The Journal of Phytopharmacology 2024; 13(6):470-477
Online at: www.phytopharmajournal.com

Research Article

ISSN 2320-480X
JPHYTO 2024; 13(6): 470-477
November- December
Received: 09-11-2024
Accepted: 11-12-2024

doi: 10.31254/phyto.2024.13608

Sheetal Koranga
Department of Botany, Dev Singh Bisht Campus, Kumaun University, Nainital-263001, Uttarakhand, India

Preeti Chaturvedi
Department of Biological Sciences, College of Basic Sciences and Humanities, Govind Ballabh Pant University of Agriculture and Technology, Pantnagar- 263145, Uttarakhand, India

Lalit M. Tewari
Department of Botany, Dev Singh Bisht Campus, Kumaun University, Nainital-263001, Uttarakhand, India

Correspondence:
Dr. Preeti Chaturvedi
Department of Biological Sciences, College of Basic Sciences and Humanities, Govind Ballabh Pant University of Agriculture and Technology, Pantnagar- 263145, Uttarakhand, India
Email: pchaturvedi.biolsc@gmail.com

Secondary metabolite profiling, antioxidant, and anti-inflammatory investigations of *Symplocos paniculata* (Thunb.) Miq., an ethnomedicinal plant from Kumaun Himalayan region

Sheetal Koranga, Preeti Chaturvedi, Lalit M. Tewari

ABSTRACT

Background: Traditional medicine relies heavily on plant-based remedies for immune system modulation, offering advantages of affordability, reduced toxicity, minimal side effects, and better accessibility compared to synthetic alternatives. *Symplocos paniculata* (Thunb.) Miq. has been traditionally used in ethnomedicine for various ailments, though its chemical composition and therapeutic properties remain inadequately studied. Objectives: Thus, this study aimed to investigate the phytochemical composition and evaluate the antioxidant and anti-inflammatory properties of leaf and bark extracts from *S. paniculata*. Material and Methods: The study employed qualitative and quantitative phytochemical screening methods, along with *in vitro* assays including DPPH radical scavenging and metal chelation for antioxidant activity, and protein denaturation for anti-inflammatory effects. GC-MS analysis was conducted to identify the phytoconstituents. Results and Conclusion: Phytochemical screening revealed diverse compounds, with leaf extracts showing higher concentrations of phenolics, flavonoids, and tannins compared to bark extracts. Bark extract demonstrated superior antioxidant properties with DPPH radical scavenging (IC_{50} = 61.37 µg/ml) and metal chelating (IC_{50} = 30.06 µg/ml) activities. Moderate anti-inflammatory activity was observed, with leaf extract showing slightly higher potency (IC_{50} = 222.81 µg/ml). GC-MS analysis identified terpenes, phytosterols, and fatty acids as major constituents in both extracts. The study provides scientific validation for traditional medicinal applications of *S. paniculata* through comprehensive characterization of its phytochemical profile and demonstration of significant antioxidant and anti-inflammatory properties, establishing its potential for pharmaceutical development.

Keywords: *Symplocos paniculata*, Ethnomedicine, Antioxidant, Anti-inflammatory, Terpenes.

INTRODUCTION

Medicinal plants have long been acknowledged as valuable sources of diverse drugs, yielding various therapeutic compounds [1]. Recognizing their importance, the World Health Organization (WHO) emphasized investigating these plants to deepen our understanding of their properties, mechanisms of action, and applications [2]. Traditional medicine relies extensively on natural remedies derived from plants and their formulations to regulate the immune system. These treatments are increasingly attractive due to their cost-effectiveness, low toxicity, reduced health risks, and ready availability compared to synthetic medications [3,4].

One such ethnomedicinal plant of interest is *Symplocos paniculata* (Thunb.) Miq., commonly known as 'Asiatic Sweet Leaf' or 'Sapphire Berry,' marketed as 'Lodhra' (Figure 1). It belongs to the Symplocaceae family, comprising just one genus, *Symplocos*, which encompasses approximately 300 to 500 species [5]. In Ayurveda, *S. paniculata* is recognized as 'Akshi bheshaja' for its traditional use in treating eye conditions. Additionally, it holds significance for its efficacy in addressing uterine complaints, vaginal issues, and menstrual disorders [6]. *S. paniculata* is a deciduous shrub or medium-sized tree that reaches up to 12 meters in height and thrives at 600 to 2,400 meters in the Himalayan region. The tree is recognizable by its rough, yellowish-brown bark. Fruits are 3-8 mm long, globose, or ovoid, turning from blue to black when ripe, typically with one seed. Flowering occurs from May to June, while fruiting occurs from October to December [7].

The bark of *S. paniculata* has a folkloric repute as an astringent, tonic, and coolant that has been traditionally employed to manage conditions such as menorrhagia, bowel complaints, eye ailments, bleeding gums, and ulcers [8,9]. Its traditional use extends to treating diarrhea and lowering blood pressure [10]. Remarkably, the plant material has a historical application in the extraction of red or yellow dye [7]. Additionally, the plant is valuable in modern medicine because of its anti-HIV, anti-tumor, and

phosphodiesterase inhibitory activities [11]. Furthermore, the juice of the bark is applied externally to sprains and muscle swellings [12]. This study focused on evaluating *in vitro* antioxidant and anti-inflammatory activities and utilizing GC-MS analysis to identify essential phytoconstituents in the methanolic leaf and bark extracts of *S. paniculata*. The primary objective of this study was to bridge the existing knowledge gap by providing valuable insights into the characteristics, and potential applications of *S. paniculata*.

MATERIALS AND METHODS

Plant material collection and Extract preparation

Plant materials (leaves and bark) were collected in June 2023 from Niglat (1628 m, 29°23'57" N, 79°30'33" E) near Bhowali, Uttarakhand, and authenticated at the Botanical Survey of India, Northern Regional Centre, Dehradun with voucher specimen number 1628. The leaves and bark were cleaned with running water, rinsed with distilled water, shade-dried, and ground into a fine powder. Extraction was performed using the hot Soxhlet method, where 12.0 g of powdered material was extracted with 250 ml methanol at 50°C for 24 hours. The obtained solvent was evaporated, and the resulting solid extract was stored at 4°C for further analysis.

Phytochemical analysis of the plant extract

Qualitative tests were carried out to detect the presence of alkaloids (Mayer's test), phenols and tannins (ferric chloride test), flavonoids (sulfuric acid test), coumarins (sodium hydroxide test), quinones (hydrochloric acid test), saponins (froth test), and proteins (xanthoproteic test) in the plant extracts following the standard methods [13,14]. For quantitative analysis of the plant extract total phenolics, total flavonoids and total tannin content of the plant extract was measured [15-17].

2,2'-Diphenylpicrylhydrazyl (DPPH) free radical scavenging activity

The free radical scavenging activity was assessed employing the 2,2-diphenyl-1-picryl-hydrazyl (DPPH) assay with subtle modifications as described [18]. The reaction mixture (3.0 ml) included 2.0 ml of DPPH (0.004%) in methanol and 1.0 ml of extract. After 60 minutes of incubation in the dark, the absorbance at 517 nm was measured using methanol as a blank. Ascorbic acid served as the positive control. The percent inhibition was determined using equation: Scavenging (%) = $1-(A_t / A_0) \times 100$

where A_0 is the absorbance of the DPPH solution and A_t is the absorbance of the test sample.

The IC_{50} was also calculated by plotting the scavenging percentage against the sample concentration.

Metal chelating activity

The metal chelating activity was determined using ferrozine, a potent metal chelator [19]. Methanol extracts were prepared at concentrations ranging from 20 to 100 µg/ml. Each sample was adjusted to 5 ml with 0.2 ml of 5 mM ferrozine and 0.2 ml of methanol, followed by incubation at room temperature for 10 minutes. EDTA (Ethylenediaminetetraacetic acid) was used as the standard. The percent inhibition of metal chelation activity was calculated using equation: Chelation (%) = $1-[(A_0-A_t)/A_0] \times 100$

where A_o is the absorbance of the control sample and A_t is the absorbance of the test sample.

To establish the IC_{50} values of the extracts and the standard, the percentage of chelating activity was plotted against the concentration. The lower the IC_{50} value was, the greater the metal chelating activity.

In vitro anti-inflammatory activity

The *in vitro* anti-inflammatory effectiveness in preventing the denaturation of egg albumin protein was determined as described [20]. A 5 ml reaction mixture comprising 0.2 ml of fresh hen egg albumin, 2.8 ml of phosphate-buffered saline (PBS) at pH 6.4, and 2 ml of various concentrations (100-500 µg/ml) of methanolic plant extracts was prepared. The control group was treated with an equivalent volume of distilled water. These mixtures were incubated at 37 ± 2°C in an incubator for 15 minutes, followed by heating for 5 minutes in a water bath at 70°C. After cooling, the absorbance of these mixtures was measured at 660 nm. Diclofenac sodium was used as the standard. The percentage of protein denaturation inhibition was calculated using equation: Inhibition (%) = $1-(A_t/A_c) \times 100$

where A_t is the absorbance of the sample and A_c is the absorbance of the control.

Gas chromatography-mass spectrometry (GC-MS) analysis

Gas chromatography-mass spectrometry (GC-MS) utilizing a GCMS-QP 2010 Ultra instrument with a Rxi-5Sil MS ultra-inert capillary GC column (30 m length, 250 µm diameter, 0.25 µm film thickness) coated with polydimethylsiloxane was used to analyze the plant extracts. Helium was used as the carrier gas at 77.6 kPa with a total flow rate of 16.3 ml/minute, a column flow rate of 1.21 ml/minute, and a purge flow of 3.0 ml/minute. The oven temperature started at 70°C, ramped at 3°C/minute to 300°C, held for 2 minutes, increased at 6°C/minute to 300°C, and held for 15 minutes. The flame thermionic detector identified compounds by comparing GC peak retention times to reference compounds and matching mass spectra fragmentation patterns with NIST-11 and WILEY-8 library data.

Statistical analysis

All the experiments were conducted in triplicate and presented as mean ± standard error. The statistical analysis was performed using IBM SPSS 19.0 software, employing analysis of variance (ANOVA) for a completely randomized design at the 5% significance level.

RESULTS

Phytochemical analysis of the plant extract

The qualitative phytochemical analysis of *S. paniculata* methanolic extracts indicated the presence of various bioactive compounds, such as flavonoids, phenols, quinones, tannins, proteins, and coumarins, in the leaf extract. Conversely, saponins and alkaloids tested negative for their presence (Table 1). Quantitative analysis indicated that methanolic extracts of leaves and bark at a concentration of 200 µg/ml had significantly higher levels of phenolic, flavonoid, and tannin contents in the leaf extract compared to the bark extract (Table 2).

2,2'-Diphenylpicrylhydrazyl free radical scavenging activity

The DPPH radical scavenging assay assesses antioxidant capacity by utilizing free radicals through a color change from dark purple to yellow, indicating the reduction of the radicals to a nonradical state [21]. The methanolic extract of the bark exhibited more potent DPPH radical scavenging activity (IC_{50} = 61.37 µg/ml) than the leaf extract (IC_{50} = 69.07 µg/ml). As a reference, ascorbic acid demonstrated an IC_{50} value of 29.78 µg/ml (Figure 2 (A) and Table 3).

Metal chelating activity

Chelating agents are vital in inhibiting lipid peroxidation by stabilizing transition metals [22]. The methanolic extract derived from the bark showed robust scavenging activity. Specifically, the IC_{50} value of the leaf extract was 51.24 µg/ml. At the same time, the bark extract had an even lower IC_{50} value of 30.06 µg/ml, comparable to

that of the standard EDTA (26.64 µg/ml) as shown in Figure 2 (B) and Table 3.

In vitro anti-inflammatory activity

Inflammation and related health conditions are becoming increasingly significant concerns for most of the population [23]. Many plant extracts inhibit denaturation as a potential strategy for anti-inflammatory effects [24]. In this study, the dose-dependent anti-inflammatory potential of plant extracts was systematically assessed (100-500 µg/ml). The leaf methanolic extract showed slightly more significant anti-inflammatory activity (IC_{50} = 222.81 µg/ml) than the bark extract (IC_{50} = 250.29 µg/ml). However, the standard drug diclofenac sodium had an IC_{50} value of 102.93 µg/ml (Table 3). Furthermore, Figure 3 presents the standard curve for diclofenac sodium at various concentrations.

GC-MS analysis

GC-MS analysis of the methanolic leaf and bark extracts of *S. paniculata* revealed the presence of a total of 18 and 22 bioactive compounds, respectively. At the same time, the phytocompounds in the leaf and bark methanolic fractions, along with their retention times (R.T.) and concentrations (peak area %), are detailed in Table 4 and 5, respectively. The significant compounds obtained from the leaf fractions were squalene (31.27%), neophytadiene (18.33%), guanosine (5.50%), 5-α-stigmasta-7,22-dien-β-ol (4.91%), n-nonadecanol-1 (4.57%), and 2-methylcyclopentyl alcohol (3.67%). Similarly, the significant compounds obtained in the bark fractions were stigmasterol (9.30%), lupeol (4.87%), ϒ-sitosterol (4.02%), and methyl commate B (3.23%).

Table 1: Preliminary phytochemical screening of *S. paniculata* methanolic extracts

Phytochemicals	Plant parts		Color/Result
	Leaf	Bark	
Alkaloids	-	+	Creamy precipitate
Flavonoids	+ +	+	Orange
Phenols	+	+	Bluish-green
Quinones	+ +	-	Yellowish-brown
Saponins	-	+ +	Foam
Tannins	++	+	Green
Proteins	+ +	+	Dark yellow
Coumarins	+ +	+	Yellow

(+): present; (+ +): highly present; (-): absent

Table 2: Quantitative phytochemical analysis of methanolic extracts of leaves and bark

Plant parts	TPC (µg GAE/mg)	TFC (µg QE/mg)	TTC (µg TAE/mg)
Leaf	52.19±0.44	22.08±0.13	7.45±0.83
Bark	30.97±0.18	10.81±0.14	1.64±0.13
C.D. at 5%	1.37	0.56	0.88

GAE: Gallic acid equivalents, QE: Quercetin equivalent, TAE: Tannic acid equivalent

Table 3: IC_{50} values (µg/ml) of S. paniculata for in vitro biological activities

Plant extracts	DPPH activity	Metal chelating activity	Anti-inflammatory activity
Leaf	69.07	51.24	222.81
Bark	61.37	30.06	250.29
Standard	Ascorbic acid: 29.78	EDTA: 26.64	Diclofenac sodium: 102.93

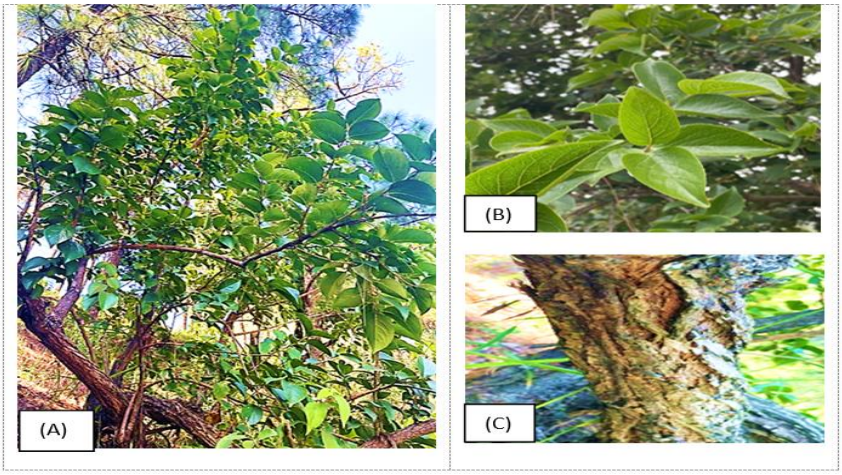

Figure 1: *S. paniculata*: (A)Tree (B) Leaves (C) Bark

Table 4: Phytochemical compounds obtained from GC-MS analysis of leaf extracts of *S. paniculata*

Peak no.	R.T. (Minutes)	Compound name	Formula	Class of compound	Area (%)
1.	7.790	L-mannose	$C_6H_{12}O_6$	Monosaccharide	2.07
2.	10.567	Geranic acid	$C_{10}H_{16}O_2$	Monoterpene	1.72
3.	11.860	Guanosine	$C_{10}H_{13}N_5O_5$	Nucleoside	5.50
4.	11.864	2-methylcyclopentyl alcohol	$C_6H_{12}O$	Cycloalkyl alcohol	3.67
5.	12.554	Stevioside	$C_{38}H_{60}O_{18}$	Glycoside	0.89
6.	12.562	D-Allose	$C_6H_{12}O_6$	Monosaccharide	1.27
7.	13.453	α-methyl-l-sorboside	$C_7H_{14}O_6$	Glycoside	1.54
8.	16.303	Neophytadiene	$C_{20}H_{38}$	Sesquiterpene	18.33
9.	17.597	Palmitic acid	$C_{16}H_{32}O_2$	Fatty acid	1.23
10.	18.780	n-nonadecanol-1	$C_{19}H_{40}O$	Primary alcohol	4.57
11.	19.015	Phytol	$C_{20}H_{40}O$	Diterpene alcohol	2.43
12.	24.666	Squalene	$C_{30}H_{50}$	Triterpene	31.27
13.	25.293	1-heptacosanol	$C_{27}H_{56}O$	Aliphatic alcohol	1.00
14.	25.423	Solanesol	$C_{45}H_{74}O$	Polyisoprenoid alcohol	0.50
15.	25.504	Glycerin tricaprylate	$C_{27}H_{50}O_6$	Triglyceride	1.48
16.	27.268	Vitamin E	$C_{29}H_{50}O_2$	Fat-soluble vitamin	0.86
17.	29.550	5-α-stigmasta-7,22-dien-β-ol	$C_{29}H_{48}O$	Phytosterol	4.91
18.	30.298	α-amyrin	$C_{30}H_{50}O$	Triterpene	0.98

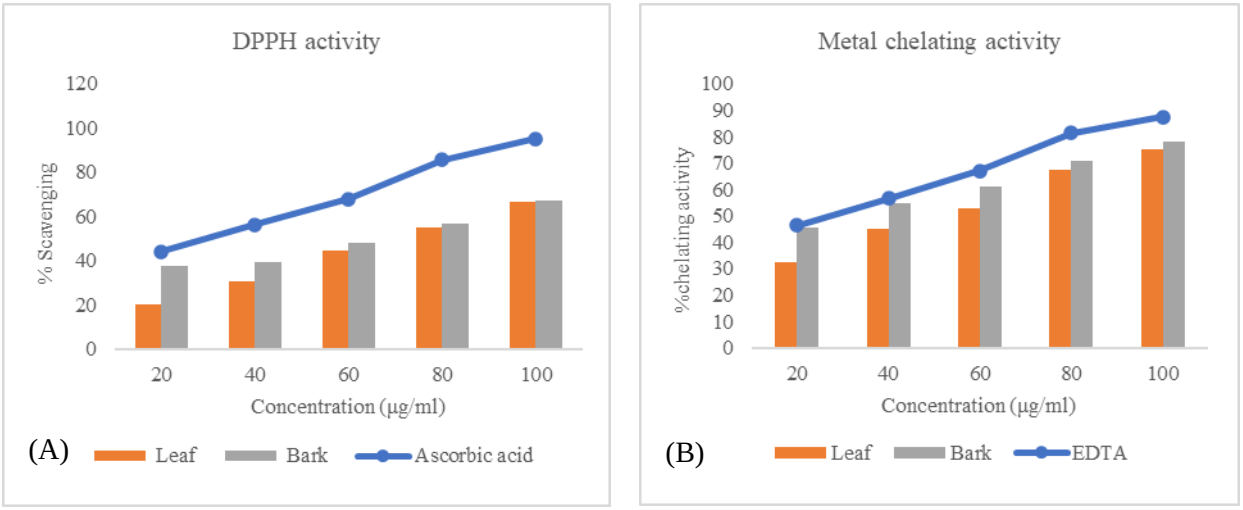

Figure 2: (A) and (B) Percent scavenging of leaf and bark extracts with the standard curve

Table 5: Phytochemical compounds obtained from GC-MS analysis of the bark extract of *S. paniculata*

Peak no.	R.T. (Minutes)	Compound name	Formula	Class of compound	Area (%)
1.	8.060	Guanosine	$C_{10}H_{13}N_5O_5$	Nucleoside	1.05
2.	13.259	Dodecanoic acid	$C_{12}H_{24}O_2$	Saturated fatty acid	0.88
3.	16.302	Neophytadiene	$C_{20}H_{38}$	Sesquiterpene	1.05
4.	16.698	8-octadecanone	$C_{18}H_{36}O$	Fatty acid	1.26
5.	16.785	Palmityl alcohol	$C_{16}H_{34}O$	Fatty alcohol	1.05
6.	17.207	Arachidic acid methyl ester	$C_{21}H_{42}O_2$	Fatty acid ester	0.69
7.	17.599	n-hexadecanoic acid	$C_{16}H_{32}O_2$	Saturated fatty acid	2.17
8.	17.889	1-nonadecene	$C_{19}H_{38}$	Unsaturated hydrocarbon	2.68
9.	18.778	n-nonadecanol-1	$C_{19}H_{40}O$	Saturated alcohol	2.86
10.	21.131	9-hexadecenoic acid, eicosyl ester	$C_{36}H_{70}O_2$	Fatty acid ester	1.48
11.	23.830	Pentatriacontane	$C_{35}H_{72}$	Saturated hydrocarbon	1.15
12.	24.669	Squalene	$C_{30}H_{50}$	Triterpene	0.90
13.	25.245	Tetracontane	$C_{40}H_{82}$	Saturated hydrocarbon	2.85
14.	25.503	Glycerol tricaprylate	$C_{27}H_{50}O_6$	Triglyceride	2.28
15.	26.535	ϒ-tocopherol	$C_{28}H_{48}O_2$	Fat-soluble vitamin	0.45
16.	28.805	Stigmasterol	$C_{29}H_{48}O$	Phytosterol	9.30
17.	29.585	ϒ-sitosterol	$C_{29}H_{50}O$	Phytosterol	4.02
18.	30.032	Ergost-1-en-3-one,12-hydroxy-, (5-β-12-α)	$C_{28}H_{46}O_2$	Sterol	2.70
19.	30.283	Methyl commate B	$C_{31}H_{50}O_3$	Triterpene	3.23
20.	30.894	Lupeol	$C_{30}H_{50}O$	Triterpene	4.87
21.	31.920	Lup-20(29)-en-3-one	$C_{30}H_{48}O$	Triterpene	1.64
22.	37.350	Ursolic aldehyde	$C_{30}H_{48}O_2$	Triterpene	0.85

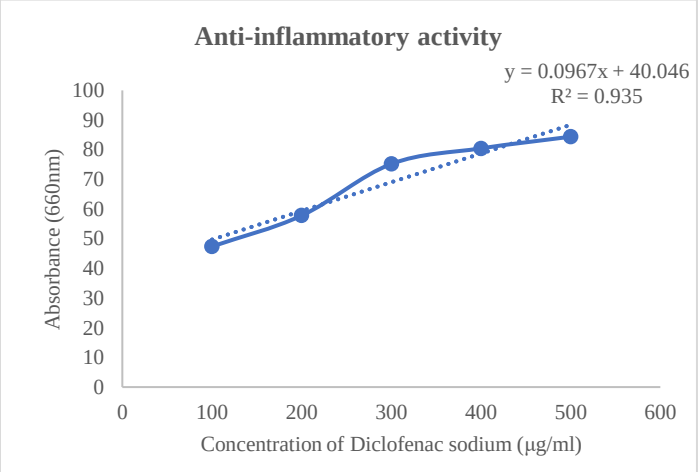

Figure 3: Standard curve graph of diclofenac sodium

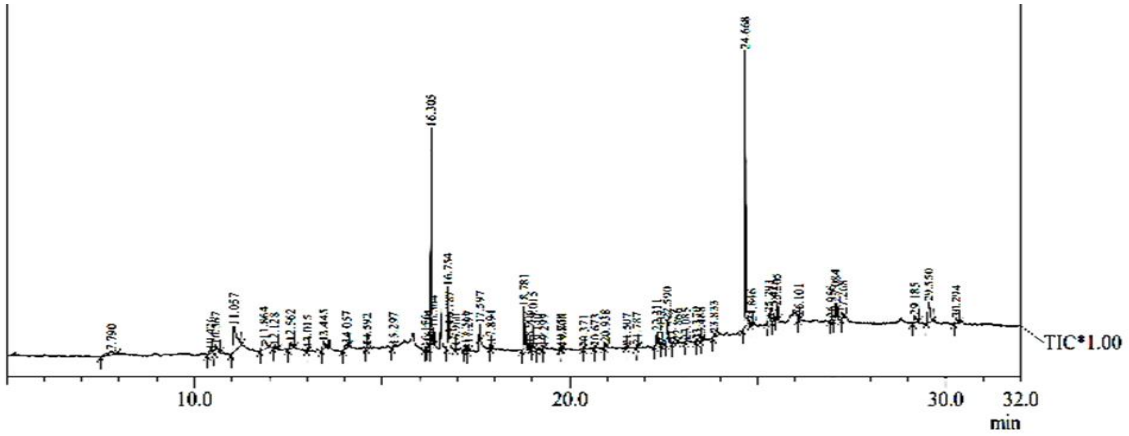

Figure 4: Gas chromatogram of leaf extract of *S. paniculata*

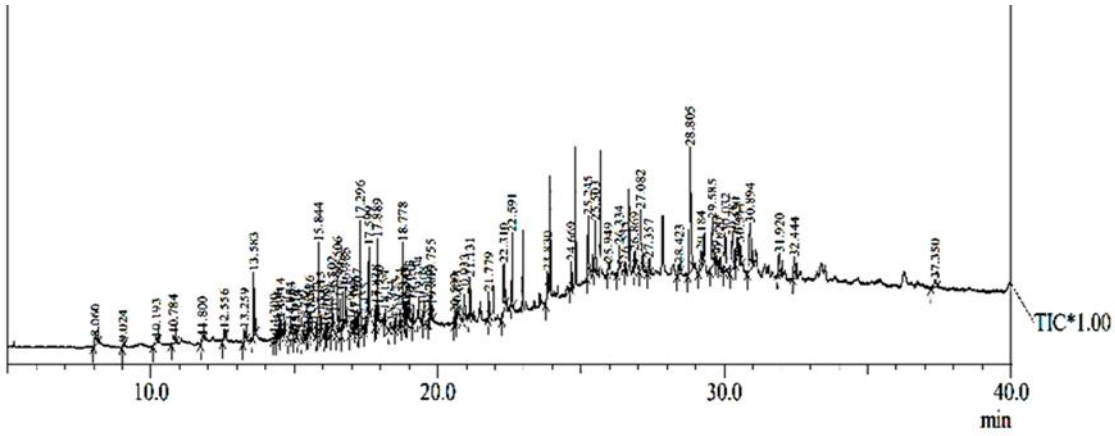

Figure 5: Gas chromatogram of bark extract of *S. paniculata*

DISCUSSION

Bioprospecting involves the rigorous pharmacognostic investigation of crude plant materials, which includes screening for phytochemicals and biological activities [25]. In this context, the study examined the phytochemical, antioxidant, and anti-inflammatory activities of *S. paniculata*, aiming to validate its pharmaceutical potential.

Previous studies have revealed the presence of various secondary metabolites in *S. paniculata* extracts, corroborating phytochemical investigations on plants from the *Symplocos* genus that reported the occurrence of terpenoids, flavonoids, lignans, phenols, steroids, alkaloids, and iridoids [26]. In this study, qualitative phytochemical screening revealed flavonoids, phenols, quinones, tannins, proteins, and coumarins in the leaf extract. At the same time, alkaloids and saponins were also detected in the bark extract, except for quinones (Table 1). This variation in phytochemical distribution across different plant parts is a commonly observed phenomenon [27].

Qualitative phytochemical analysis alone is inadequate for validating the pharmaceutical potential of extracts. Therefore, quantitative investigations targeting essential compounds such as phenols, flavonoids, and tannins are crucial. The quantitative analysis demonstrated more significant phenolic (52.19±0.44 µg GAE/mg), flavonoid (22.08±0.13 µg QE/mg) and tannin (7.45±0.83 µg TAE/mg) contents in the leaf extracts than in the bark (Table 2), which could be attributed to more active biosynthetic pathways or specific metabolic processes in the leaves[28].

The presence of phenols and flavonoids has been linked to potent antioxidant activities [28,29]. Tannin-containing drugs are used in medicine because of their astringent properties and have shown potential therapeutic applications, including antiviral, antibacterial, and antiparasitic effects [30]. Due to the diverse nature of free radicals, assessing antioxidant potential through a single assay is insufficient. Hence, this study utilized DPPH and metal chelation assays to thoroughly evaluate the antioxidant capacity of the extract through different mechanisms. These two assays offer insight into the various antioxidant mechanisms demonstrated by the extracts. The methanolic extracts from the leaves and bark exhibited potent *in vitro* antioxidant activity, as evidenced by their ability to scavenge DPPH radicals (IC_{50} = 69.07 µg/ml for leaves and 61.37 µg/ml for bark) and chelate metals (IC_{50} = 51.24 µg/ml for leaves and 30.06 µg/ml for bark) (Table 3). Notably, the bark extract showed comparatively superior potency, particularly in metal chelation. This superior antioxidant effect of the bark could be attributed to the presence of phytosterols such as stigmasterol (9.30%) and γ-sitosterol (4.02%), triterpenes such as squalene (0.90%), methyl commate B (3.23%), and lupeol (4.87%) and fatty acids such as n-hexadecanoic acid (2.17%) (Table 5).

Moreover, oxidative stress and free radicals are implicated in the pathogenesis of inflammatory diseases [31]. The assessment of anti-inflammatory effects in addition to antioxidant activity offers insights into the potential of the extracts to modulate oxidative stress-mediated inflammatory pathways. As observed in protein denaturation inhibition assays, leaf (IC_{50} = 222.81 µg/ml) and bark (IC_{50} = 250.29 µg/ml) extracts showed moderate effects compared to the standard diclofenac sodium (IC_{50} = 102.93 µg/ml) (Table 3). The leaf extract had a slightly more significant anti-inflammatory effect, likely attributed to the presence of compounds such as squalene (31.27%), neophytadiene (18.33%), and phytosterols such as 5-α-stigmasta-7,22-dien-β-ol (4.91%). Additionally, Semwal *et al*. [32] reported the presence of various triterpenoids in *S. paniculata* that exhibited potent anti-inflammatory activities, complementing the findings of the current investigation.

GC-MS analysis provided insights into the phytochemical composition of the leaf and bark extracts of *S. paniculata*. In the leaf extract, eighteen compounds were identified, representing 83.22% of the total composition. Terpenes, including squalene (31.27%),

neophytadiene (18.33%), and geranic acid (1.72%), collectively constituted 51.32% of the total terpenes. Additionally, guanosine (5.50%) and 5-α-stigmasta-7,22-dien-β-ol (4.91%) were identified as the other predominant compounds (Figure 4 and Table 4). Squalene, a triterpene possesses anticancer, antioxidant, detoxifying, skin-hydrating, and emollient activities [33]. Neophytadiene is known for its analgesic, antipyretic, anti-inflammatory, and antioxidant properties [34], and the guanosine in methanol leaf extracts is known for its neuroprotective properties [35].

Similarly, in the bark extract, a total of twenty-two compounds were identified from the GC-MS analysis of the methanol fraction, revealing the presence of a significant number of bioactive constituents, including terpenoids (12.54%), phytosterols (13.32%), and fatty acids (4.31%) (Figure 5 and Table 5). Notably, phytosterols such as stigmasterol and γ-sitosterol are well-known to have antioxidant potential [36]. Hexadecanoic acid, recognized for its robust antimicrobial and anti-inflammatory activities, is also in the bark extract [37]. Govindarajan *et al*. [6] reported the presence of fifty-seven compounds in the n-hexane bark extract of *S. paniculata*, with undecane, isopropyl myristate, dodecane, 1,2,4-trimethyl-benzene, octacosane, 2-methyl-decane, 2-ethyl-1,2-dimethyl-benzene, and 1,2,3,5-tetramethyl-benzene as the significant compounds. Nevertheless, it is worth mentioning herein that GC-MS studies are insufficient for conclusive phytochemical investigations because they cannot determine the structure of bioactive compounds. NMR spectroscopy is necessary for providing insight into the structures and functionalities of various components. This technique is crucial in drug development for molecular identification and structural elucidation [38, 39].

CONCLUSION

This study revealed that *S. paniculata* possesses various secondary metabolites, including alkaloids, coumarins, proteins, quinones, saponins, and tannins, as well as high phenolic and flavonoid contents. Thus, *S. paniculata* exhibits striking scavenging activities in its leaf and bark extracts. GC-MS analysis identified essential bioactive compounds, such as squalene, neophytadiene, phytosterols, and fatty acids, which are known to contribute to potent *in vitro* biological activities. These findings underscore the potential pharmaceutical applications of *S. paniculata*, supporting its traditional use in medicine. However, further *in vivo* studies are imperative to validate and explore the therapeutic efficacy of these extracts and their active constituents.

Acknowledgments

The authors acknowledge G.B. Pant University of Agriculture and Technology, Pantnagar for providing all the necessary research facilities. Appreciation is extended to the Advance Instrumentation Research Facility (AIRF), JNU, New Delhi, for facilitating the GC-MS analysis.

Conflict of interest

The authors declared no conflict of interest.

Financial Support

None declared.

ORCID ID

Preeti Chaturvedi: https://orcid.org/0000-0002-2408-3299

Sheetal Koranga: https://orcid.org/0000-0002-6951-8655

Lalit M. Tewari: https://orcid.org/0000-0001-6398-333X

REFERENCES

1. Sarvanandaa L, Premarathna AD, Karunarathnad SC. Immunomodulatory effect of Cardiospermum halicacabum against cancer. Biomed J. 2018;1(4):97-109.
2. Alamgir M, Uddin SJ. Recent advances on the ethnomedicinal plants as immunomodulatory agents. Ethnomedicine: A Source of Complementary Therapeutics. 2010;37(661):2.
3. Ganju L, Karan D, Chanda S, Srivastava KK, Sawhney RC, Selvamurthy W. Immunomodulatory effects of agents of plant origin. Biomed Pharmacother. 2003;57(7):296-300.
4. Samant SS, Jagtap VA, Kalangutkar P, Morye R, Gadekar A, Rane R, et al. Phytochemistry and therapeutic uses of Albizia lebbeck. Int J Herbal Med. 2023;11(5):22-6.
5. Manimaran P, Solai Senthil Kumar K, Prithiviraj M. Investigation of physico-chemical, mechanical, and thermal properties of the Albizia lebbeck bark fibers. J Nat Fibers. 2021;18(8):1151-62.
6. Sharma T, Rani C, Kumar D, Kumar H, Deep A. Phytochemical screening and traditional medicinal potential of Albizia lebbeck (L.) Benth: An update. TMR Mod Herb Med. 2022;5(4):23.
7. Shirisha K, Priyanka B, Rahman H, Bardalai D, Ali F. Review on Albizia lebbeck (L.) Benth: A plant possessing diverse pharmacological activities. Res J Pharmacogn Phytochem. 2013;5(5):263-8.
8. Avoseh ON, Mtunzi FM, Ogunwande IA, Ascrizzi R, Guido F. Albizia lebbeck and Albizia zygia volatile oils exhibit anti-nociceptive and anti-inflammatory properties in pain models. J Ethnopharmacol. 2021;268:113676.
9. Al-Snafi AE. A review on Lawsonia inermis: A potential medicinal plant. Int J Curr Pharm Res. 2019;11(5):1-3.
10. Mikhaeil BR, Badria FA, Maatooq GT, Amer MM. Antioxidant and immunomodulatory constituents of henna leaves. Z Naturforsch C. 2004;59(7-8):468-76.
11. Dikshit V, Dikshit J, Saraf M, Thakur V, Sainis K. Immunomodulatory activity of naphthoquinone fraction of Lawsonia inermis Linn. Phytomedicine. 2000;7(2):102-3.
12. Stéphane FF, Jules BK, Batiha GE, Ali I, Bruno LN. Extraction of bioactive compounds from medicinal plants and herbs. Nat Med Plants. 2021:1-39.
13. Zieliński H, Ceglińska A, Michalska A. Antioxidant contents and properties as quality indices of rye cultivars. Food Chem. 2007;104(3):980-8.
14. Demiray S, Pintado ME, Castro PM. Evaluation of phenolic profiles and antioxidant activities of Turkish medicinal plants: Tilia argentea, Crataegi folium leaves, and Polygonum bistorta roots. Int J Pharmacol Pharm Sci. 2009;3(6):74-9.
15. Ambwani S, Dolma R, Sharma R, Kaur A, Singh H, Ruj A, et al. Modulation of inflammatory and oxidative stress biomarkers due to dexamethasone exposure in chicken splenocytes. Vet Immunol Immunopathol. 2023;262:110632.
16. Creed TJ, Lee RW, Newcomb PV, di Mambro AJ, Raju M, Dayan CM. The effects of cytokines on suppression of lymphocyte proliferation by dexamethasone. J Immunol. 2009;183(1):164-71.
17. Ambwani S, Kandpal D, Arora S, Ambwani TK. Cytotoxic effects of gold nanoparticles exposure employing in vitro animal cell culture system as part of nanobiosafety. AIP Conf Proc. 2016;1724(1).
18. Mishra SS, Gothecha VK, Sharma A. Albizia lebbeck: A short review. J Herb Med Toxicol. 2010;4(2):9-15.
19. Desai TH, Joshi SV. Anticancer activity of saponin isolated from Albizia lebbeck using various in vitro models. J Ethnopharmacol. 2019;231:494-502.
20. Saleem U, Raza Z, Anwar F, Chaudary Z, Ahmad B. Systems pharmacology-based approach to investigate the in vivo therapeutic efficacy of Albizia lebbeck (L.) in an experimental model of Parkinson's disease. BMC Complement Altern Med. 2019;19:1-6.

21. Balkrishna A, Sakshi, Chauhan M, Dabas A, Arya V. A comprehensive insight into the phytochemical, pharmacological potential, and traditional medicinal uses of Albizia lebbeck (L.) Benth. Evid Based Complement Altern Med. 2022;2022:5359669.
22. Kamala Lakshmi B, Valarmathi S. In vitro anti-inflammatory activity of aqueous extract of Albizia lebbeck leaf (L.). J Phytopharmacol. 2020;9:356-60.
23. Sangeetha G, Vidhya R. In vitro anti-inflammatory activity of different parts of Pedalium murex (L.). Inflammation. 2016;4(3):31-6.
24. Meshram GG, Kumar A, Rizvi W, Tripathi CD, Khan RA. Evaluation of the anti-inflammatory activity of the aqueous and ethanolic extracts of the leaves of Albizia lebbeck in rats. J Tradit Complement Med. 2016;6(2):172-5.
25. Kuhl K, Hanania NA. Targeting IgE in asthma. Curr Opin Pulm Med. 2012;18(1):1-5.
26. Raja W, Ovais M, Dubey A. Phytochemical screening and antibacterial activity of Lawsonia inermis leaf extract. Med. 2013;6(8).
27. Muhammad HS, Muhammad S. The use of Lawsonia inermis Linn. (henna) in the management of burn wound infections. Afr J Biotechnol. 2005;4(9).
28. Dasgupta T, Rao AR, Yadava PK. Modulatory effect of henna leaf (Lawsonia inermis) on drug-metabolizing phase I and phase II enzymes, antioxidant enzymes, lipid peroxidation, and chemically induced skin and forestomach papillomagenesis in mice. Mol Cell Biochem. 2003;245:11-22.
29. Khatun R, Khanam F, Juhi SA, Khatune NA, Rahman MA. In vitro evaluation of anti-diabetic and anti-inflammatory activities of ethanolic extract of Lawsonia inermis L. leaves (Family: Lythraceae). J Pharmacogn Phytochem. 2022;11(1):252-5.
30. Aremu A, Idris JF, Akorede GJ, Olatumji AO, Basiru A, Ahmed AO. Lawsonia inermis possesses significant analgesic activity compared to Waltheria indica, Moringa oleifera, Nigella sativa, and diclofenac in female Wistar rats. Iran J Vet Sci Technol. 2023;15(2):48-55.
31. Pisoschi AM, Pop A, Cimpeanu C, Predoi G. Antioxidant capacity determination in plants and plant-derived products: A review. Oxid Med Cell Longev. 2016;2016:9130976.
32. Pandey Y, Ambwani S. Nano metal-based herbal theranostics for cancer management: Coalescing nature's boon with nanotechnological advancement. Curr Pharm Biotechnol. 2022;23(1):30-46.
33. Mathesius U. Flavonoid functions in plants and their interactions with other organisms. Plants. 2018;7(2):30.
34. Ambwani S, Tandon R, Ambwani TK, Malik YS. Current knowledge on nanodelivery systems and their beneficial applications in enhancing the efficacy of herbal drugs. J Exp Biol Agric Sci. 2018;6(1):87-107.

HOW TO CITE THIS ARTICLE

Koranga S, Chaturvedi P, Tewari LM. Secondary metabolite profiling, antioxidant, and anti-inflammatory investigations of *Symplocos paniculata* (Thunb.) Miq., an ethnomedicinal plant from Kumaun Himalayan region. J Phytopharmacol 2024; 13(6):470-477. doi: 10.31254/phyto.2024.13608