matter from a pure solvent, increasing solvent extraction efficiency. Before extraction, the solid material is crushed to enhance the solid-liquid contact area. After that, the solid material is placed in a filter paper container and extracted. The bottom end of the extractor is attached to a reflux condenser and a round bottom flask containing a solvent. The solvent is boiled in the bottom flask, and the vapour rises through the extractor's branch pipe, condenses, and falls into the extractor, where the solvent is contacted with the solid for extraction. The solvent holding the extract is syphoned back when the solvent surface surpasses the highest point of the syphon. The flask is repeated, removing a portion of the material and concentrating it in the flask.

PHYTOCHEMICAL ANALYSIS

The presence and absence of primary and secondary metabolites such as starch, tannins, oil, mucilage, and lignin were revealed by histochemical examination using various reagents. Air-dried powdered leaves' identity, purity, and strength were tested using physicochemical methods, including ash and extractive values. The phytochemical screening of chemical constituents of plants in various solvents studied,

such as Methanol, Petroleum ether and aqueous extracts, revealed that alkaloids, glycosides, flavonoids, phenols, steroids, proteins, and amino acids are present in all sections, but the amounts vary.

RESULTS AND DISCUSSION

Table 6.1: Physicochemical Analysis of powdered drug of *Plumeria obtusa*

| Name of Test | Extractive Ash Value (gm) | Acid Insoluble Ash Value (gm) |
|------------------------|---------------------------|-------------------------------|
| Weight of empty dish | 60.70 | 60.70 |
| Weight of powder taken | 0.25 | 0.18 |
| Weight of ash + dish | 60.95 | 60.88 |
| Percentage of ash | 99.58 | 99.70 |

Table 6.2: Phytochemical Tests of *Plumeria obtusa*

| Sr. No. | Ingredients | <i>Plumeria obtusa</i> |
|---------|---|------------------------|
| 1. | Carbohydrates: Molisch Test | + |
| 2. | Monosaccharides: Barfoed's Test | - |
| 3. | Pentose Sugars | + |
| 4. | Hexose Sugars: Selwinoff's Test | + |
| 5. | Non-reducing Polysaccharides - Starch: Iodine Test | - |
| 6. | Proteins: Millon's Test | + |
| 7. | Amino Acids: Tyrosine Test | - |
| 8. | Fats and Oil: Saponification Test | + |
| 10. | Steroids: Salkowski Test | + |
| 11. | Glycosides: Bontrager's Test | + |
| 12. | Flavonoids: Sulphuric Acid Test | + |
| 13. | Alkaloids: Mayer's Test | - |

Calculation of the extracting value

Extractable value in water in a closed flask, 1 g of coarsely ground air-dried medication was macerated with 100 ml of distilled water for 24 hours, stirring frequently. The filtered solution was evaporated in a tarred flat bottom shallow dish, dried at 100°C, and weighed. The percentage of water-soluble extractives was estimated using air-dried medicines as a reference.

ANTIMICROBIAL ACTIVITY

Principle

Natural products (secondary metabolites) are the major sources of drugs, and these products have greater structural diversity than drugs or

compounds from standard combinatorial chemistry. The use of medicinal plants to treat microbial diseases has been well-known and well-documented since ancient times. Medicinal plants synthesize many defensive compounds to protect themselves and predators, and these compounds have antimicrobial activity. The ability of the plant extract to reduce or inhibit the growth of microorganisms or kill pathogenic organisms is known as antimicrobial activity or efficacy. Several plant species have been the antimicrobial activity of plant extracts that may be tested by agar diffusion or cup-plate methods.



Figure 6.1: Zone of Inhibition of *E. coli*



Figure 6.2: Zone of Inhibition of *S. aureus*

Table 6.3: Antimicrobial activity, Zone of Inhibition of *E. coli*

| Concentration($\mu\text{g/ml}$) | Zone of Inhibition (cm) |
|-----------------------------------|-------------------------|
| 50 | 0.4 |
| 100 | 0.5 |
| 200 | 0.7 |
| STD (Streptomycin) | 0.6 |

Table 6.4: Antimicrobial activity, Zone of Inhibition of *S. aureus*

| Concentration($\mu\text{g/ml}$) | Zone of Inhibition (cm) |
|-----------------------------------|-------------------------|
| 50 | 0.4 |
| 100 | 0.5 |
| 200 | 0.9 |
| STD (Streptomycin) | 0.8 |

CONCLUSIONS

Extraction, Phytochemical investigation and antimicrobial activity of *Plumeria obtusa* are executed. In Phytochemical analysis, Alkaloids, Carbohydrates, Glycosides, Steroids, Proteins, and Amino acids are present in *Plumeria obtusa*.

In Antimicrobial activity, *E. coli* and *S. aureus* show significant inhibition of growth in different concentrations in which *E. coli* is more potent than *S. aureus*.

FUTURE PROSPECTS

Plumeria obtusa. (*Apocynaceae*) is a deciduous, ornamental, tropical plant grown in premises, parks, gardens, and graveyards because of its attractive and fragrant flowers. The plant's different parts are traditionally used to treat diseases like diabetes mellitus, wounds and skin

disease, diuretics, purgative, and abortion. They are also used in cosmetics, aromatherapy, necklaces and as offerings.

The toxicological studies of *Plumeria obtusa* need to be completed and paid more attention to. A couple of pharmacological activities are proven by using higher doses of plant extracts. So, the studies are essential to investigate the long-term *in vivo* toxicity of the plant. Further relation systemic toxicity and safety evaluation studies are needed to be studied. We hope that the information discussed or provided here could make people more aware of the plant *Plumeria obtusa* and can be beneficial for further research.

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