



RESEARCH ARTICLE

Unravelling the bioactive potential: Phytochemical and antimicrobial potential of leaf and bark extracts of *Pterocarpus dalbergioides* Roxb. ex DC. (Andaman Padauk)

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Abstract

Therapeutic herbs are recognized as valuable sources of bioactives that are utilized in pharmaceutical research. *Pterocarpus dalbergioides* Roxb. ex DC., commonly called as Andaman padauk or narra, is a medicinal tree endemic to the Andaman Islands, renowned for its therapeutic properties against various health conditions. In this study, phytochemical constituents were quantitatively analysed through Gas Chromatography- Mass Spectrometry (GC-MS) and antibacterial activity was evaluated. The GC-MS analysis indicated a presence of about 100 metabolites with major metabolites including bis(2-ethylhexyl) phthalate, alpha-methyl mannofuranoside, stigmasterol and dotriacontane. The antibacterial assay revealed inhibition zones ranging from 10-13mm and 8-11mm against *Staphylococcus aureus*, *Escherichia coli* respectively, with the bark extract of *P. dalbergioides* while the leaf extract showed inhibition zones of 8-9mm. This pioneering research on the endemic species provides valuable information for further pharmacological research.

Keywords: Andaman; antimicrobial activity; GC-MS; metabolites; *P. dalbergioides*

Introduction

Traditional tree-based medicines have witnessed a surge in global interest over the past decade and have been widely acknowledged for their essential role in supporting human health and improving quality of life. It is estimated that nearly one-quarter of the global population, approximately 1.42 billion people, relies on traditional remedies to address various health issues (1). Phytochemicals such as alkaloids, corticosteroids, tannins and phenolic compounds, which are naturally synthesized and stored in specific plant parts, exhibit significant pharmacological properties. These bioactive molecules play a crucial role in the development of new therapeutic agents, offering promising potential for drug discovery (2). In addition to the role as protective agents for plants against microbial infections and pest infestations, many endangered species and numerous plants in nature remain largely unexplored for their medicinal potential. The genus *Pterocarpus*, consisting of approximately 35 to 46 species, has been traditionally utilized in herbal medicine for treating various health conditions, including malaria, gonorrhea, diarrhea, mouth ulcers and skin disorders (3).

P. dalbergioides, widely recognized as Andaman Padauk, is a large evergreen tree native to the Andaman Islands and belongs to the Fabaceae family. It occurs throughout both the southern and northern parts of the islands. In Ayurveda, it is referred to as Vijaysar and has been relied upon for treating diverse medical conditions. The alkaline substance derived from the water-soluble ash of *P. dalbergioides* is used to treat haemoptysis (Raktapitta), while a decoction of its heartwood is reported to be beneficial in treating filariasis (Slipada), obesity (Sthoulya and *Diabetes mellitus* (Madhumeha). Additionally, it helps manage symptoms such as reduced urine quality, complications associated with hyperglycemia and regulates blood sugar levels (4). Another study demonstrated the chemical profile, *in vivo* effects and anti-inflammatory properties of *P. dalbergioides* fruit extract (3).

In this study, we assess the phytochemical composition of bark and leaf extracts of *P. dalbergioides* and evaluate their antimicrobial efficacy.

Materials and Methods

Crude extraction from the leaf and bark of *P. dalbergioides* was conducted at the Department of Medicinal and Aromatic Crops, Horticultural College and Research Institute, Coimbatore. Phytochemical profiling of *P. dalbergioides* and its antimicrobial activity were carried out at the Department of Plant Biotechnology, Centre for Plant Molecular Biology and Biotechnology, Tamil Nadu Agricultural University, Coimbatore and in the Department of Plant Pathology, Tamil Nadu Agricultural University, Coimbatore respectively.

Plant material procurement and identification

Bark & leaf samples of *P. dalbergioides* were obtained from the Central Island Agricultural Research Institute, Andaman and Nicobar Islands, India. The study was conducted in December 2023.

Preparation of plant material

The collected plant samples were meticulously washed and dried in a shaded area to remove moisture. The leaves and barks were finely ground using an electric mortar and the resulting powder was carefully sieved to remove any coarse particles. The fine, uniformly pulverized substance was then securely stored in a vacuum-sealed canister for subsequent use.

Preparation of crude extract

The air-dried bark and leaf powder (100g) were successively extracted using the Hot Soxhlet extraction with two solvents of different polarity i.e. methanol and hexane. The collected crude extracts were further concentrated using rotary evaporator (Fig. 1).

Gas Chromatography- Mass Spectrum

The analysis was conducted using a GC-MS instrument that featured a silica-capillary non-polar Rxi-5 Sil MS column, 30 m long, with an internal diameter of 0.25 mm and a film thickness of 0.25 μ m. Helium was used as the carrier gas at a flow rate of 0.3 mL/min, with an injection volume of 1 μ L at a temperature of 240 °C. The oven temperature was initially set at 70 °C for 8 min and was then increased gradually at a rate of 2 °C/ min until reaching 250 °C, where it was maintained for 15 min. The chemical components were identified by matching the mass spectra with those in the National Institute of Standards and Technology (NIST) library database.

Determination of antibacterial assay

Inoculum Preparation: Three to five distinct colonies exhibiting identical morphologies were chosen from the agar plate. The

top of each colony was gently contacted by a loop and the bacterial cells were transferred into a tube containing 4–5 mL of Nutrient-rich broth or another suitable broth type. The broth culture was incubated at 35 °C for 2 to 6 hr, or until it reached or exceeded the required turbidity. To achieve the desired turbidity, sterile saline or broth was added to the rapidly growing broth culture. This procedure produced a suspension with an estimated concentration of about $1-2 \times 10^8$ CFU/mL for both *E. coli* (Gram -ve) and *S. aureus* (Gram +ve), following the guidelines put forth by the Kirby-Bauer (5).

Inoculation of Test Plates: An inoculated cotton swab was ideally inserted into the modified suspension for 15 min. After rotating the swab a few times, it was pressed firmly against the tube's interior well, above the fluid level to remove excess inoculum. The dried nutrient agar plate was inoculated by swabbing the entire sterile surface. To ensure uniform distribution of the inoculum, this process was repeated twice, rotating the plates approximately 60 ° between each streaking. The final step involved swabbing the rim of the agar. For 3-5 min, the lid may be shaken prior to placing the drug-impregnated disks, no longer than 15 min, to facilitate the absorption of any surplus surface moisture. A 6 mm diameter well was made in the media, which was filled with 20 -100 μ L of the sample. In addition, the petri dishes were then placed inversely in order to promote complete diffusion and zones of inhibition were assessed by determining the diameter (mm) that developed surrounding the well following a 24 hr of incubation at 37 °C. A standard (Hi-Media) scale was used to measure the zones.

Results and Discussion

Gas Chromatography-Mass Spectrum

Hexane and methanol extracts of *P. dalbergioides* bark and leaves were analyzed using GC-MS, which identified several metabolites, as detailed in Table 1. The respective GC-MS chromatograms are illustrated in Fig. 2(a-d). The predominant compounds detected were α -methyl mannofuranoside (26.91 %), stigmasterol (10.78 %), bis(2-ethylhexyl) phthalate (56.71 %), dotriacontane (13.47 %), γ -sitosterol (8.11 %), 1-triacontanol, tert-butyldimethylsilyl (TBDMS) derivative (7.81 %), glyoxylic acid-meto-TMS (7.18 %), methyl myristoleate (6.74 %), N-butyrylglycine-TMS (6.09 %), dihydrophytol, trimethylsilyl (TMS) derivative (5.79 %), neophytadiene (5.55 %), margaric acid-TMS (5.02 %) and phytol (4.66 %), as shown in

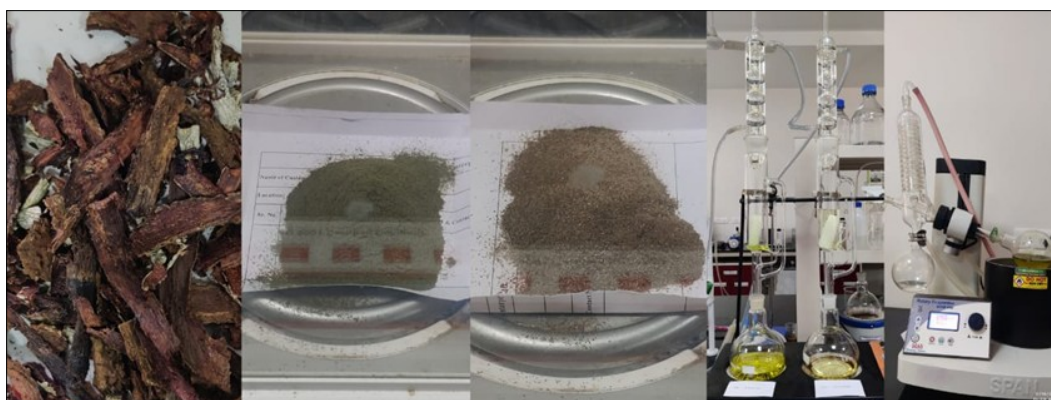


Fig. 1. Preparation of extracts from leaves and barks of *P. dalbergioides*.

Table 1. Bioactive compounds identified in hexane and methanolic extracts of the leaves and bark of *P. dalbergioides*

S. No	Compounds	<i>P. dalbergioides</i> (Methanol)		<i>P. dalbergioides</i> (Hexane)	
		Area %		Area %	
		Bark	Leaf	Bark	Leaf
1.	alpha.-Methyl mannofuranoside	2.6	26.91	-	-
2.	Hexadecanoic acid, methyl ester	0.71	0.67	-	-
3.	Phytol	0.54	4.66	-	1.22
4.	2-Furoic acid-TMS	0.69	-	-	-
5.	Squalene	1.76	-	1.3	-
6.	Valproic acid-TMS	0.67	-	-	-
7.	Arachidonic acid-TMS	1.06	-	-	-
8.	Glycolic acid-2TMS	0.55	-	-	2.11
9.	Octacosanol	2.6	-	-	-
10.	dl-.alpha.-Tocopherol	3.17	1.34	-	-
11.	Methyl margarate	0.99	-	-	-
12.	Arachidonic acid-TMS	1.09	-	-	-
13.	Elaidic acid-TMS	2.41	-	-	-
14.	7-Hydroxooctanoic acid-2TMS	1.91	-	-	-
15.	Margaric acid-TMS	5.02	-	-	-
16.	Azelaic acid-2TMS	1.72	-	2.45	-
17.	Stigmasterol	10.78	2.39	-	1.68
18.	Methyl myristoleate	6.74	0.73	-	-
19.	Phetyl dodecanoate	4.76	-	-	-
20.	gamma.-Sitosterol	8.11	2.06	-	-
21.	Linoleic acid-TMS	2.51	-	-	-
22.	Methyl elaidate	2.12	-	-	-
23.	Methyl eicosa-8,11,14-trienoate	2.86	-	-	-
24.	Dimethyl(bis[(4,8,8-trimethyldecahydro-1,4-me	5.14	-	-	-
25.	9,19-Cyclolanostan-3-ol, acetate, (3.beta.)-	3.21	-	-	-
26.	Lup-20(29)-en-3-ol, acetate, (3.beta.)-	8.5	3.16	-	-
27.	3-Hydroxybenzoic acid-2TMS	1.14	-	-	-
28.	Cholest-4-en-3-one	1.4	-	-	-
29.	Glyoxylic acid-meto-TMS	7.18	-	-	-
30.	N-Butyrylglycine-TMS	6.09	-	-	-
31.	Neophytadiene	5.55	-	-	0.75
32.	3,7,11,15-Tetramethylhexadec-2-ene	1.22	-	-	-
33.	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	1.83	-	-	-
34.	Hexanoic acid, undecyl ester	0.58	-	-	-
35.	2,6,10,15,19,23-Pentamethyl-2,6,18,22-tetracosatetraen-10,15-diol	4.67	-	-	-
36.	Hexacontane	1.01	-	-	-
37.	gamma-Tocopherol	0.67	-	-	-
38.	Tetrapentacontane	0.7	-	0.89	1.17
39.	Lup-20(29)-en-3-one	8.94	-	2.39	2.8
40.	Epilupeol	3.38	-	-	-
41.	Palmitic acid-TMS	-	-	1.13	1.85
42.	Stearic acid-TMS	-	-	-	0.86
43.	Cholesterol-TMS	-	-	0.99	-
44.	Lupeol	-	-	0.72	0.95
45.	Hexadecanoic acid, butyl ester	-	-	0.62	-
46.	Bis(2-ethylhexyl) phthalate	-	-	56.71	35.15
47.	Dotriacontane	-	-	-	13.47
48.	Tetracontane-1,40-diol	-	-	1.14	-
49.	Heptacosanal	-	-	1.16	-
50.	1-Octacosanol, TMS derivative	-	-	-	1.32
51.	Stigmast-5-ene, 3.beta.-(trimethylsiloxy)-, (24S)	-	-	-	3.85
52.	Dihydrophytol, TMS derivative	-	-	-	5.79
53.	Betulin	-	-	-	1.83
54.	Tetracontane	-	-	0.83	-
55.	Silane, trimethyl(tetratriacontyloxy)-	-	-	2.12	-
56.	1-Triacontanol, TBDMS derivative	-	-	7.81	-
57.	Methyl cis-13,16-Docosadienate	-	-	1.12	-
58.	Phenol, 2,4-bis(1,1-dimethylethyl)-, phosphite	-	-	1.28	-
59.	Dodecanedioic acid-2TMS	-	-	1.07	-
60.	Methyl linolelaidate	-	-	1.69	-

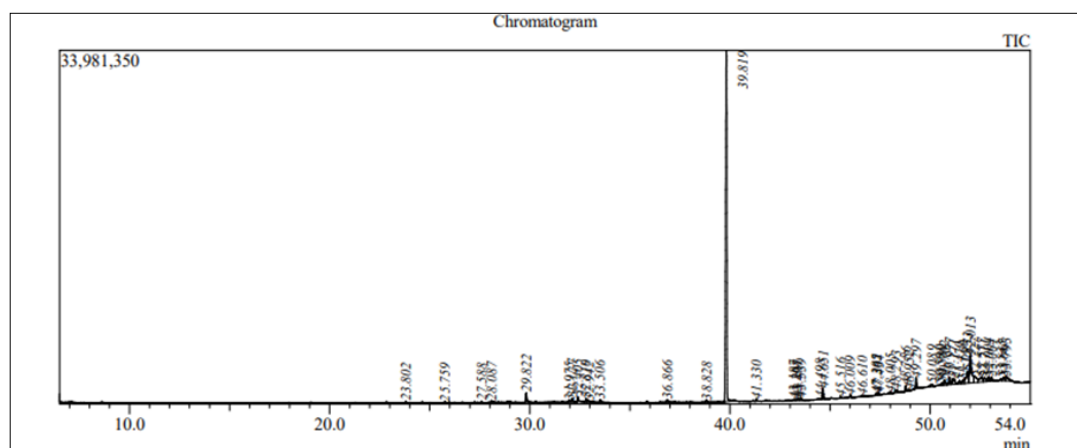


Fig. 2. (a) The chromatogram of bark extract from *P. dalbergioides* (He).

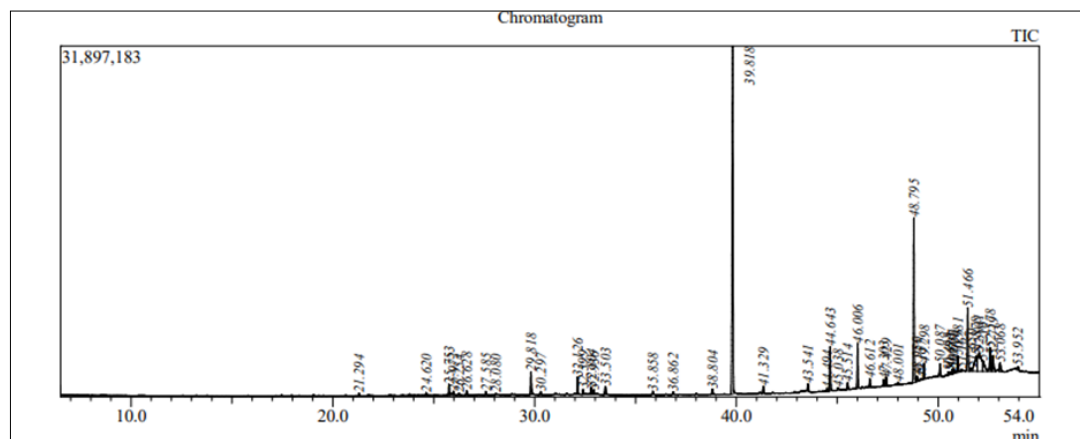


Fig. 2. (b) The chromatogram of leaf extract from *P. dalbergioides* (He).

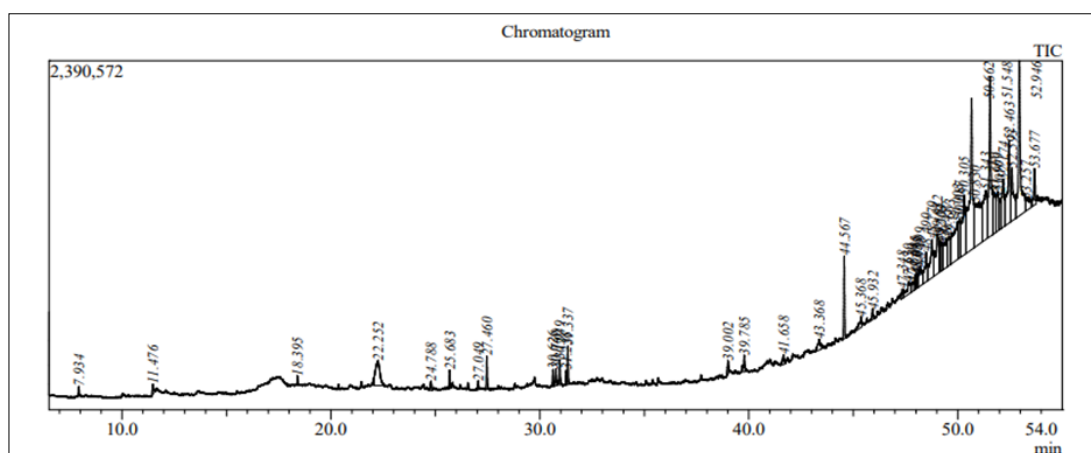


Fig. 2. (c) The chromatogram of bark extract from *P. dalbergioides* (Me).

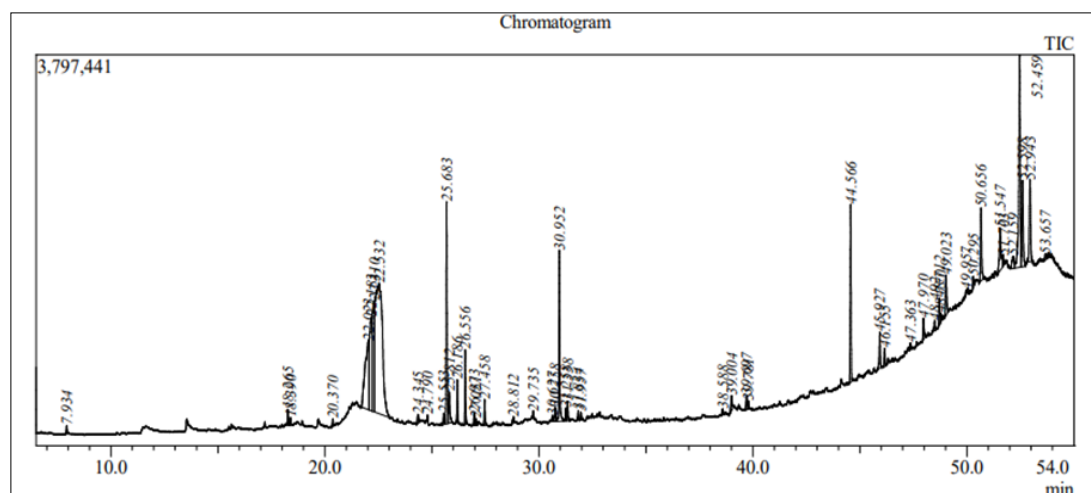


Fig. 2. (d) The chromatogram of leaf extract from *P. dalbergioides* (Me).

Fig. 3. The biological activity and chemical structures are presented in Table 2. Additionally, several compounds identified in the methanolic extract of *P. dalbergioides* fruit, include heptadecanoic acid, 3,4,9-trimethoxypterocarpan, 5-hydroxysalicylic acid and 3,4,5-trihydroxybenzoic acid (3).

Stigmasterol, a plant sterol abundant in various medicinal plants, is associated with multiple therapeutic effects, including anti-osteoarthritic, anti-inflammatory, anticancer, immunomodulatory, neuroprotective, antimicrobial, antidiabetic and antiparasitic activities (6). Its broad spectrum of biological activities suggests that it may contribute significantly to the anti-inflammatory and immune-supportive properties attributed to *P. dalbergioides* in traditional Ayurvedic medicine. Bis(2-ethylhexyl) phthalate, although predominantly used industrial applications, has been reported to possess antimicrobial, larvicidal, leukemia-preventive and mutagen-inhibiting effects (7, 8).

Dotriacontane, a long-chain alkane, has been identified in several plant species and contributes to their antioxidant and antimicrobial properties (9). Lup-20(29)-en-3-one, a lupane-type triterpenoid, is known for its anti-inflammatory, antioxidant, anticancer and antimicrobial properties (10). These effects complement the bioactivity of stigmasterol and may play a critical role in the plant's therapeutic potential, particularly in inflammation and oxidative stress-related conditions. γ -Sitosterol, which is structurally similar to stigmasterol, is primarily known for its antihyperglycemic effect (11). This supports the potential role of *P. dalbergioides* in managing metabolic disorders like diabetes, aligning with its use in traditional formulations aimed at regulating blood glucose levels. 1-Triacontanol is a long-chain alcohol derivative reported to possess antitumor properties (12). These compounds are known to exhibit a broad spectrum of pharmacological activities that collectively validate the traditional medicinal use of this species.

Antibacterial activity

The bacterial inhibition of *P. dalbergioides* leaf and bark extracts was tested against *S. aureus* (Gram +ve) and *E. coli* (Gram -ve) using the inhibition zone test, with results detailed in Table 3.

Among the two pathogens, the bark extract exhibited the highest inhibition, with zones measuring 13 mm (30 μ L) against *S. aureus* and 11 mm (30 μ L) against *E. coli*, whereas the leaf extract showed a maximum inhibition of 9 mm (30 μ L) against both microbes, as shown in Fig. 4. These findings indicate that both the extracts possess antibacterial activity against *S. aureus* and *E. coli*, with varying effectiveness depending on the microbial strain as well as the concentration of the extract. Similarly, other species of *Pterocarpus*, including *P. indicus*, *P. erinaceus*, *P. santalinus*, *P. angolensis*, *P. macrocarpus* and *P. soyauxi*, have demonstrated antibacterial activity against various bacteria, such as *S. aureus*, *E. coli*, *Bacillus subtilis*, *Pseudomonas aeruginosa*, *Pantoea agglomerans*, *B. pumilus*, *Proteus mirabilis*, *B. cereus* and *Enterococcus faecalis* (13-18).

The antibacterial properties of these *Pterocarpus* species are attributed to the presence of various secondary metabolites, such as tannins, terpenoids, flavonoids and phenols (19). These findings suggest that the plants in the *Pterocarpus* genus may serve as a promising source of natural antimicrobial agents, potentially offering novel mechanisms of action compared to conventional antibiotics.

Conclusion

The present study confirms that *Pterocarpus dalbergioides* contains a diverse range of bioactive secondary metabolites, including bis(2-ethylhexyl) phthalate, stigmasterol, dotriacontane, γ -sitosterol and 1-triacontanol, as identified through GC-MS analysis. The collective presence of these compounds underscores the multi-targeted therapeutic potential of *P. dalbergioides*, including antimicrobial, anti-inflammatory, antioxidant and anticancer effects. The antimicrobial efficacy of both bark and leaf extracts against specific microbial strains further supports the plant's therapeutic potential. To advance its therapeutic application, future studies should focus on compound isolation, *in vivo* validation of pharmacological activity, toxicity profiling and elucidation of the mechanisms of action of key bioactive compounds. These approaches will be crucial for developing standardized extracts or pure compounds for potential drug development.

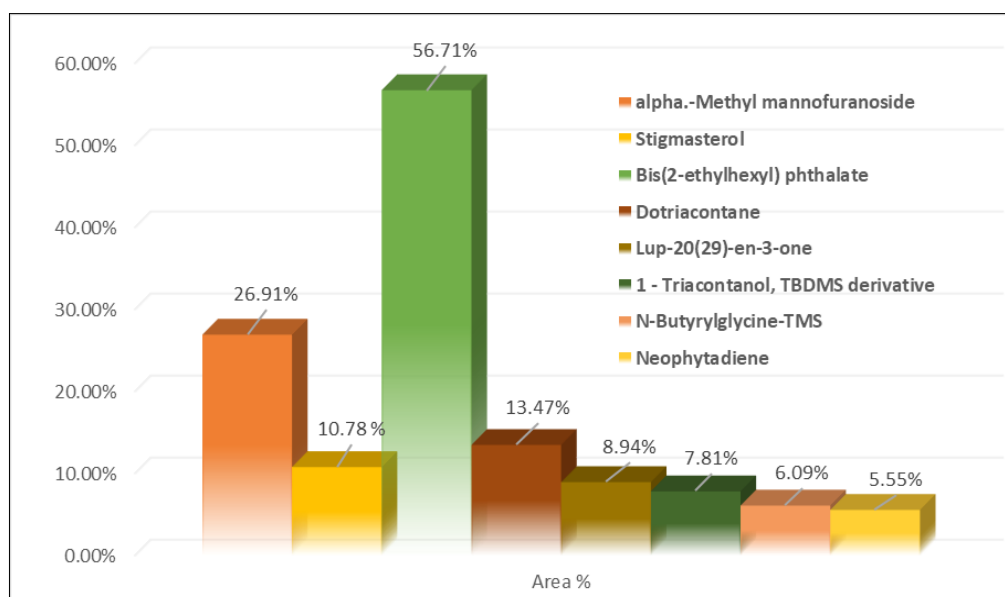
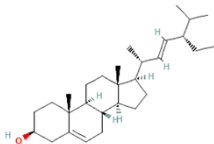
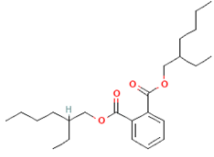

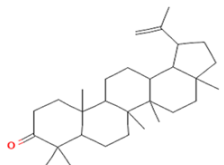
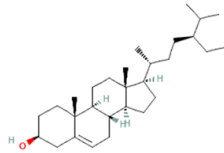
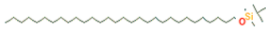
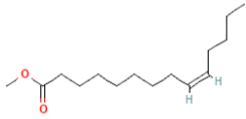
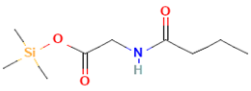
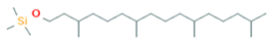
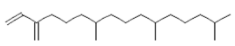
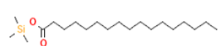
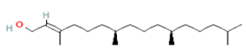


Fig. 3. Predominant compounds of *P. dalbergioides* extract.

Table 2. Biological activity and chemical structure of predominant compounds of *P. dalbergioides* extracts

Compounds	Biological activities	Structure	Reference
Stigmasterol	Anti-osteoarthritis, anti-inflammatory, anticancer, immunomodulatory effects, neuroprotective effects, antimicrobial activity, anti-diabetic and antiparasitic properties		(6)
Bis(2-ethylhexyl) phthalate	Antimicrobial activity, larvicidal activity, leukemia Preventive and mutagen-inhibiting effects		(7, 8)
Dotriacontane	Antimicrobial and antioxidant properties		(9)
Lup-20(29)-en-3-one	Anti-inflammatory, antioxidant, anticancer and antimicrobial effects		(10)
gamma-Sitosterol	Antihyperglycemic		(11)
1-Triacontanol, TBDMS derivative	Antitumor activity		(12)
Methyl myristoleate	Antifungal		(20, 21)
N-Butyrylglycine, TMS	-		-
Dihydrophytol, TMS derivative	-		-
Neophytadiene	Anxiolytic effects and convulsion-preventing properties		(22)
Margaric acid-TMS	-		-
Phytol	Antiparasitic, anti-anxiety, antioxidant, anti-inflammatory, antinociceptive and anticancer effects		(23)

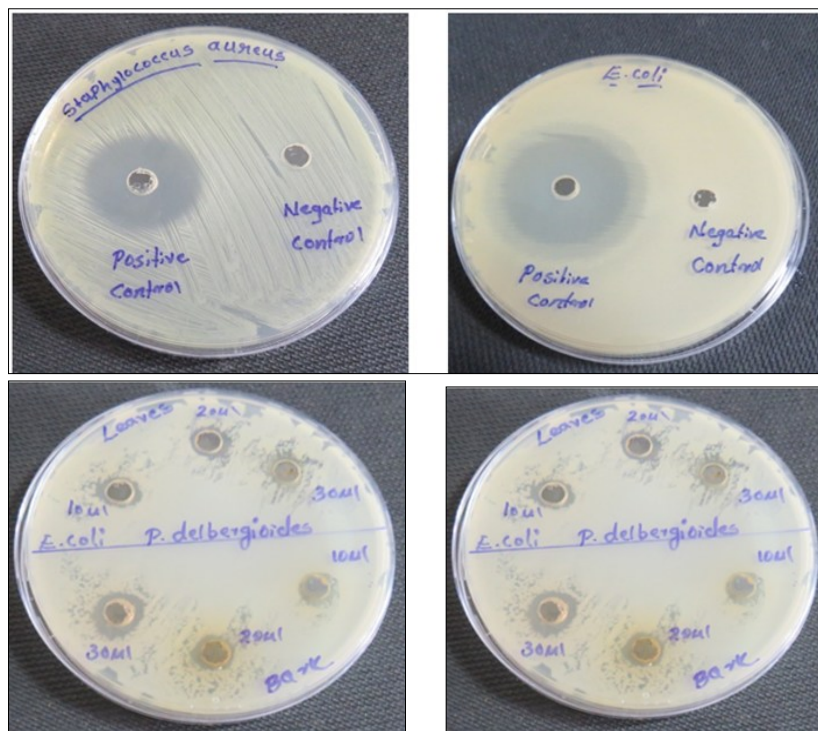


Fig. 4. Antibacterial efficacy of bark and leaf extracts of *P. dalbergioides*.

Table 3. Antibacterial effectiveness of *P. dalbergioides* against two microbial strains

Sample	Inhibition Zone	
	<i>S. aureus</i>	<i>E. coli</i>
Standard - (Gentamicin)	28 mm	31 mm
Negative control- Methanol	0 mm	0 mm
<i>P. dalbergioides</i> - Bark		
10µL	10 mm	8 mm
20µL	8 mm	8 mm
30µL	13 mm	11 mm
<i>P. dalbergioides</i> - Leaves		
10µL	8 mm	9 mm
20µL	8 mm	8 mm
30µL	9 mm	9 mm

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Authors' contributions

HP contributed in conceptualization of the review, providing critical insights, final revision and approval of manuscript for submission, RV helped in literature search, initial drafting of the manuscript, creation of tables and figures, KM writing and summarizing sections on specific subtopics, SVP supported the GC-MS study and tabulation of data, KS contributed in writing and revising sections of the manuscript, JI provided support in collecting plant samples and framing the methodology, ST assisted in forming a framework for reviewing the literature, RR verified data and references to ensure accuracy.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interests to declare.

Ethical issues: None

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