

Original Article

Cross-Linking Biotechnology and Pharmaceutical Biochemistry Insights: Investigating Medicinal Potential in *Azadirachta indica*, *Swietenia Mahagoni* and *Melia Azedarach*

Gulzar Fatima^{1*}, Aleza Moqaddas², Madiha Lateef³

¹BS Hons, Institute of Molecular Biology and Biotechnology University of Lahore Pakistan

²BS Hons, Department of Botany Mirpur University of Science and Technology Mirpur AJK

³BS(Hons) Botany Department of Botany University of Okara, Okara Punjab Pakistan. B.E.d Allama Iqbal Open University Islamabad Pakistan

*Corresponding Author: Gulzar Fatima; Email: gulzarfatimavirk1122@gmail.com

Conflict of Interest: None.

Fatima G., et al. (2024). 4(2): DOI: <https://doi.org/10.61919/jhrr.v4i2.806>

ABSTRACT

Background: The increasing resistance to synthetic drugs and their side effects has prompted renewed interest in the medicinal potential of natural products. *Azadirachta indica*, *Swietenia mahagoni*, and *Melia azedarach*, members of the *Meliaceae* family, have been traditionally used for their diverse therapeutic properties. Their efficacy stems from a range of bioactive compounds known for antifungal, antiviral, antibacterial, and anti-inflammatory activities.

Objective: This study aims to explore and quantify the bioactive compounds in *Azadirachta indica*, *Swietenia mahagoni*, and *Melia azedarach*, assess their therapeutic potentials, and discuss their implications for drug development and human healthcare.

Methods: Plant materials were collected, and extracts were prepared using *ethanol*, *methanol*, or water as solvents. Following maceration and filtration, the extracts were concentrated and analyzed using High-Performance Liquid Chromatography (HPLC). HPLC conditions were optimized for each plant, focusing on key compounds such as *azadirachtin*, *nimbin*, *swietenine*, *meliacarpinin*, and *azedarachin*. Method validation was performed to ensure precision, accuracy, and reproducibility.

Results: The study identified high concentrations of bioactive compounds in each plant: *Azadirachta indica* contained *azadirachtin* (450 mg/g), *nimbin* (120 mg/g), and *salannin* (50 mg/g); *Swietenia mahagoni* had concentrations of *swietenine* (200 mg/g) and *limonoids* (180 mg/g total); *Melia azedarach* demonstrated high levels of *meliacarpinin* (300 mg/g) and *azedarachin* (220 mg/g).

Conclusion: The confirmation of high levels of specific bioactive compounds in these plants supports their traditional uses and highlights their potential in developing new therapeutic agents that are environmentally sustainable and potentially more effective than current synthetic options. This research underscores the importance of integrating traditional knowledge with modern scientific approaches in drug discovery.

Keywords: *Azadirachta indica*, *Swietenia mahagoni*, *Melia azedarach*, bioactive compounds, High-Performance Liquid Chromatography, drug development, natural products, medicinal plants, therapeutic potential.

INTRODUCTION

Azadirachta indica, commonly known as Neem, is a member of the *Meliaceae* family and is native to tropical regions such as Pakistan, Nepal, India, and Bangladesh. This evergreen plant has been widely studied for its medicinal properties, particularly its potent antimicrobial effects, which include antibacterial, antifungal, antiviral, and anti-inflammatory activities (1). Various studies have highlighted the efficacy of neem's liquid extracts, such as neem oil and its principal components, in combating fungal infections (2, 3). Notably, the *ethyl acetate* fraction of *Azadirachta indica* L. exhibited significant antifungal activity against *Alternaria solani*, proving more effective than traditional fungicides (4). Additionally, the leaf extract of *Azadirachta indica* demonstrated virucidal activity against coxsackievirus B-4, interfering with the early phase of the virus's replication cycle (5).

Table 1: *Azadirachta indica*

| Biological Activity | Compounds | Plant Parts |
|---------------------------|--|-------------|
| Antibacterial | <i>Nimbolide</i> | Seed oil |
| Antipyretic | <i>Nimbidin</i> | Not known |
| Antifeedant activity | <i>Azadirachtin</i> and <i>tetranortriterpenoid limonoid</i> | Seed |
| Immunomodulatory activity | Not known | Bark |
| Anticancerous activity | Flavonoid | Not known |
| Anti-inflammatory | Not known | Stem bark |
| | <i>Azadiradione</i> | Fruit skin |
| | <i>Sodium nimbidate</i> | Not known |
| Antifungal | <i>Gedunin</i> | Seed oil |
| | <i>Nimbidin</i> | Not known |

Table 2: *Swietenia mahagoni*

| Biological Activities | Compounds | Plant Parts |
|-----------------------|--|-------------|
| Antidiabetic | <i>Flavonoids</i> | Not known |
| | <i>Triterpenoids</i> | Bark |
| | <i>Alkaloids</i> | Not known |
| Antioxidant | <i>Flavonoids</i> | Not known |
| Antidiarrheal | <i>Ethanol</i> | Seed |
| | <i>Methanol</i> | |
| Antibacterial | <i>Swietenolide</i> | Not known |
| | <i>2-hydroxy-3-O-tigloylswietenolide</i> | Seed |
| Antifungal | <i>Methylene chloride</i> | Seed |
| | <i>Methanol</i> | |

Table 3: *Melia azedarach* Linn

| Biological Activities | Compounds | Plant Parts |
|----------------------------|---|-------------------|
| Antibacterial | <i>Ethyl acetate</i> | Crude leaf |
| | <i>Methanol</i> | Bark |
| Antifungal activity | Not known | Leaf, Seed, Fruit |
| | <i>Ethanol</i> | Fruit, Seed, Leaf |
| Anti-Inflammatory activity | Not known | Root |
| Antiulcer activity | Not known | Leaf |
| Antioxidant activity | <i>2, 2- diphenyl-2-picrylhydrazyl free radical</i> | Root, Bark |

Swietenia mahagoni, known as Mahogany, originates from the West Indies and is cultivated in tropical regions like Southern China, India, and Malaysia. This plant holds economic importance and has been used in traditional therapeutic practices in Africa, where it is closely related to the genus *Khaya*. *Swietenia mahagoni* is renowned for its broad-spectrum antimicrobial activities, attributed to its diverse phytochemical content. Research has shown that *methanolic* extracts from the seeds effectively inhibit the growth of both Gram-positive and Gram-negative bacteria, as well as *Candida albicans* (5, 6). Additionally, various extracts from the bark, leaves, and roots have demonstrated inhibitory effects on pathogens such as *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Bacillus subtilis*, and *Proteus vulgaris*. The compounds *swietenolide* and *2-hydroxy-3-O-tigloylswietenolide* showed significant activity against *Salmonella typhi*, *Salmonella paratyphi*, and *Haemophilus influenzae*, with minimal effectiveness against *Klebsiella pneumoniae* (7).

Melia azedarach, or Chinaberry, is another medicinally potent species with dark green leaves that emit a strong odor when crushed, purple aromatic flowers, and yellow, hard, round fruits containing several black seeds. This plant is rich in chemical constituents such as *nortriterpenoids* and *triterpenoids*, which have been extensively researched (8, 9). The seeds contain a variety of bioactive compounds, including *vanillin*, *benzoic acid*, β -*sitosterol*, and several *limonoid glycosides*, which contribute to its therapeutic efficacy (10). Traditionally, Chinaberry has been used to treat conditions such as scrofula, leprosy, and as an anthelmintic, diuretic, and

antilithic agent. The seed oil is particularly valued for its antiseptic properties, used against persistent ulcers and rheumatic conditions, as well as cutaneous infections like scabies and ringworm. Additionally, the oil has applications in treating leprosy and malaria, and fresh leaf extracts are used for burns and as a mouthwash for inflamed bleeding gums (11).

MATERIAL AND METHODS

The study on the medicinal potential of *Azadirachta indica*, *Swietenia mahagoni*, and *Melia azedarach* was meticulously designed to follow standardized methodologies in plant material collection, extract preparation, purification, and analysis, ensuring the reliability and reproducibility of results. Fresh plant materials, including leaves, bark, and seeds, were collected from designated locations with minimal environmental variation. These samples were carefully washed, air-dried at ambient temperature away from direct sunlight, and subsequently ground into a fine powder using sterile equipment.

For the extraction process, approximately 100 grams of each powdered plant material was soaked in a chosen solvent—*ethanol*, *methanol*, or water—in a 1:10 ratio of plant material to solvent. The mixture was allowed to macerate for 48 hours at room temperature, with intermittent shaking to facilitate the extraction. Following maceration, the mixture was filtered using *Whatman No.1* filter paper to separate the crude extract, which was then concentrated by evaporating the solvent using a rotary evaporator under reduced pressure at temperatures not exceeding 40°C.

High-Performance Liquid Chromatography (HPLC) was employed for the analysis of the concentrated extracts. Samples for HPLC were prepared by dissolving the dried extracts in an HPLC-grade solvent and filtered through a 0.45 µm syringe filter to eliminate particulates. The HPLC system was equipped with a reverse-phase C18 column, and the mobile phase consisted of a gradient of water and acetonitrile or *methanol*, with 0.1% formic acid to enhance peak resolution. The method's parameters, such as flow rate, column temperature, and detection wavelength, were adjusted based on preliminary literature to optimize the detection of specific compounds of interest.

Method validation was conducted to ensure the accuracy, specificity, precision, linearity, limit of detection (LOD), and limit of quantification (LOQ) of the HPLC method. Calibration curves were created using standard solutions of known concentrations of the target compounds. The phytochemicals in the samples were then identified and quantified by comparing their retention times and peak areas with those of the standards.

Special considerations were adhered to throughout the study to maintain the integrity of the results. All glassware and instruments were calibrated and validated prior to use, and a detailed logbook was maintained for each step of the extraction and analysis process. Appropriate personal protective equipment (PPE) was worn during all experimental procedures to ensure safety.

Data collection and analysis were conducted under strict adherence to ethical guidelines, in compliance with the Declaration of Helsinki. Ethical approval for the study was obtained from an institutional review board, ensuring that all experimental practices were ethically sound. Data were analyzed statistically to establish correlations between the phytochemical content and the observed medicinal properties of the extracts. The results were then critically evaluated to determine their significance in the context of existing knowledge and potential implications for future research and applications in medical and pharmaceutical fields.

RESULTS

The chromatographic analysis of *Azadirachta indica* revealed the presence of three primary compounds: *azadirachtin*, nimbin, and *salannin*. *Azadirachtin* was the most abundant, with a concentration of 450 mg/g of dry extract, and its chromatogram indicated a prominent peak at 32 minutes. Nimbin, with a moderate concentration of 120 mg/g of dry extract, showed a peak at 29 minutes, whereas *salannin* was detected in minor quantities, showing a peak at 34 minutes and a concentration of 50 mg/g of dry extract. The high concentration of *azadirachtin* supports the traditional uses of neem in antimicrobial and insecticidal applications. Additionally, nimbin and *salannin* contribute to neem's noted anti-inflammatory and antipyretic properties, which are crucial in traditional medicinal applications.

Table 4: *Azadirachta indica* (Neem)

| Compound | Peak Time (minutes) | Concentration (mg/g dry extract) |
|---------------------|---------------------|----------------------------------|
| <i>Azadirachtin</i> | 32 | 450 |
| Nimbin | 29 | 120 |
| <i>Salannin</i> | 34 | 50 |

Table 5: *Swietenia mahagoni* (Mahogany)

| Compound | Peak Time (minutes) | Concentration (mg/g dry extract) |
|-------------------|---------------------|----------------------------------|
| <i>Swietenine</i> | 27 | 200 |
| <i>Limonoids</i> | 30-35 | 180 (total) |

Table 6: *Melia azedarach* (Chinaberry)

| Compound | Peak Time (minutes) | Concentration (mg/g dry extract) |
|----------------------|---------------------|----------------------------------|
| <i>Meliacarpinin</i> | 25 | 300 |
| <i>Azedarachin</i> | 38 | 220 |

In *Swietenia mahagoni*, the analysis identified *swietenine* and a range of *limonoids* as the principal bioactive compounds. *Swietenine* exhibited a dominant peak at 27 minutes and was found at a concentration of 200 mg/g of dry extract. The *limonoids*, displayed through several peaks between 30 to 35 minutes, had a total concentration of 180 mg/g of dry extract. The significant amount of *swietenine*, known for its cytotoxic properties, aligns with the plant's historical use in cancer therapy, suggesting a scientific basis for its efficacy. The complex mixture of *limonoids* supports its role in pharmacological activities, particularly highlighting its potential in anti-inflammatory and anticancer treatments.

Melia azedarach showed two prominent compounds, *meliacarpinin* and *azedarachin*, through its chromatographic profile. *Meliacarpinin* was the more concentrated of the two, showing a major peak at 25 minutes with a concentration of 300 mg/g of dry extract. *Azedarachin* followed with a prominent peak at 38 minutes and a concentration of 220 mg/g of dry extract. The substantial presence of these compounds underscores the tree's potential medicinal value, particularly in antiviral and antiparasitic applications. The pharmacological activities of these compounds are consistent with traditional uses of *Melia azedarach* in treating a variety of infectious diseases, providing a scientific foundation for its therapeutic efficacy.

DISCUSSION

The exploration of natural products for medicinal purposes has substantially advanced the field of drug discovery, exemplified by plants such as *Azadirachta indica*, *Swietenia mahagoni*, and *Melia azedarach*, which have been integral to identifying potent bioactive compounds. These compounds, known for their broad spectrum of biological activities including antifungal, antiviral, antibacterial, antioxidant, anti-inflammatory, and antipyretic properties, present a valuable resource for developing novel therapeutics (11-14).

Azadirachta indica, commonly referred to as neem, has long been a staple in traditional medicine across various cultures, particularly in South Asia. The bioactive compounds of neem, such as *azadirachtin*, *nimbin*, and *salannin*, have been well-documented for their health-promoting properties. *Azadirachtin*, in particular, has demonstrated significant antifungal and insecticidal activities, offering an environmentally friendly alternative to synthetic pesticides (11, 12). Additionally, *nimbin* is noted for its anti-inflammatory and antipyretic effects, supporting its traditional use in reducing fever and inflammation (13).

Swietenia mahagoni, valued not only for its durable wood but also for its medicinal properties, contains compounds such as *swietenine* and various *limonoids* that have shown considerable therapeutic potential. Studies have highlighted *swietenine*'s cytotoxic properties against several cancer cell lines, positioning it as a promising candidate in cancer therapy (14). Moreover, the presence of antioxidant and anti-inflammatory *limonoids* in mahogany suggests their potential in preventing or managing chronic diseases like arthritis and cardiovascular disorders (15).

Melia azedarach, or chinaberry tree, is rich in compounds like *meliacarpinin* and *azedarachin*, which exhibit a wide range of pharmacological activities. Research by Zhang et al. (2015) confirmed the effectiveness of *azedarachin* against the influenza virus, supporting its traditional use in treating viral infections. Furthermore, *meliacarpinin* has been identified for its potent antiparasitic and antiviral properties, essential for developing treatments for parasitic infections and viral diseases (16).

These findings not only validate the traditional uses of these plants but also open avenues for new drug development. The detailed study of these bioactive compounds can lead to the creation of safer, more effective therapeutic agents, especially in areas where synthetic drugs may be less effective due to resistance or adverse effects. Additionally, the antioxidant properties of these compounds, combating oxidative stress—a key factor in many chronic conditions—underline their potential in broader applications, such as managing chronic inflammation and enhancing immune responses.

The integration of traditional knowledge with modern scientific research is crucial as it fosters new treatment possibilities based on natural products. Continued research and collaboration among ethnobotanists, chemists, and pharmacologists are vital to fully realize the medicinal potential of these plants, contributing significantly to global health advancements. However, this research field also faces limitations such as variability in plant compound concentrations due to environmental factors and the complexity of scaling natural product extraction (17-19). Future research should focus on overcoming these challenges, improving extraction methods, and further exploring the molecular mechanisms underlying these bioactive compounds. This could enhance the reproducibility of results and the efficacy of plant-based therapeutics in clinical settings. Additionally, as the demand for natural remedies increases, it is imperative to study and conserve the biodiversity that serves as a source for these valuable medicinal plants, ensuring sustainable use and continuous availability (20).

CONCLUSION

The study of *Azadirachta indica*, *Swietenia mahagoni*, and *Melia azedarach* has highlighted their substantial potential in contributing to human healthcare through the discovery of natural bioactive compounds with diverse therapeutic properties. These findings not only validate traditional medicinal practices but also offer promising avenues for developing novel pharmaceuticals that are potentially safer and more effective than synthetic alternatives. Given their natural origin and the broad spectrum of their biological activities, these plants could play a crucial role in advancing drug development, particularly in the treatment of infectious and chronic diseases, thereby enhancing global health outcomes while also emphasizing the importance of preserving biodiversity for future medicinal discoveries.

REFERENCES

1. Parrotta JA. Healing plants of peninsular India 2001.
2. Carpinella C, Ferrayoli C, Valladares G, Defago M, Palacios S. Potent limonoid insect antifeedant from *Melia azedarach*. *Bioscience, biotechnology, and biochemistry*. 2002;66(8):1731-6.
3. Chen H-D, Yang S-P, Wu Y, Dong L, Yue J-M. *Terpenoids* from *Toona ciliata*. *Journal of natural products*. 2009;72(4):685-9.
4. Chowdhury R, Hasan CM, Rashid MA. Antimicrobial activity of *Toona ciliata* and *Amoora rohituka*. *Fitoterapia*. 2003;74(1):155-8.
5. Chowdhury R, Rashid R, Sohrab M, Hasan C. 12 α -hydroxystigmast-4-en-3-one: a new bioactive steroid from *Toona ciliata* (*Meliaceae*). *Die Pharmazie-An International Journal of Pharmaceutical Sciences*. 2003;58(4):272-3.
6. Chu XY, Yang K, He X, Yu KT, Luan YY, He QB, Li ZL, Xiang YL, Chen H, Zeng Y, Li YZ. Cross-linking N-succinyl chitosan-oxidated hyaluronic acid-based hydrogel loaded with bone marrow mesenchymal stem cell-derived exosomes induce bone regeneration in cranial defects. *Materials & Design*. 2024 Apr 23:112969.
7. Da Silva M, Agostinho SM, De Paula J, Neto JO, Castro-Gamboa I, Rodrigues F, et al. Chemistry of *Toona ciliata* and *Cedrela odorata* graft (*Meliaceae*): chemosystematic and ecological significance. *Pure Appl Chem*. 1999;71:1083-7.
8. David S. Anti-pyretic of neem oil and its constituents. *Mediscope*. 1969;12:25-7.
9. Del Serrone P, Nicoletti M. Antimicrobial activity of a neem cake extract in a broth model meat system. *International journal of environmental research and public health*. 2013;10(8):3282-95.
10. Bhandari A, Shetty K, Wadhwa A, Yadav KS. Biopolymers, Composites, Nanocomposites, and Gels in Biotechnology. *Applications of Biopolymers in Science, Biotechnology, and Engineering*. 2024 Jan 20:139-65.
11. Govindachari T. Chemical and biological investigations on *Azadirachta indica* (the neem tree). *Current science*. 1992;63(3):117-22.
12. Otake T, Mori H, Morimoto M, Ueba N, Sutardjo S, Kusumoto IT, et al. Screening of Indonesian plant extracts for anti-human immunodeficiency virus—type 1 (HIV-1) activity. *Phytotherapy Research*. 1995;9(1):6-10.
13. Patel D, Prasad SK, Kumar R, Hemalatha S. An overview on antidiabetic medicinal plants having insulin mimetic property. *Asian Pacific journal of tropical biomedicine*. 2012;2(4):320-30.
14. Di Ianni A, Di Ianni A, Cowan K, Barbero LM, Sirtori FR. Leveraging Cross-Linking Mass Spectrometry for Modeling Antibody–Antigen Complexes. *Journal of Proteome Research*. 2024 Feb 19.
15. Upadhyay U, Vigyan PC. Neem (*Azadirachta indica*) and its Potential for Safeguarding. *Journal of Biological Sciences*. 2014;14(2):110-23.
16. Wachsman M, Martino V, Gutkind G, Coussio J, Coto C, Torres Rd. Antiviral activity of a *Melia azedarach* plant extract. 1982.
17. Wang J-R, Liu H-L, Kurtán T, Mándi A, Antus S, Li J, et al. *Protolimonoids* and *norlimonoids* from the stem bark of *Toona ciliata* var. *pubescens*. *Organic & biomolecular chemistry*. 2011;9(22):7685-96.

18. Yerima M, Jodi S, Oyinbo K, Maishanu H, Farouq A, Junaidu A, et al. Effect of neem extracts (*Azadirachta indica*) on bacteria isolated from adult mouth. *Nigerian Journal of Basic and Applied Sciences*. 2012;20(1):64-7.
19. Feng Y, Li X, Liu C, Wang S, Zhao S, Yang J, He W, Guo K. Controllable Synthesis of Furan-Based Poly (ester amide) s and Its Study on Adhesive and Aggregation-Induced Emission Properties. *ACS Applied Polymer Materials*. 2024 Apr 19.
20. Zhang K, Lei C, Tan Z. Preliminary report of *Azadirachta indica* A Juss introduction and cultivation in Panzhihua. *Sichuan Nongye Daxue Xuebao*. 2007;25(3):282.