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Original Article

PHYTOCHEMICAL PROFILE AND PHYSICOCHEMICAL CHARACTERIZATION OF THE LEAVES OF *BAUHINIA RACEMOSA*

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ABSTRACT

The current study focuses on the comprehensive evaluation of the phytochemical and physicochemical properties of *Bauhinia racemosa* (*B. racemosa*) leaves, a plant widely used in traditional medicine across Asia. Fresh leaves were collected, authenticated, air-dried, and powdered before undergoing Soxhlet extraction with solvents such as acetone, water, chloroform, dichloromethane, ethanol, ethyl acetate, and petroleum ether. Physicochemical parameters — including ash values, extractive values, moisture content, and potential of hydrogen (pH) — were measured to ensure quality and stability. Qualitative phytochemical screening revealed the presence of bioactive compounds, including carbohydrates, proteins, glycosides, steroids, flavonoids, tannins, and phenolic compounds, known for their antioxidant, anti-inflammatory, and antimicrobial properties. Notably, the identification of flavonoids and phenolic compounds underscores the plant's potential for managing oxidative stress and related health conditions. We establish a foundational profile of *B. racemosa* leaves, contributing to their standardization and supporting their medicinal value for potential applications in herbal medicine and pharmaceutical development.

KEYWORDS: *Bauhinia racemosa*, phytochemical screening, bioactive compounds, physicochemical analysis, ash value, extractive value.

1. Introduction

Plants have been central to human healthcare for centuries, serving as the foundation of traditional medicine and a valuable resource for modern drug development [1,2]. The World Health Organization (WHO) estimates that nearly 80% of the global population relies on plant-based medicines for primary healthcare, highlighting their enduring relevance [3]. Advances in phytochemical research have identified numerous bioactive compounds from plants that are integral to nutraceuticals, cosmetics, and pharmaceuticals [4,5]. These natural compounds are often favored for their minimal toxicity, compatibility with human biochemistry, and fewer side effects compared to synthetic drugs [6-8].

Among medicinal plants, *Bauhinia racemosa* (*B. racemosa*), commonly known as the "Bidi leaf tree," stands out for its rich phytochemical composition and extensive use in traditional medicine. This small deciduous tree is native to India, Sri Lanka, China, and other parts of Asia. Its leaves are utilized in South India for making beedis (traditional Indian cigarettes), while its bark and leaves are employed in treating headaches, fever, skin diseases, and dysentery [9,10]. The plant is also renowned for its anti-inflammatory, antimicrobial, and antioxidant properties, making it a valuable component of Ayurvedic and traditional medicine systems [9,11].

Phytochemical screening is essential for understanding the therapeutic potential of medicinal plants. Compounds such as flavonoids, tannins, alkaloids, and phenolic compounds are often the key contributors to the biological activities of plants [6,12]. Profiling these compounds aids in identifying therapeutic applications, ensuring the quality and consistency of herbal formulations, and potentially discovering new drug candidates [13,14]. This is particularly important in regions with limited access to modern medicine, where medicinal plants serve as primary healthcare resources [15,16].

Despite the documented medicinal properties of *B. racemosa*, comprehensive profiling of its phytochemical composition across different solvents is limited. Addressing this gap is critical for standardizing herbal formulations and ensuring consistent therapeutic efficacy. The current study systematically analyzes the bioactive compounds in the leaves of *B. racemosa*, offering insights to support the development of novel

therapeutic applications and enhance the utility of traditional remedies, particularly in regions where such plants are a primary healthcare resource.

2. Materials and Methods

2.1. Plant Material

Fresh leaves of *B. racemosa* were collected during August and September from the Dharashiv (Osmanabad) district, Maharashtra, India. The plant material was authenticated by Dr. A. S. Linge, Head of the Department of Botany at Venkatesh Mahajan Senior College, Dharashiv, which is affiliated with Dr. Babasaheb Ambedkar Marathwada University, Chhatrapati Sambhaji Nagar, Maharashtra. Voucher specimens were deposited with herbarium no. 128(B) 2022-23.

2.2. Preparation of Powder and Extracts

The collected leaves were washed thoroughly with reverse osmosis-purified water to remove surface impurities. After washing, the leaves were dried in a house loft for 21 days to prevent photodegradation. Once dried, they were ground into a powder using an electric grinder (Philips HL7756/01 750 Watt Mixer Grinder).

Soxhlet extraction was conducted using 100 grams of the powdered leaf material. The material was placed in a thimble and loaded into a Soxhlet extractor purchased from Dolphin Pharmacy Instruments Pvt Ltd, Mumbai, Maharashtra, India. A 500 mL round-bottom flask containing 300 mL of laboratory-grade solvent (acetone, water, chloroform, dichloromethane, ethanol, ethyl acetate, or petroleum ether) sourced from Research Lab Fine Chem Industries, Mumbai, Maharashtra, India, was attached to the Soxhlet apparatus, along with a condenser.

The setup was positioned on a heating mantle, and the solvent was heated to its boiling point. The evaporated solvent passed through the Soxhlet extractor to the condenser, where it was cooled and condensed back into liquid form. The condensed solvent dripped into the chamber containing the plant sample in the thimble. When the solvent level reached the siphon, it was siphoned back into the round-bottom flask, recycling the solvent-extract mixture. This process was repeated continuously for 24 hours for each solvent [17].

After extraction, the solvent-extract mixture was cooled to ambient temperature. The solvent was evaporated under reduced pressure using a Superfit Rotary Vacuum Evaporator (PBU 6D) supplied by Superfit Continental Private Limited, Mumbai, Maharashtra, India. The concentrated extracts were stored in airtight containers at 4°C for further analysis. The extraction process was repeated three times for each solvent to ensure reliability and reproducibility [17].

2.3. Physicochemical Analysis

The powdered leaf material underwent various physicochemical analyses, including determination of ash content, extractive values in different solvents, moisture content, and the potential of hydrogen (pH) measurement. These analyses followed the methodologies described by Khandelwal and Kokate [18,19].

2.4. Qualitative Phytochemical Screening

The different extracts (acetone, water, chloroform, dichloromethane, ethanol, ethyl acetate, and petroleum ether) were screened for the presence of secondary metabolites, such as carbohydrates, proteins, fixed oils, glycosides, flavonoids, tannins, and phenolic compounds. Screening was performed using methodologies outlined by Khandelwal and Kokate [18,19].

3. Results

3.1. Physicochemical Properties

Key physicochemical parameters, including ash content, extractive values, and moisture content of the *B. racemosa* leaves, are summarized in Table 1.

Table 1. Physicochemical Properties of *B. racemosa* Leaf Powder

Parameter	Result	Unit
Ash value		
Total Ash	5.5	% (w/w)
Acid-Insoluble Ash	3.5	% (w/w)
Water-Soluble Ash	2.5	% (w/w)
Sulfate Ash	3	% (w/w)
Extractive values		
Water	0.64	% (w/w)
Ethanol	0.36	% (w/w)
Acetone	0.40	% (w/w)
Ethyl acetate	0.32	% (w/w)
Dichloromethane	0.44	% (w/w)
Chloroform	0.28	% (w/w)
Petroleum ether	0.36	% (w/w)
Moisture content		
Loss on drying at 110°C	6.65	%
pH		
pH of 1.00% w/v aqueous solution	6	–
pH of 10.00% w/v aqueous solution	7	–

3.1. Qualitative Phytochemical Screening

Phytochemical screening of various solvent extracts of *B. racemosa* leaves identified a range of bioactive compounds, as shown in Table 2.

Table 2. Phytochemical Screening of *B. racemosa* Leaf Extracts in Various Solvents

Phytochemicals	Test/ Reagent	WE	AE	CE	DE	EE	EAE	PEE
Carbohydrates	Molisch's	+	-	+	+	+	+	-
Gums and mucilage	Alcoholic precipitation	+	-	-	-	-	-	-
Proteins	Millon's	+	+	-	+	+	-	-
Amino acids	Ninhydrin	+	+	-	+	+	-	-
//Fixed oils/fats	Spon	-	-	-	-	-	-	-
Volatile oils	Stain	-	-	-	-	-	-	-
Steroids	Salkowski reaction	+	+	-	+	+	+	-
Glycosides	Legal's	+	+	+	+	+	+	-
Saponins	Foam	-	-	-	-	-	-	-
Phytosterols	Liebermann's	-	+	-	+	+	+	+
Flavonoids	Sulphuric acid and lead acetate	-	-	-	+	+	-	+
Phenolic compounds and tannins	5% Ferric chloride and lead acetate	+	+	-	+	+	-	-
Alkaloids	Dragendorff's	-	-	+	+	-	+	-
Enzymes	Catalase (Hydrogen peroxide)	+	-	-	-	+	-	-
Acidic compounds	Sodium bicarbonate	-	-	-	-	-	-	-
Organic acids	Calcium chloride	-	-	-	-	+	-	-
Vitamins	Water + sodium bicarbonate + ferrous sulphate + sulphuric acid	+	-	-	-	+	-	-

AE, acetone extract; CE, chloroform extract; DE, dichloromethane extract; EAE, ethyl acetate extract; EE, ethanol extract; PEE, petroleum extract; WE, water extract.

Note: '+' denotes presence, and '-' denotes absence. All solvents used for extraction were of analytical grade, with a purity exceeding 95%.

4. Discussion

The comprehensive analysis presented in this study highlights the therapeutic potential of *B. racemosa* leaf extracts. The detection of flavonoids, tannins, phenolic compound, and glycosides aligns with prior research and supports the plant's medicinal applications [20-22]. Among the bioactive compounds identified, flavonoids and phenolic compounds stand out due to their potent antioxidant properties, which are crucial for mitigating oxidative stress associated with aging and chronic diseases [23].

The physicochemical properties of the leaf powder, including total ash (5.5%), acid-insoluble ash (3.5%), water-soluble ash (2.5%), and moisture content (6.65%) are within the acceptable limits for medicinal plant materials, indicating good quality and stability [19,24]. These findings are consistent with standards outlined by the WHO for the quality control of herbal materials [25]. The moisture content was 6.65%, which is relatively low and suggests good stability and shelf life for the dried leaves [13,26]. Additionally, the pH values (6 for a 1% solution and 7 for a 10% solution) demonstrate the plant's compatibility with therapeutic formulations, enhancing its safety and efficacy profile [18].

The identified phytochemicals, particularly glycosides, flavonoids, and steroids, have well-documented pharmacological activities. Glycosides are known for their cardioprotective and anti-inflammatory properties, while steroids exhibit anti-inflammatory and antimicrobial effects [3,27,28]. Flavonoids and phenolic compounds, in addition to their antioxidant roles, have shown potential as anti-cancer agents by modulating oxidative stress pathways and inducing apoptosis in tumor cells [29]. These bioactive compounds collectively underscore the potential of *B. racemosa* as a source of therapeutic agents for conditions such as inflammation, infections, and oxidative stress-related disorders [30].

A notable observation in this study is the influence of solvent polarity on the extraction efficiency of phytochemicals. Ethanol and water extracts exhibited a broader spectrum of bioactive compounds, consistent with earlier findings [12] that polar solvents effectively extract flavonoids, phenolics, and other polar phytochemicals, including flavonoids and phenolics, which are crucial for antioxidant activity [31]. This highlights the importance of selecting appropriate solvents for optimizing the yield of specific bioactive compounds, which is critical for standardizing herbal formulations [32].

Despite these promising findings, the study has certain limitations. While qualitative phytochemical screening provides valuable insights, quantitative estimation of key bioactive compound would offer a more comprehensive understanding of their therapeutic potential. For instance, antioxidant activity assays (e.g., 2,2-diphenyl-1-picrylhydrazyl [DPPH] and 3-ethylbenzothiazoline-6-sulphonic acid [ABTS] assays) and in vitro anti-inflammatory studies could confirm the bioactivity of the extracts [33]. Furthermore, isolating and characterizing the active compounds, along with

assessing their mechanisms of action, would strengthen the evidence for their medicinal applications.

Future research should focus on isolating and characterizing the active compounds, determining their mechanisms of action, and assessing their efficacy in *in vivo* models. Additionally, exploring the synergistic effects of these phytochemicals may reveal enhanced therapeutic potential, as seen in other plant-based formulations [34]. The integration of omics technologies, such as metabolomics and proteomics, could further elucidate the phytochemical pathways and bioactivity profiles of *B. racemosa*, paving the way for its application in drug discovery [35].

5. Conclusions

This study provides a preliminary yet comprehensive phytochemical and physicochemical profile of *B. racemosa* leaves, highlighting their rich content of bioactive compounds such as flavonoids, phenolic compounds, glycosides, and tannins. The physicochemical properties confirm the high quality and stability of the leaf powder, supporting its use in pharmacological applications.

Our findings validate existing literature while offering new insights into the phytochemical diversity of *B. racemosa* across different solvent extractions. Such profiling is crucial for the standardization and quality control of herbal materials, ensuring the purity and efficacy of their therapeutic formulations.

Overall, this study lays the groundwork for future research aimed at unlocking the full therapeutic potential of *B. racemosa*. By integrating traditional knowledge with modern scientific approaches, the medicinal applications of this plant can be expanded, contributing to advancements in phytotherapy and drug discovery.

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References:

1. Ale, Y.; Rana, S.; Nainwal, N.; Rawat, S.; Butola, M.; Zainul, R.; Jakhmola, V. Phytochemical Screening and Green Synthesis of Antibacterial Silver Nanoparticles of *Sapindus mukorossi* Fruit Extracts. *Res. J. Pharm. Technol.* **2023**, *16(12)*, 5643–5649. DOI: 10.52711/0974-360X.2023.00912
2. World Health Organization. WHO Traditional Medicine Strategy 2014–2023. *World Health Organization: Geneva, Switzerland*, **2013**. Available online: https://www.who.int/publications/i/item/9789241506096 (accessed on 31 November 2024).
3. Newman, D.J.; Cragg, G.M. Natural Products as Sources of New Drugs over the Last 25 Years. *J. Nat. Prod.* **2007**, *70(3)*, 461–477. DOI: 10.1021/np068054v
4. Tabassum, S.; Ahmad, S.; Ali, B.; Usman, F.; Jabeen, Q.; Sajid-ur-Rehman, M.; Ahmed, M.; Zubair, H.M.; Alkazmi, L.; Batiha, G.E.; Basit, A. Chemical Profiling and Evaluation of Toxicological, Antioxidant, Anti-Inflammatory, Anti-Nociceptive and Tyrosinase Inhibitory Potential of *Portulacaria afra* Using In-Vitro, In-Vivo and In-Silico Studies. *Arab. J. Chem.* **2023**, *16(6)*, Art. No: 104784. DOI: 10.1016/j.arabjc.2023.104784
5. Rates, S.M. Plants as Source of Drugs. *Toxicon.* **2001**, *39(5)*, 603–613. DOI: 10.1016/s0041-0101(00)00154-9
6. Adil, M.; Filimban, F.Z.; Ambrin; Quddoos, A.; Sher, A.A.; Naseer, M. Phytochemical Screening, HPLC Analysis, Antimicrobial and Antioxidant Effect of *Euphorbia parviflora* L. (Euphorbiaceae Juss.). *Sci. Rep.* **2024**, *14(1)*, Art. No: 5627. DOI: 10.1038/s41598-024-55905-w
7. Kulshreshtha, M.; Shukla, K.S.; Tiwari, G.A.; Singh, M.P.; Singh, A. Pharmacognostical, Phytochemical and Pharmacological Aspects of *Quisqualis indica*: An Update. *J. Nat. Sci. Med.* **2018**, *1(2)*, 41–47. **DOI:** 10.4103/JNSM.JNSM_20_18.
8. Gurib-Fakim, A. Medicinal Plants: Traditions of Yesterday and Drugs of Tomorrow. *Mol. Aspects Med.* **2006**, *27(1)*, 1–93. DOI: 10.1016/j.mam.2005.07.008
9. Prabhu, S.; Vijayakumar, S.; Ramasubbu, R.; Praseetha, P.K.; Karthikeyan, K.; Thiyagarajan, G.; Sureshkumar, J.; Prakash, N. Traditional Uses, Phytochemistry and Pharmacology of *Bauhinia racemosa* Lam.: A Comprehensive Review. *Futur. J. Pharm. Sci.* **2021**, *7*, Art. No: 101. DOI: 10.1186/s43094-021-00251-1
10. Panda, P.; Das, D.; Dash, P.; Ghosh, G. Therapeutic Potential of *Bauhinia racemosa*—A Mini Review. *Int. J. Pharm. Sci. Rev. Res.* **2015**, *32*, 169–179.
11. Kamboj, V.P. Herbal Medicine. *Curr. Sci.* **2000**, *78(1)*, 35–39.
12. Cowan, M.M. Plant Products as Antimicrobial Agents. *Clin. Microbiol. Rev.* **1999**, *12(4)*, 564–582. DOI: 10.1128/CMR.12.4.564
13. Arokiarajan, M.S.; Christopher, J.J.; Akram, S.A.W.; Ahmed, N.Z.; Meena, R.; Ahmed, K.K. Phyto-Chemical, Analytical Evaluation and Biological Attributes of Rare Medicinal Species of Flowering Shrub *Bauhinia racemosa* L. *Adv. Tradit. Med. (ADTM)* **2024**, 1181–1203. DOI: 10.1007/s13596-024-00761-2
14. Heinrich, M.; Williamson, E.M.; Gibbons, S.; Barnes, J. Fundamentals of Pharmacognosy and Phytotherapy E-BOOK. Elsevier Health Sciences: London, UK, **2012**.
15. Adugna, H.; Ezez, D.; Guadie, A.; Tefera, M. Phytochemical Profiling, Evaluation of Heavy Metal Load, Antioxidant and Antibacterial Activity of Various Medicinal Plants. *J. Agric. Food Res.* **2024**, *16*, Art. No: 101190. DOI: 10.1016/j.jafr.2024.101190

16. Fabricant, D.S.; Farnsworth, N.R. The Value of Plants Used in Traditional Medicine for Drug Discovery. *Environ. Health Perspect.* **2001**, *109(Suppl. 1)*, 69–75. DOI: 10.1289/ehp.01109s169

17. Mokaizh, A.A.; Nour, A.H.; Alazaiza, M.Y.; Mustafa, S.E.; Omer, M.S.; Nassani, D.E. Extraction and Characterization of Biological Phytoconstituents of *Commiphora gileadensis* Leaves Using Soxhlet Method. *Processes* **2024**, *12(8)*, Art. No: 1567. DOI: 10.3390/pr12081567

18. Khandelwal, K.R.; Sethi, V. Practical Pharmacognosy. 32nd ed.; Nirali Prakashan: Pune, India, **2022**.

19. Kokate, C.K. Practical Pharmacognosy. 5th ed.; Vallabh Prakashan: New Delhi, India, **2022**.

20. Tiwari, P.; Kumar, B.; Kaur, M.; Kaur, G.; Kaur, H. Phytochemical Screening and Extraction: A Review. *Int. Pharm. Sci.* **2011**, 1, 98–106.

21. Sharma, A.; Shanker, C.; Tyagi, L.K.; Singh, M.; Rao, C.V. Herbal Medicine for Market Potential in India: An Overview. *Acad. J. Plant Sci.* **2008**, *1(2)*, 26–36.

22. Verma, S.; Singh, S.P. Current and Future Status of Herbal Medicines. *Vet. World.* **2008**, *1(11)*, 347.

23. Lobo, V.; Patil, A.; Phatak, A.; Chandra, N. Free Radicals, Antioxidants and Functional Foods: Impact on Human Health. *Pharmacogn. Rev.* **2010**, *4(8)*, 118-126. DOI: 10.4103/0973-7847.70902

24. World Health Organization. Quality Control Methods for Medicinal Plant Materials. World Health Organization: Geneva, Switzerland, **1998**. Available online: https://www.who.int/publications/i/item/9241545100 (accessed on 31 November 2024).

25. Sofowora, A.; Ogunbodede, E.; Onayade, A. The Role and Place of Medicinal Plants in the Strategies for Disease Prevention. *Afr. J. Tradit. Complement. Altern. Med.* **2013**, *10(5)*, 210–229. DOI: 10.4314/ajtcam.v10i5.2

26. Dogara, F.; et al. Phytochemical and Antimicrobial Analysis *of Bauhinia racemosa*. *Merit Res. J. Food Sci. Technol.* **2023**, *8(2)*, 024–032. DOI: 10.5281/zenodo.8398740

27. Fürst, R.; Zündorf, I.; Dingermann, T. New Knowledge About Old Drugs: The Anti-Inflammatory Properties of Cardiac Glycosides. *Planta Med.* **2017**, *83 (12/13)*, 977–984. DOI: 10.1055/s-0043-105390.

28. Bucki, R.; Leszczyńska, K.; Byfield, F. J.; Fein, D. E.; Won, E.; Cruz, K.; Namiot, A.; Kułakowska, A.; Namiot, Z.; Savage, P. B.; Diamond, S. L.; Janmey, P. A. Combined Antibacterial and Anti-Inflammatory Activity of a Cationic Disubstituted Dexamethasone-Spermine Conjugate. *Antimicrob. Agents Chemother.* **2010**, *54*, 2525–2533. DOI: 10.1128/aac.01682-09

29. Harborne, J.B. Phytochemical Methods: A Guide to Modern Techniques of Plant Analysis. 3rd ed.; Springer: New York, USA, **1998**.

30. Patel, D.K.; Patel, K.; Dhanabal, S.P. Phytochemical Standardization of *Aloe vera* Extract by HPTLC Techniques. *J. Acute Dis.* **2012**, *1(1)*, 47–50. DOI: 10.1016/S2221-6189(13)60011-6

31. Sultana, B.; Anwar, F.; Ashraf, M. Effect of Extraction Solvent/Technique on the Antioxidant Activity of Selected Medicinal Plant Extracts. *Molecules* 2009, *14*(6), 2167–2180. DOI: 10.3390/molecules14062167

32. Tiwari, B.K.; Valdramidis, V.P.; O'Donnell, C.P.; et al. Application of Natural Antimicrobials for Food Preservation. *J. Agric. Food Chem.* 2009, *57* (*14*), 5987–6000. DOI: 10.1021/jf900668n

33. Singleton, V.L.; Rossi, J.A. Colorimetry of Total Phenolics with Phosphomolybdic-Phosphotungstic Acid Reagents. *Am. J. Enol. Vitic.* **1965**, *16*, 144–158. DOI: 10.5344/ajev.1965.16.3.144

34. Williamson, E.M. Synergy and Other Interactions in Phytomedicines. *Phytomedicine* **2001**, *8(5)*, 401–409. DOI: 10.1078/0944-7113-00060

35. Dixit, T.; Tiwari, A.; Bose, S.; Kulkarni, H.; Suthar, J.; Ravindran, S. Metabolism of Phytochemicals. In: *Drug Metabolism*. Dunnington, K. Ed. **2021.** IntechOpen. doi: 10.5772/intechopen.100569**.** Available from: http://dx.doi.org/10.5772/intechopen.100569