

Phytochemical Characterization and Antibacterial Activity of a Hydroalcoholic Extract of *Lagerstroemia Speciosa* Leaves

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ABSTRACT

The present study investigated the antimicrobial potential of the hydroalcoholic leaf extract of *Lagerstroemia speciosa* (LSLE) against representative Gram-positive and Gram-negative bacterial strains. Antibacterial activity was evaluated by the well diffusion method using *Escherichia coli* (NCIM 2832) and *Bacillus subtilis* (ATCC 6051), with streptomycin serving as the standard reference. LSLE exhibited moderate activity, producing inhibition zones of 15 mm against *E. coli* and 9 mm against *B. subtilis*, compared with 20 mm and 19 mm, respectively, for streptomycin. Preliminary phytochemical screening confirmed the presence of flavonoids and alkaloids, while spectroscopic analyses (FTIR and UV-Vis) indicated functional groups characteristic of bioactive polyphenols. These findings suggest that *L. speciosa* leaves contain phytoconstituents with antibacterial activity. This has implications as a natural source for the development of complementary antimicrobial agents.

Keywords: *Lagerstroemia speciosa*; Antimicrobial activity; Hydroalcoholic extract; *Escherichia coli*; *Bacillus subtilis*; Phytochemicals

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1. INTRODUCTION

The antimicrobial resistance (AMR) is a major global health concern. The reduced effectiveness of antibiotics against resistant pathogens complicates infection treatment, increasing mortality and healthcare costs. Resistant strains such as vancomycin-resistant enterococci, methicillin-resistant *Staphylococcus aureus* (MRSA), and multidrug-resistant Gram-negative organisms are particularly worrisome [1], [2]. In developing regions, bacterial infections still account for significant mortality [3]. Biofilm formation further protects bacteria from conventional antibiotics, leading to hospital-acquired and device-associated infections [4], [5], [6]. These challenges underscore the urgent need for new antimicrobial options.

Lagerstroemia speciosa (L.) Pers., commonly known as Banaba or Jarul, is a deciduous tree in the family Lythraceae, distributed across tropical Asia, including India, the Philippines, and Malaysia. Its leaves are widely

used in Southeast Asian traditional medicine, particularly for managing diabetes [7]. Scientific studies have confirmed a range of pharmacological activities in this plant. Extracts and isolated compounds have shown antioxidant [8], anti-obesity [9], anti-gout [10], anti-inflammatory [8], antiviral [11], and antidiabetic effects [12]. Corosolic acid, a triterpenoid in the leaves, exhibits hypoglycemic action [13], while ethyl acetate extracts protect against cisplatin-induced nephrotoxicity in animal models [14]. Phytochemical studies report proteins, amino acids, tannins, and triterpenoids [7], and chemical analyses reveal ellagitannins, ellagic acid, gallic acid, caffeic acid, p-coumaric acid, kaempferol, quercetin, and isoquercitrin [15], many of which are known for their antioxidant and antimicrobial potential.

Evidence suggests antibacterial activity of *L. speciosa* leaves. Leaf powder extracts have been tested against *Staphylococcus aureus*, *Bacillus subtilis*, *Pseudomonas aeruginosa*, and *Escherichia coli*, with ampicillin as the

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reference standard [16]. Singh et al. [17] reported antibacterial activity, including inhibition of quorum sensing in *P. aeruginosa*. Ragasa et al. [18] identified bioactive compounds such as sitosterol, ellagic acid, campesterol, and stigmasterol, further supporting therapeutic value. However, compared with extensive studies on antidiabetic and antioxidant effects, research on antibacterial activity remains limited.

The rising threat of AMR emphasizes the importance of exploring such plants. WHO reports indicate resistance to third-generation cephalosporins in *E. coli* exceeding 50% in several regions [19], highlighting the need for alternative strategies. Studies on natural products, including essential oils and resin acids, show that functional groups such as hydroxyl and carbonyl contribute to antimicrobial activity [20], [21]. These findings underline the importance of combining phytochemical characterization with antibacterial screening. Techniques like Fourier transform infrared (FTIR) and ultraviolet-visible (UV-Vis) spectroscopy allow functional group analysis, strengthening links between phytochemical composition and biological effects [22].

Although *L. speciosa* has been extensively studied for metabolic disorders, very few works focus on its antibacterial properties, particularly hydroalcoholic leaf extracts. Studies integrating phytochemical screening with spectral characterization and antibacterial testing are scarce. The present study was therefore designed to prepare a hydroalcoholic extract of *L. speciosa* leaves, perform preliminary phytochemical screening and FTIR and UV-Vis analysis, and evaluate antibacterial activity against *E. coli* (NCIM 2832) and *B. subtilis* (ATCC 6051). By combining chemical profiling with antimicrobial assays, this work aims to provide preliminary evidence guiding further studies on bioactive fractionation and identification of active constituents.

2. MATERIALS AND METHODS

2.1. Plant material collection and identification

Fresh leaves of *Lagerstroemia speciosa* L. were collected from Undale, Tal. Karad, District Satara, Maharashtra, India. The species was identified and authenticated at the Department of Pharmacognosy, Shree Sant Krupa College of Pharmacy, Ghogaon, Tal. Karad, Satara, and further confirmed at Sant Gadgamaraj College, Karad, Maharashtra. The voucher specimen was deposited in the herbarium for future reference.

2.2. Preparation of plant extract

Collected leaves were thoroughly washed with distilled water, shade-dried, and powdered using an electronic grinder. About 60 g of dried leaf powder was subjected to maceration with methanol:water (1:3 v/v) as solvent. The

mixture was left at room temperature with intermittent shaking, filtered, and concentrated to obtain the hydroalcoholic extract, which was stored in airtight containers at 4 °C until use.

2.3. Preliminary phytochemical screening

The crude extract was subjected to qualitative phytochemical analysis using standard protocols. Tests were performed for major classes of secondary metabolites, including alkaloids (Wagner's test), flavonoids (Shinoda test), tannins (ferric chloride test), phenolic compounds, saponins (froth test), glycosides, proteins, and carbohydrates. Results were recorded as presence (+) or absence (-).

2.4. Spectroscopic characterization

FTIR spectra of the extract were recorded using a Bruker ALPHA-II spectrophotometer in the range of 4000–400 cm^{-1} to identify major functional groups. UV-Visible absorption spectra were obtained using a Shimadzu UV-1900 spectrophotometer in the range of 200–400 nm.

2.5. Microorganisms and inoculum preparation

Two bacterial strains were used in this study: *Escherichia coli* (NCIM 2832) and *Bacillus subtilis* (ATCC 6051). Test cultures were revived from stock and maintained on nutrient agar slants at 4 °C. For assays, fresh bacterial suspensions were prepared in nutrient broth.

2.6. Antibacterial assay

The antibacterial activity of the hydroalcoholic extract was evaluated by the agar well diffusion method [19]. Sterile nutrient agar plates (15 mL) were prepared, and 100 μL of bacterial inoculum was spread evenly onto the agar surface. Wells of 6 mm diameter were punched aseptically, and 100 μL of the plant extract solution (100 $\mu\text{L}/\text{mL}$ in DMSO) was added to each well. Streptomycin (1 mg/mL) served as a positive control, and DMSO was used as a negative control. Plates were incubated at 37 °C for 24 h, after which the diameters of inhibition zones were measured in millimeters. All experiments were carried out in triplicate.

2.7. Statistical analysis

Data obtained from all experiments were expressed as mean \pm standard deviation (SD). Statistical comparisons between treated and control groups were made using Student's *t*-test, and differences were considered statistically significant at $p < 0.05$.

3. RESULTS

3.1. Phytochemical screening

Qualitative phytochemical analysis of the hydroalcoholic extract of *Lagerstroemia speciosa* leaves (LSLE) revealed the presence of flavonoids and alkaloids, while tests for carbohydrates, starch, proteins, phenolics, glycosides, and tannins were negative (Table 1).

Table 1: Phytochemical screening of LSLE

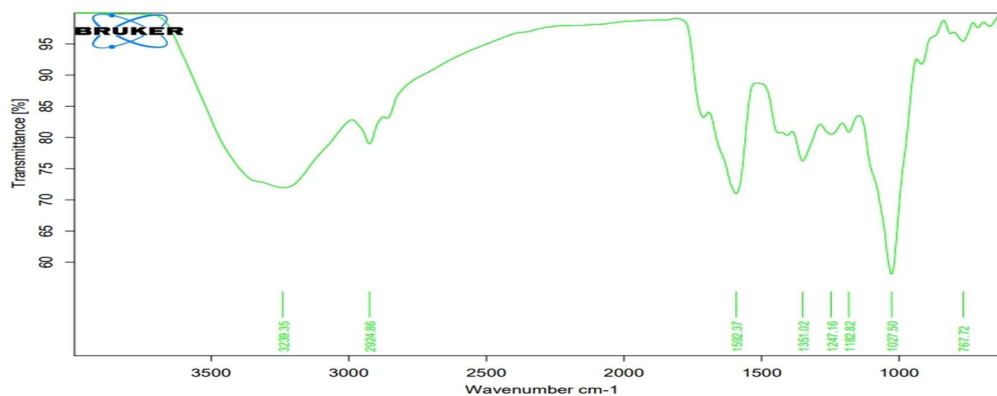
Phytochemical group	Tests performed	Result	Inference
Carbohydrates	Molisch, Benedict, Barfoed, Fehling	-	Absent
Starch	Iodine test	-	Absent
Proteins	Millon, Xanthoproteic	-	Absent
Flavonoids	Sulfuric acid, Lead acetate, Alkaline reagent	+	Present
Phenolic compounds	Lead acetate, Gelatin	-	Absent
Alkaloids	Wagner, Hager, Dragendorff	+	Present
Glycosides	Borntrager, Keller-Killiani, Legal's	-	Absent
Tannins	Ferric chloride, Bromine water	-	Absent

(+ present, - absent)

3.2. FTIR spectroscopy

The FTIR spectrum of LSLE showed characteristic absorption peaks at 3239.35, 2924.86, 1592.37, 1247.16,

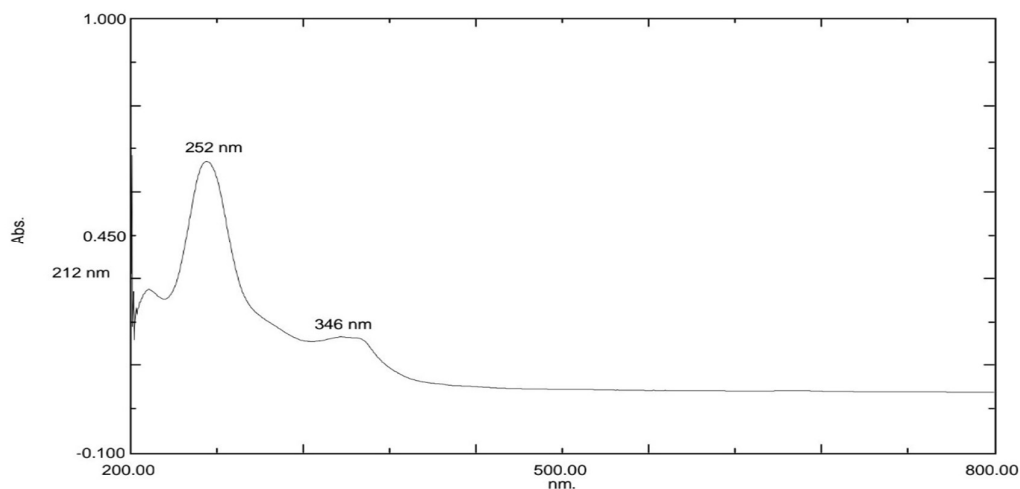
and 1027.50 cm^{-1} (Figure 1). These correspond to carboxylic acids, C-H groups, diketones, ester carbonyl groups, and C-O stretching/O-H deformation, respectively, indicating the presence of functional groups typical of flavonoids and alkaloids.



3.3. UV-Visible spectroscopy

The UV-Vis spectrum of LSLE revealed three major absorption peaks at 346.5 nm ($A=0.192$), 252.5 nm

($A=0.638$), and 212.5 nm ($A=0.312$) (Figure 2). The absorption maxima suggest the presence of conjugated systems, consistent with polyphenolic constituents.



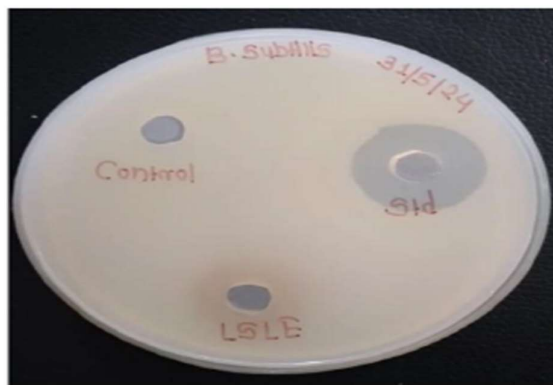
3.4. Antibacterial activity

The hydroalcoholic extract exhibited antibacterial activity against both *Escherichia coli* and *Bacillus subtilis* in the agar well diffusion assay (Table 2). Against *E. coli*, LSLE produced a zone of inhibition of 15 mm, compared to 20

mm for streptomycin (Figure 3A). Against *B. subtilis*, LSLE produced a 9 mm zone, compared to 19 mm for streptomycin (Figure 3B). No inhibition was observed with the DMSO control.

Table 2: Antibacterial activity of LSLE compared with streptomycin

Organism	Control (mm)	LSLE (mm)	Streptomycin (mm)
<i>Escherichia coli</i>	0	15	20
<i>Bacillus subtilis</i>	0	9	19



4. DISCUSSION

The present study evaluated the phytochemical profile, spectral characteristics, and antibacterial potential of a hydroalcoholic extract of *Lagerstroemia speciosa* leaves (LSLE). Qualitative screening indicated the presence of flavonoids and alkaloids, while carbohydrates, proteins, tannins, phenolics, and glycosides were absent. These findings are consistent with earlier phytochemical reports on *L. speciosa*, which highlighted flavonoids, ellagitannins, and triterpenoids as the predominant classes of metabolites [7], [15], [20]. Such compounds are widely recognized for their antimicrobial and antioxidant activities, suggesting a plausible role in the activity observed in this study [8], [22].

The FTIR and UV-Vis spectra further supported the presence of functional groups typical of polyphenols and alkaloids. Peaks at 3239 cm^{-1} (carboxylic acids) and 1592 cm^{-1} (diketones) correspond well with polyphenolic structures, while absorption maxima at 346.5, 252.5, and 212.5 nm in the UV spectrum are consistent with conjugated systems commonly found in flavonoids. Similar associations between FTIR/UV bands and bioactive phytoconstituents have been reported in prior studies of medicinal plants [22], [23]. These data suggest that flavonoid- and alkaloid-rich fractions may be largely responsible for the antibacterial activity of LSLE.

In antibacterial assays, LSLE produced inhibition zones of 15 mm against *E. coli* and 9 mm against *B. subtilis*. While these values were lower than those of the reference drug streptomycin (20 mm and 19 mm, respectively), they clearly indicate moderate antibacterial activity. Earlier investigations also reported inhibition of Gram-positive and Gram-negative organisms by *L. speciosa* extracts, including effects against *S. aureus*, *P. aeruginosa*, and *E. coli* [16], [17]. Singh et al. [17] additionally demonstrated

quorum-sensing inhibition in *P. aeruginosa*, suggesting that the antibacterial action of *L. speciosa* may involve multiple mechanisms beyond direct growth inhibition.

The relatively greater inhibition observed against *E. coli* compared with *B. subtilis* in this study indicates that the hydroalcoholic extract may be more effective against Gram-negative organisms. This is notable, as Gram-negative bacteria generally possess a more complex cell wall structure that reduces susceptibility to antimicrobials. The activity against both bacterial types nevertheless demonstrates the broad potential of LSLE as a source of antibacterial compounds.

Despite these encouraging results, certain limitations should be acknowledged. The study employed only two bacterial strains and a single concentration of crude extract, without determination of minimum inhibitory concentration (MIC) or minimum bactericidal concentration (MBC). Moreover, the extract was not fractionated to identify specific compounds responsible for the activity. Future work should therefore focus on systematic fractionation, quantitative antimicrobial assays, and broader microbial panels, including clinically relevant multidrug-resistant strains. Additionally, testing for synergistic interactions with existing antibiotics may be worthwhile, given the growing importance of combination therapies in addressing antimicrobial resistance.

Overall, the results strengthen the pharmacological profile of *L. speciosa*, adding preliminary antibacterial data to its well-documented antidiabetic and antioxidant properties. The moderate zones of inhibition obtained suggest that although LSLE may not match conventional antibiotics in potency, it represents a promising source of complementary antimicrobial agents, particularly when further purified or optimized.

5. CONCLUSION

This study demonstrated that the hydroalcoholic leaf extract of *Lagerstroemia speciosa* (LSLE) contains flavonoids and alkaloids and exhibits moderate antibacterial activity. Spectroscopic characterization supported the presence of functional groups associated with bioactive phytochemicals. In vitro assays revealed that LSLLE inhibited *Escherichia coli* (15 mm zone) and *Bacillus subtilis* (9 mm zone), though activity was lower than the reference antibiotic streptomycin. These findings confirm the potential of *L. speciosa* leaves as a source of antimicrobial agents and warrant further studies, including MIC/MBC determination, bioassay-guided fractionation, and evaluation against resistant bacterial strains.

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