PROSPECTS IN PHARMACEUTICAL SCIENCES

Prospects in Pharmaceutical Sciences, 22(4), 155-159 https://prospects.wum.edu.pl/

Original Article

PHYTOCHEMICAL PROFILE AND PHYSICOCHEMICAL ANALYSIS OF THE LEAVES OF ZIZIPHUS MAURITIANA

Deepak Jha^{*}, Ashish Singh Parihar

Department of Pharmacy, Faculty of Pharmacy, Oriental University, 453555 Indore, India.

* Correspondence, e-mail: drdbjmw@gmail.com

Received: 12.11.2024 / Accepted: 27.11.2024 / Published: 31.12.2024

ABSTRACT

Ziziphus mauritiana Lam. (Indian jujube) is traditionally used to treat asthma, digestive disorders, skin conditions, and ulcers. Despite its wide medicinal applications, comprehensive phytochemical and physicochemical profiling of Z. mauritiana leaves remains limited. The current study aimed to establish a detailed phytochemical and physicochemical profile of Z. mauritiana leaves across different solvent extracts to aid in standardization and quality control. Fresh Z. mauritiana leaves were collected, authenticated, dried, and powdered. Soxhlet extraction was performed using seven solvents: acetone, water, chloroform, dichloromethane, ethanol, ethyl acetate, and petroleum ether. Physicochemical analyses included ash content, extractive values, moisture content, and pH. Qualitative phytochemical screening was conducted to identify bioactive compounds such as carbohydrates, proteins, flavonoids, tannins, and alkaloids. Physicochemical analysis revealed total ash (7.5%), acid-insoluble ash (2.5%), and moisture content (7.3%). Extractive values varied across solvents, with ethyl acetate showing the highest yield (0.76%). Phytochemical screening demonstrated the presence of carbohydrates, proteins, amino acids, steroids, flavonoids, phenolic compounds, tannins, glycosides, and alkaloids, with variation across solvents. Z. mauritiana leaves exhibit a diverse range of bioactive compounds with potential antioxidant, anti-inflammatory, and antimicrobial activities, supporting their traditional medicinal use. Future research should focus on quantifying these compounds using advanced analytical methods and exploring other plant parts for a comprehensive profile.

KEYWORDS: *Ziziphus mauritiana*, phytochemical screening, bioactive compounds, physicochemical analysis, ash value, extractive value.

Article is published under the CC BY license.

1. Introduction

Nature serves as a significant source of diverse resources, with plants representing one of its most remarkable gifts. Medicinal plants, in particular, play a crucial role in the treatment of various ailments due to their primary and secondary metabolites, forming the foundation of traditional medicine systems [1-4]. Plant-derived drugs are widely popular for their minimal side effects, making them a preferred choice in many therapeutic contexts [1,5].

The preservation of medicinal plants within their natural habitats is essential due to their wide-ranging applications and valued active compounds across multiple sectors [6]. *Ziziphus mauritiana Lam.* (*Z. mauritiana*), commonly known as Indian jujube or Ber, belongs to the Rhamnaceae family and is a large evergreen shrub or small tree, typically growing to a height of 9 to 15 meters [7,8]. The plant is widely distributed across regions including India, Nepal, Afghanistan, Pakistan, China, Malaysia, Australia, Japan, the Philippines, and southern Africa [8].

Z. mauritiana has been traditionally used for the treatment of various conditions such as asthma, allergies, diabetes, digestive disorders, obesity, skin infections, ulcers, and urinary issues [9-12]. In Ayurvedic medicine, the roots of this plant are recommended for ailments like biliousness, cough, headaches, and nausea, and are valued for their cooling and bitter tonic properties [8,13,14]. Formulations made from the aqueous and ethanolic extracts of Z. mauritiana demonstrate antiemetic, anti-inflammatory, analgesic, and antipyretic properties [7]. Furthermore, the leaves of this plant are known for their cooling, antipyretic, and anthelminthic effects, which make them beneficial for conditions such as asthma, stomatitis, and typhoid fever [8,11,13].

Although some literature exists on the phytochemical and physicochemical profile of *Z. mauritiana* leaf extracts, comprehensive profiling remains limited. Therefore, this investigation was undertaken to establish a detailed profile of the leaves across different solvents, aiding in the standardization and quality control of this valuable medicinal plant material.

2. Materials and Methods

2.1. Plant Material

Fresh leaves of Z. mauritiana were collected in August and September from the Dharashiv (Osmanabad) district, Maharashtra, India. Dr. A. S. Linge, Head of the Department of Botany at Venkatesh Mahajan Senior College, Dharashiv – affiliated with Dr. Babasaheb Ambedkar Marathwada University, Chhatrapati Sambhaji Nagar, Maharashtra – authenticated the plant material.

2.2. Preparation of Powder and Extracts

The collected leaves were thoroughly rinsed with reverse osmosis-purified water to remove surface impurities. The clean leaves were air-dried in a shaded area for several days to prevent photodegradation and then finely powdered using an electric grinder.

For Soxhlet extraction, 100 grams of the powdered leaf material was extracted sequentially with 300 mL of each solvent: acetone, water, chloroform, dichloromethane, ethanol, ethyl acetate, and petroleum ether. The extraction process was carried out for 24 hours per solvent at a consistent temperature appropriate for each solvent's boiling point. The procedure was conducted in triplicate to ensure reproducibility and reliability of the results.

Post extraction, the mixtures were filtered using Whatman filter paper No. 1, and the solvents were removed under reduced pressure using a rotary evaporator. The resulting extracts were stored in airtight containers at $4^{\circ}C$ for further analysis.

2.3. Physicochemical Analysis

The powdered leaves were subjected to several physicochemical tests, including the assessment of ash content, extractive values in various solvents, moisture content, and potential of hydrogen (pH) measurement.

2.4. Qualitative Phytochemical Screening

Different extracts (acetone, chloroform, dichloromethane, ethanol, ethyl acetate, petroleum ether, and water) were screened for secondary metabolites, such as carbohydrates, fixed oils, flavonoids, glycosides, proteins, phenolic compounds, and tannins, following methodologies outlined by Khandelwal et al. and Kokate et al. [15,16].

3. Results

3.1. Physicochemical Properties

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

3.2. Qualitative Phytochemical Screening

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

Table 1. Physicochemical Properties of Z. mauritianaLeaf Powder

Parameter	Result	Unit
Ash value		
Total Ash	7.5	% (w/w)
Acid-Insoluble Ash	2.5	% (w/w)
Water-Soluble Ash	4	% (w/w)
Sulfate Ash	5	% (w/w)
Water	0.57	% (w/w)
Ethanol	0.12	% (w/w)
Acetone	0.64	% (w/w)
Ethyl acetate	0.76	% (w/w)
Dichloromethane	0.40	% (w/w)
Chloroform	0.40	% (w/w)
Petroleum ether	0.52	% (w/w)
Moisture content		
Loss on drying at 110°C	7.3	%
pН		
pH of 1.00% w/v aqueous solution	6	_
pH of 10.00% w/v aqueous solution	7	_

 Table 2. Phytochemical Screening of Z. mauritiana 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 current study presents a comprehensive phytochemical profile and physicochemical analysis of *Z. mauritiana* leaves. Qualitative screening revealed a diverse array of bioactive compounds, including carbohydrates, proteins, amino acids, steroids, glycosides, saponins, phytosterols, flavonoids, phenolic compounds, tannins, alkaloids, enzymes, acidic compounds, organic acids, and vitamins. These findings highlight the potential medicinal properties of *Z. mauritiana*, particularly its antioxidant, anti-inflammatory, and antimicrobial activities.

The phytochemical composition of *Z. mauritiana* observed in this study is consistent with previous reports on other *Ziziphus* species. For instance, *Z. jujuba* and *Z. spina-christi* are known to possess high levels of flavonoids and phenolic compounds with potent antioxidant properties [17,18]. This study, however, extends existing knowledge by providing a detailed profile across multiple solvent extracts, offering a broader understanding of the phytochemical diversity in *Z. mauritiana*. The detection of saponins and tannins aligns with findings from other studies that emphasize their anti-inflammatory and antimicrobial properties [19]. Such comprehensive profiling is crucial for the standardization and quality control of this medicinal plant.

The presence of flavonoids and phenolic compounds in *Z. mauritiana* suggests a strong antioxidant potential. These compounds are known to scavenge free radicals and protect against oxidative stress, which is crucial for managing oxidative stress-related diseases [20]. Studies have shown that the antioxidant capacity of *Z. mauritiana* leaves is comparable to other species like *Z. jujuba* and *Z. spina-christi*, which are rich in flavonoids and phenolic compounds with potent antioxidant properties [18,22]. The high levels of these compounds in *Z. mauritiana* support its traditional use in preventing and treating conditions associated with oxidative damage [23].

Saponins and tannins detected in *Z. mauritiana* are known for their anti-inflammatory properties. These compounds inhibit the production of pro-inflammatory cytokines and enzymes, thereby reducing inflammation. Research indicates that the anti-inflammatory effects of *Z. mauritiana* are significant and align with findings from other studies on *Ziziphus* species [21,24]. For instance, the anti-inflammatory activity of *Z. mauritiana* has been demonstrated in various in vitro and in vivo models, highlighting its potential in managing inflammatory conditions [25]. The antimicrobial properties of *Z. mauritiana* are attributed to its rich phytochemical composition, including alkaloids, flavonoids, and saponins. These compounds exhibit broad-spectrum antimicrobial activity against various pathogens. Studies have shown that extracts from *Z. mauritiana* leaves are effective against bacterial strains such as *Staphylococcus aureus*, *Escherichia coli*, and *Salmonella typhimurium* [26,27]. The antimicrobial activity of *Z. mauritiana* supports its traditional use in treating infections and highlights its potential as a natural antimicrobial agent [21,28].

A key limitation of this study is the qualitative nature of the phytochemical screening, which does not quantify the concentrations of identified compounds. Future studies should utilize quantitative analytical techniques such as high-performance liquid chromatography or mass spectrometry for accurate compound quantification. Additionally, this study focused solely on the leaves of *Z. mauritiana*. Future research should explore other plant parts, such as bark and roots, to provide a more comprehensive phytochemical profile.

Future studies should adopt advanced analytical methods to quantify identified phytochemicals and evaluate their specific biological activities, especially their antioxidant, anti-inflammatory, and antimicrobial properties. Comparative analyses across different parts of *Z. mauritiana* and other *Ziziphus* species are also recommended.

5. Conclusions

This study offers a detailed phytochemical and physicochemical profile of *Z. mauritiana* leaves, highlighting their rich composition of bioactive compounds, including carbohydrates, proteins, amino acids, steroids, glycosides, flavonoids, phenolic compounds, tannins, and alkaloids across multiple solvent extracts. These findings reinforce the documented medicinal properties of *Z. mauritiana* and its traditional uses in treating ailments such as asthma, fever, and skin infections. The presence of flavonoids and phenolic compounds indicates strong antioxidant potential, while glycosides and saponins suggest anti-inflammatory and antimicrobial properties, validating its use in herbal medicine.

While the qualitative nature of this screening is a limitation in determining the exact concentrations of these compounds, future research should focus on quantitative analysis and advanced analytical techniques to accurately assess compound concentrations and validate pharmacological activities. Exploring other plant parts and conducting bioactivity assays would further substantiate the medicinal potential of *Z. mauritiana*, supporting the development of standardized herbal formulations and nutraceutical products.

Author Contributions: Conceptualization, D.J. and A.S.P.; methodology, D.J.; validation, D.J., and A.S.P.; investigation, D.J.; resources, D.J.; data curation, D.J.; writing—original draft preparation, D.J.; writing—review and editing, A.S.P.; visualization, D.J.; supervision, A.S.P.; project administration, D.J.; funding acquisition, D.J. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Acknowledgments: I would like to thank Dr. Amol Joshi, Principal of ASPM's K. T. Patil College of Pharmacy, Dharashiv, Maharashtra, for granting me permission to conduct the experiments and for providing the necessary facilities. I also extend my thanks to Swati Pawar (Assistant Professor) and Priyanshu Ram (Pharm.D student) for their assistance with the experiments.

Conflicts of Interest: The authors declare no conflict of interest.

References:

- Mendhekar, S. Y.; Rachh, P. R. Pharmacognostic, Physicochemical and Preliminary Phytochemical Investigation of *Trapa natans* Linn. Root. *Res. J. Pharm. Technol.* 2023, *16* (11), 5444-5452. DOI: 10.5958/0974-360X
- 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
- Davidson-Hunt, I. Ecological Ethnobotany: Stumbling Toward New Practices and Paradigms. MASA J. 2000, 16 (1), 1-3.
- Mohammed, F. S.; Sevindik, M.; Uysal, I.; Sevindik, E.; Akgül, H. A Natural Material for Suppressing the Effects of Oxidative Stress: Biological Activities of *Alcea kurdica*. *Biol. Bull.* **2022**, *49* (Suppl 2), S59-S66. DOI: 10.1134/S1062359022140102
- Mohammed FS, Pehlivan M, Sevindik E, Akgul H, Sevindik M, Bozgeyik I, Yumrutas O. Pharmacological properties of edible Asparagus acutifolius and Asparagus officinalis collected from North Iraq and Turkey (Hatay). Acta Alimentaria 2021, 50(1), 136-43. DOI: 10.1556/066. 2020.00204
- Alsaedi, S.; Aljeddani, G. Phytochemical Analysis and Bioactivity Screening of Primary and Secondary Metabolic Products of Medicinal Plants in the Valleys of Medina Region Saudi Arabia. Adv. Biol. Chem. 2022, 12 (4), 92-115. DOI: 10.4236/abc.2022.124009
- Siddique, N.; Ved, A.; Shukla, K. S.; Nigam, A. K. Standardization and Pharmacological Evaluation of Ziziphus mauritiana Extract for Sedative and Anticonvulsant Activity in Mice and Rat. Anti-Inflamm. Antiallergy Agents Med. Chem. 2024, 23 (1), 31-38. doi: 10.2174/0118715230276586231215045816
- 8. Ambrin, A.; Adil, M.; Filimban, F. Z.; Naseer, M. Chemical Profiling and Biological Activities of *Ziziphus*

mauritiana var. spontanea (Edgew.) R. R. Stewart ex Qaiser & Nazim. and Oenothera biennis L. J. Food Qual. 2024, 2024 (1), 7318407. DOI: 10.1155/2024/ 7318407

- Paudel, M. R.; Poudeyal, M. R.; Devkota, H. P. Ziziphus spp. (Ziziphus jujuba Mill., Ziziphus mauritiana Lam.). In Himalayan Fruits and Berries; Academic Press: 2023; pp 491-497.
- Mohankumar, R.; Prakash, S. E.; Irfan, N.; Mohanraj, S.; Kumarappan, C. Evaluation of Analgesic, Anti-Inflammatory, and Antipyretic Activities of *Ziziphus mauritiana* Lam Leaves in Animal Models. *Pharmacol. Res.-Mod. Chin. Med.* **2022**, *4*, Art. No: 100153. DOI: 10.1016/j.prmcm.2022.10015
- Ramar, M. K.; Chidambaram, K.; Chandrasekaran, B.; Kandasamy, R. Standardization, In-Silico and In-Vivo Safety Assessment of Methanol Extract of Ziziphus mauritiana Lam Leaves. Regul. Toxicol. Pharmacol. 2022, 131, Art. No: 105144. 10.1016/j.yrtph.2022. 105144
- Pandey, J.; Gaire, S.; Sharma, K.; Pun, D.; Gyawali, A.; Lamichhane, G.; Budean, D.; Devkota, H. P. Exploring the Potentials of *Ziziphus mauritiana* Lam. Seed Kernel Oil as Pharmaceutical Oil Base: Physicochemical Characterization and Ketoconazole Soap Formulation. *Heliyon* 2023, 9 (11), Art. No: e21034. DOI: 10.1016/j.heliyon.2023.e2103
- Khan, U. A.; Qadri, R.; Akram, M. T.; Khan, M. A.; Nisar, N.; Azam, M.; Khan, M. M.; Hussain, K. Physio-Chemical Assessment of Jujube (*Ziziphus* mauritiana L.) Genotypes at Three Ripening Stages Grown Under Cholistan Desert, Pakistan. J. Anim. Plant Sci. 2023, 33 (2), 285-295. DOI: 10.36899/JAPS. 2023.2.0620
- 14. Goyal, M.; Nagori, B. P.; Sasmal, D. Review on Ethnomedicinal Uses, Pharmacological Activity and Phytochemical Constituents of Ziziphus mauritiana (Z. jujuba Lam., non Mill). Spatula DD 2012, 2 (2), 107-116.
- 15. Khandelwal, K. R.; Sethi, V. *Practical Pharmacognosy*; 32nd ed.; Nirali Prakashan: **2022**.
- 16. Kokate, C. K. *Practical Pharmacognosy*; 5th ed.; Vallabh Prakashan: 2022.
- Sinan, K. I.; Dall'Acqua, S.; Sut, S.; Uba, A. I.; Etienne, O. K.; Ferrante, C.; Ahmad, J.; Zengin, G. Ziziphus mauritiana Lam. Bark and Leaves: Extraction, Phytochemical Composition, In Vitro Bioassays and In Silico Studies. *Plants* 2024, *13* (16), Art. No: 2195. DOI: 10.3390/plants13162195
- Sakna, S. T.; Maghraby, Y. R.; Abdelfattah, M. S.; Farag, M. A. Phytochemical Diversity and Pharmacological Effects of Triterpenes from Genus Ziziphus: A Comprehensive Review. Phytochem. Rev. 2023, 22 (6), 1611-1636. DOI: 10.1007/s11101-022-09835-y
- Aldhanhani, A. R.; Kaur, N.; Ahmed, Z. F. Antioxidant Phytochemicals and Antibacterial Activities of Sidr (*Ziziphus* spp.) Leaf Extracts. In XXXI Int. Hortic. Congr. (IHC2022): Int. Symp. Integr. Approaches Prod. Qual. 2022, 1353, 323-332.

- Mohammed FS, Günal S, Pehlivan M, Doğan M, Sevindik M, Akgül H. Phenolic content, antioxidant and antimicrobial potential of endemic *Ferulago platycarpa*. *Gazi Univ. J. Sci.* 2020, 33(4), 670-677. DOI: 10.35378/ gujs.707555
- Butt, S. Z.; Hussain, S.; Munawar, K. S.; Tajammal, A.; Muazzam, M. A. Phytochemistry of *Ziziphus mauritiana*; Its Nutritional and Pharmaceutical Potential. *Sci. Ing. Rev.* 2021, 5 (2), 1-5. DOI:10.32350/sir.52.01
- Naaz, F.; Agari, N.; Singh, A. Medicinal Properties of Ziziphus mauritiana: A Review Article. Int. J. Pharm. Pharm. Res. 2020, 19, 219-230.
- 23. Jeet, S. Ziziphus mauritiana Leaf: Africa's Skin Rejuvenation Secret. NewsByte, 2024. https://www.newsbytesapp.com/news/lifestyle/ziziphu s-mauritiana-leaf-africas-skin-rejuvenation-secret/story (accessed 2024-11-23).
- Talmale, S.; Bhujade, A.; Patil, M. Anti-Allergic and Anti-Inflammatory Properties of *Ziziphus mauritiana* Root Bark. *Food Funct.* 2015, 6 (9), 2975-2983. DOI: 10.1039/c5fo00270b
- 25. Basu, S. Zizyphus Aka Ziziphus jujuba: Health Benefits, Uses, Ayurvedic Indications, Side Effects and More! Netmeds.com, 2024.https://www.netmeds.com/healthlibrary/post/zizyphus-aka-ziziphus-jujuba-healthbenefits-uses-ayurvedic-indications-side-effects-andmore (accessed 2024-11-23).
- 26. Salih, R.; Gibreel, H.; Ahmed, D.; Hammad, Z.; Ahmed, A. Antimicrobial Effect of Aqueous and Ethanolic Leaf Extracts of *Ziziphus* Species Against Animal Bacterial Pathogens. *Curr. Trends For. Res.* **2019**, *3*, Art. No: 1036. DOI: 10.29011/2638-0013.100036
- Akanda, M. K.; Hasan, A. N. Characterization of Pharmacological Properties of Methanolic Seed and Stem Bark Extracts of *Ziziphus mauritiana* (BAU Kul) Using In-Vitro and In-Vivo Animal (Swiss Albino Male Mice) Model. *Clin. Phytosci.* 2021, 7, 1-3. DOI: 10.1186/s40816-020-00246-0
- Yahia, Y.; Benabderrahim, M. A.; Tlili, N.; Bagues, M.; Nagaz, K. Bioactive Compounds, Antioxidant, and Antimicrobial Activities of Extracts from Different Plant Parts of Two Ziziphus Mill. Species. PLoS One 2020, 15 (5), Art. No: e0232599. doi: 10.1371/journal.pone. 0232599