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Integrating Neem's Medicinal Properties into Microbiology Education: A Systematic Review

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Abstract: *This systematic review investigates the integration of Azadirachta indica (Neem) as potential learning resources into Microbiology topics in Biology learning, focusing on its bioactive compounds and medicinal properties such as antimicrobial agent. Neem's bioactive compounds, such as azadirachtin, nimbolide, and gedunin, exhibit potent antimicrobial activity against a broad spectrum of pathogens, including E. coli, S. aureus, and Candida albicans. These properties make Neem a compelling natural alternative in the fight against antimicrobial resistance (AMR), a growing global health concern. Drawing on 20 studies published between 2014 and 2024, this review highlights Neem's values in microbiology curricula by offering students hands-on opportunities to study natural antimicrobial agents while fostering critical thinking and practical skills in microbiology. Additionally, incorporating Neem into education promotes environmental sustainability whilst acts as an appreciation for traditional local wisdom.*

Keywords: *Antimicrobial, Education, Medicinal plants, Microbiology, Neem*

1. INTRODUCTION

Incorporating medicinal plant studies into microbiology education is a compelling approach to bridging traditional pharmacological knowledge with modern scientific methods. Among the most researched medicinal plants, Azadirachta indica, commonly known as Neem, holds a prominent position in traditional medicine across Asia and Africa due to its broad-spectrum medicinal properties (Vimala *et al*., 2014). Neem has long been valued for its antimicrobial, antiviral, antifungal, and anti-inflammatory effects, making it a natural alternative to synthetic treatments (Herrera-Calderon *et al*., 2019; Wylie and Merrell, 2022). Key bioactive compounds in Neem, such as azadirachtin, nimbin, nimbolide, and quercetin, exhibit unique mechanisms of action that target microbial cell structures, DNA synthesis, and metabolic pathways (Alzohairy, 2016). Neem also has a potential role in cosmetics. For instance, most studies explained the antimicrobial properties in health conditions, such as acne, dandruff and personal health care. (Baby *et al*., 2022) summarized not only the most common cosmetic claims to treat acne but also mitigating other skin disorders related to inflammatory and oxidant processes. These characteristics position Neem as an ideal subject for Microbiology studies, providing students with hands-on opportunities to explore natural antimicrobial agents and understand their potential within the context of phytomedicine.

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The study of Neem's medicinal properties has become especially pertinent in light of the global rise in antimicrobial resistance (AMR). According to the World Health Organization (WHO), AMR is one of the top ten public health threats worldwide, with resistant strains of bacteria, fungi, and viruses rendering conventional treatments increasingly ineffective (World Health Organization (WHO), no date). AMR has spurred interest in exploring alternative treatments, particularly those derived from plants with historical medicinal use, such as Neem. Studies show that Neem's active compounds, particularly azadirachtin, can inhibit the growth of multiple bacterial strains, including Staphylococcus aureus, Escherichia coli, and Pseudomonas aeruginosa (Krishnan, Y & Kui.W., 2015), (Mohapatra, Ranjan and Sandra Horta Monica, 2014) which are responsible for a significant number of hospital-acquired infections (Wylie and Merrell, 2022; Romão *et al.*, 2023). According to a study by Muhammad (Mohammed, Fadhil and Omer, 2015) leaf extract of A. indica (Neem) has a potent antibacterial activity against various strains of bacterial pathogens. Results from (Faujdar, Bisht and Sharma, 2020) show that ethanolic extract of Neem leaves showed good antibacterial activity against all uropathogenic isolates. Maximum zone of inhibitions and lowest minimum inhibitory concentration and minimum bactericidal concentration values were revealed by Acinetobacter baumannii. Saveral other studies conducted by Maleki (Maleki et al., 2017) and (Altayb *et al.*, 2022), demonstrated that methanolic extract of Neem inhibits the growth of various bacterial strains including nosocomial strain of E. faecalis. Another study by Rawat (Rawat et al., 2017) demonstrated that Neem oil exhibits significant antifungal activity against a range of fungi, including Schizophyllum commune and Fusarium oxysporum, with effective concentrations starting above 2%. This highlights the potential of Neem as a natural alternative to synthetic antifungal agents. Supplementation of Neem's bark and leaves also performed effective activity against pathogenic bacteria EC0157 in a dairy manure in less than 10 days (Ravva and Korn, 2015). The same supplementation was used by (Jamge *et al.*, 2014; Akhter and Sarker, 2019) and it showed more inhibition zone against Vibrio cholerae and Bacillus subtilis, E. coli and Salmonella spp respectively.

Beyond its antimicrobial potential, Neem also represents a sustainable and accessible resource that aligns with educational initiatives to promote environmental awareness and cultural respect. Dubbed the "village pharmacy," Neem is an integral part of healthcare in various cultures, where it is used not only for treating infections but also for wound healing, digestive health, and immunological benefits (Khan, 2017; Blum, Singh and Merrell, 2019). Integrating Neem in the learning of microbiology for students could cultivate an appreciation for ethnobotany and sustainable health practices, allowing students to evaluate traditional practices within rigorous scientific frameworks. Students can examine these natural compounds' antimicrobial effects through experimentation and case studies. Moreover, by investigating the antimicrobial properties of Neem, students can develop a comprehensive understanding of alternative antimicrobial agents while gaining insight into the intersection of traditional knowledge and modern science (Wylie and Merrell, 2022). Incorporating Neem in microbiology as a learning resource is highly relevant for students, especially those preparing to address critical health challenges like antimicrobial resistance (AMR). By studying Neem's antimicrobial properties, students can develop key skills, such as understanding microbial mechanisms, analyzing bioactive compounds, and exploring natural alternatives to synthetic drugs. These competencies are essential for future microbiologists working in areas like clinical diagnostics, pharmaceutical research, or public health. Additionally, this approach not only equips students with practical scientific skills but also promotes a sense of appreciation toward traditional medicinal knowledge and its role in sustainable healthcare solutions. Integration of such topics into microbiology education helps bridge the gap between scientific innovation and real-world applications, help preparing students to tackle pressing global issues effectively. Those potencies highlight the potential of Neem as learning resources in Microbiology subject.

The primary objective of this review is to compile and synthesize existing research on Neem's medicinal properties, with a specific

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focus on its antimicrobial application. This review discusses Neem's bioactive compounds and their effects on microbial agents, providing a robust foundation for incorporating Neem into Microbiology curricula. Subsequently, the strategies to integrate Neem potencies in Microbiology learning are also discussed by analysing and synthesizing relevant literature on similar topics but with different medicinal plants. Studies have demonstrated the effectiveness of integrating medicinal plants into biology education. For instance, Prayitno and Suryadarma (Prayitno and Suryadarma, 2019) developed and validated LKPDs focusing on medicinal plant diversity using a group investigation model, which significantly improved student comprehension and engagement. Similarly, the ethnobotanical study of the Baduy tribe led to the development of a contextual-based module, allowing students to explore plant diversity and medicinal uses through inquiry-based learning, thereby enhancing scientific understanding and cultural awareness (Khastini *et al*., 2019). Another study on Dayak medicinal plants in Kayu Ara Village identified 21 species used for ailments like kidney pain and malaria, resulting in a biodiversity-focused learning module that fostered scientific and cultural appreciation (Sunarya, Rahayu and Sunandar, 2024). In Pasti Jaya Village, a validated booklet integrating traditional medicinal plants for postpartum care into reproductive health topics was developed, enhancing students' understanding of human biology while promoting traditional knowledge (Hartika, Panjaitan and Tenriawaru, 2024). Additionally, Pit'ay et al (Fonny Pit, Ulung Anggraito and Ngabekti, 2019) identified 33 medicinal plant species in Nasinoah Forest and created teaching materials validated as highly effective for enhancing learning, conserving biodiversity, and preserving local wisdom.

While the antimicrobial properties of Neem have been extensively studied, its potential integration into microbiology education remains largely unexplored. Most studies are focused on its medicinal applications, yet no study analyze how it can be used as a learning resource to enhance microbiology education. Previous research on medicinal plants in education, such as those involving ethnobotanical studies and learning modules, has demonstrated improvements in student engagement and scientific understanding. However, no systematic review has specifically addressed Neem's role as a microbiology learning tool. This review aims to bridge that gap by synthesizing studies on Neem's bioactive properties and proposing strategies to incorporate them into microbiology curricula. By contextualizing its antimicrobial effects within an educational framework, this review provides insights into how traditional pharmacological knowledge can be integrated into microbiology learning. This interdisciplinary approach not only enhances microbiology education but also fosters awareness of antimicrobial resistance (AMR) and sustainable health practices.

2. RESEARCH METHOD

This systematic review followed rigorous PRISMA guidelines to ensure accuracy and replicability in synthesizing relevant studies on Neem's (Azadirachta indica) medicinal properties and its applications for Microbiology learning. The review included peer-reviewed articles published within the last ten years (2014–2024) that are accessible and available in full text. The studies should discuss Neem's medicinal properties, particularly antimicrobial applications in microbiology. Studies exclusively focused on the chemical properties of Neem without microbiological relevance were excluded from this review. The literature search was conducted in three primary databases: PubMed, Scopus, and Google Scholar, chosen for their comprehensive coverage of life sciences, biomedical studies, and interdisciplinary research. These sources ensured that high-quality, peer-reviewed studies relevant to Neem's medicinal properties were included. A combination of keywords and Boolean operators was used, such as "Neem," "Azadirachta indica," "medicinal properties," "microbiology education," and "antimicrobial effects," following established methods for systematic search refinement (Buckley *et al*., 2013). Truncations (e.g., "microbiolog*") were applied to capture a broad scope of relevant studies. To further discuss the strategies to incorporate Neem into Microbiology learning, similar searching strategies were also employed to identify and screen relevant articles published during the same duration of year in the same databases, using citation tracking and snowballing techniques.

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Once retrieved, all references were managed using Mendeley software, facilitating duplicate removal and structured organization. The data collection followed a three-phase process (Mateen *et al*., 2013): First, titles and abstracts were screened based on the inclusion and exclusion criteria. Second, articles passing initial screening underwent a full-text review to confirm relevance to the review objectives. Third, data were extracted systematically from each selected study, capturing essential information on study design, sample size, interventions, and outcomes related to Neem's integration in microbiology education.

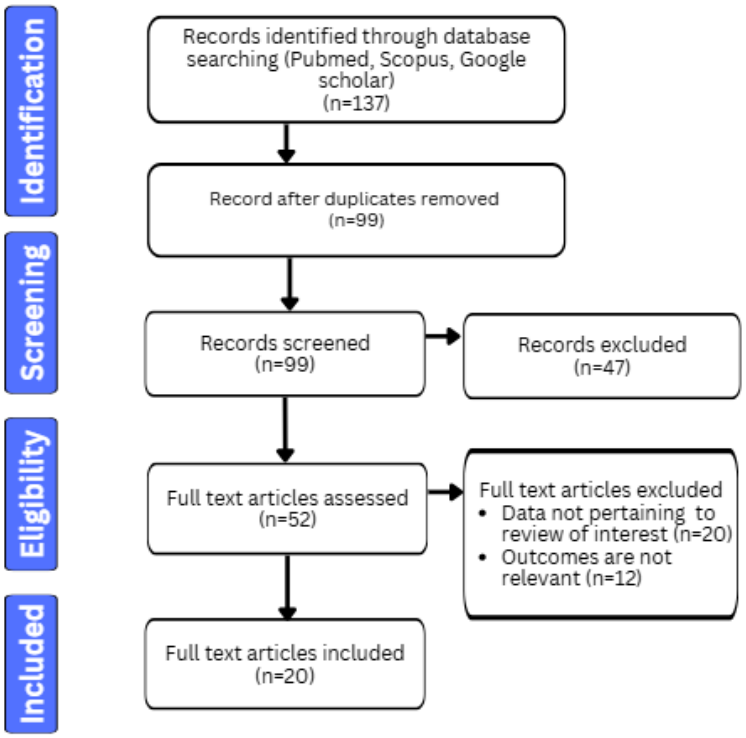

Figure 1. Flow Diagram of Article Selection

The data were analysed and synthesized from the findings both descriptively and thematically (Castleberry and Nolen, 2018), with quantitative results summarized in tabular form and qualitative insights grouped by recurring themes. Studies highlightting *Neem*'s efficacy in microbiological applications were further analyzed to understand pedagogical trends, effectiveness, and implications for future research. This process ensures a comprehensive and reliable review of current literature, providing insights into how *Neem*'s medicinal properties can be integrated into microbiology curricula.

3. RESULTS AND DISCUSSION

This systematic review synthesized research findings on *Neem* (*Azadirachta indica*), focusing on its antimicrobial properties and potential applications in Microbiology learning. The review found consistent evidence that *Neem* exhibits robust antimicrobial and antifungal properties, attributed to its bioactive compounds like *azadirachtin*, *nimbolide*, and *gedunin*, which are effective against pathogens commonly encountered in microbiology, including *Escherichia coli*, *Staphylococcus aureus*, and *Candida albicans* (Quelemes *et al*., 2015; Bansal *et al*., 2019; Wylie and Merrell, 2022). These properties make *Neem* an excellent candidate for educational modules in microbiology, providing students with hands-on experience in natural antimicrobial studies. Study by (Muhammad *et al*., 2019; Ali *et al*., 2021) demonstrated *Neem*'s bactericidal activity in both aqueous and ethanol extracts, suggesting that its application in lab exercises could vividly illustrate plant-based antimicrobial efficacy, thereby reinforcing theoretical concepts.

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A total of 20 articles were analyzed to explore the use of *Neem* (Azadirachta indica), with a particular focus on its antimicrobial properties. Among these, four articles were comprehensive reviews that synthesized existing knowledge on the antimicrobial efficacy and broader applications of *Neem* extracts. The remaining articles presented original research findings, primarily emphasizing the antimicrobial activity of *Neem* extracts against a range of bacterial and fungal pathogens. These studies employed diverse methodologies, such as zone of inhibition, minimum inhibitory concentration (MIC), and minimum bactericidal concentration (MBC), to evaluate the efficacy of *Neem* extracts. The inclusion of both review and research articles provided a balanced perspective, highlighting the consistency of antimicrobial evidence. Together, these studies underscore the potential of *Neem* as a valuable natural resource in antimicrobial applications and an engaging topic for educational settings.

Table 1. Analysis of review articles focusing on antimicrobial efficacy of Neem

Author and Reference	Article title	Objectives and Design	Study Results
(Alzohairy, 2016)	Therapeutics Role of *Azadirachta indica* (*Neem*) and Their Active Constituents in Diseases Prevention and Treatment.	To review the therapeutic potential of Azadirachta indica (*Neem*) in disease prevention and treatment through its pharmacological effects and active compounds. **Design:** The article is a review of existing studies highlighting *Neem*'s therapeutic applications and its active constituents in disease management, focusing on their biochemical mechanisms and pharmacological activities.	*Neem* exhibits diverse therapeutic activities, including antimicrobial, antioxidant, anticancer, anti-inflammatory, hepatoprotective, antidiabetic, and wound-healing effects. Active compounds such as azadirachtin, nimbolide, and nimbin regulate molecular pathways, including apoptosis, angiogenesis, and transcription factors, to inhibit disease progression. *Neem*'s components demonstrate significant potential in addressing microbial resistance and enhancing overall health management with minimal side effects.
(Wylie and Merrell, 2022)	The Antimicrobial Potential of the *Neem* Tree (*Azadirachta indica*)"	To explore the broad-spectrum antimicrobial properties of Azadirachta indica (*Neem*) against drug-resistant pathogens and biofilm-forming organisms and evaluate its potential as a source for novel therapeutics. **Design:** A comprehensive review synthesizing recent findings on *Neem*'s antimicrobial, antiviral, antifungal, and antiparasitic activities, focusing on its applications in medicine, dentistry, and food safety.	*Neem* demonstrated significant activity against antibiotic-resistant pathogens, biofilms, and various viruses, fungi, and parasites. It has applications in dentistry, food preservation, and medicine, with active compounds like azadirachtin and nimbolide showing therapeutic potential. Future research should focus on mechanisms of action, in vivo studies, and standardization of *Neem*-derived products for clinical applications.
(Baby *et al.*, 2022)	Azadirachta indica (*Neem*) as a Potential	To evaluate the potential of Azadirachta indica (*Neem*) for	The review highlighted A. indica's antimicrobial, anti-

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	Natural Active for Dermocosmetic and Topical Products: A Narrative Review	use in dermocosmetics by reviewing its biological activities, phytochemical properties, and recent technological advancements. **Design:** A narrative review summarizing existing literature, patents, and studies on *Neem*'s cosmeceutical applications and safety.	inflammatory, antioxidant, and wound-healing properties, making it suitable for applications in acne treatment, dandruff control, antiaging, and skin-whitening products. Advances in cell culture and nanotechnology were noted for enhancing targeted compound production. Toxicological evaluations supported its safety for dermocosmetic use, though further studies on phototoxicity and carcinogenicity are recommended.
(Herrera-Calderon *et al.*, 2019)	Azadirachta indica: Antibacterial activity of *Neem* against different strains of bacteria and their active constituents as preventive in various diseases	**Objectives:** To investigate the antibacterial activity of Azadirachta indica (*Neem*) against various bacterial strains and evaluate its active compounds for their role in preventing and treating different diseases. **Design:** A review compiling studies on the antimicrobial properties of *Neem* extracts, focusing on their effects on Gram-positive and Gram-negative bacteria, and the bioactive compounds responsible for these activities.	*Neem* exhibited significant antibacterial activity, particularly with methanolic extracts being highly effective. Its active compounds, such as nimbidin, azadirachtin, and nimbolide, displayed broad-spectrum antimicrobial effects. *Neem*'s extracts were also effective in combating antibiotic-resistant strains and showed potential in treating infections, promoting wound healing, and acting as a safe alternative to synthetic antibiotics.

Numerous studies highlight the remarkable antimicrobial properties of Azadirachta indica (Neem), showcasing its potential as a versatile therapeutic agent. (Wylie and Merrell, 2022) reviewed Neem's broad-spectrum efficacy against antibiotic-resistant pathogens, biofilm-forming organisms, and various viruses, fungi, and parasites, emphasizing its application in dentistry, food safety, and medicine. Similarly, Alzohairy (Alzohairy, 2016) detailed the pharmacological activities of Neem's active compounds, such as azadirachtin and nimbolide, which exhibit antimicrobial, anti-inflammatory, and antioxidant properties with minimal side effects. Herrera-Calderon (Herrera-Calderon *et al.*, 2019) further demonstrated Neem's effectiveness against multidrug-resistant bacterial strains, highlighting its role as a potential alternative to synthetic antibiotics. These findings, summarized in **Table 1**, underscore Neem's significance as a natural source for developing novel, sustainable therapeutics and emphasize the need for further investigation into its mechanisms of action and clinical applications.

The antimicrobial activity of *Azadirachta indica (Neem)* aqueous extracts against various microbial strains is illustrated in **Figure 2**, with zones of inhibition representing efficacy of *Neem* aqueous extract. The highest activity is observed against *Vibrio cholerae* and *Bacillus subtilis*, with inhibition zones of approximately 17–18 mm, while moderate activity is noted against *Escherichia coli, Staphylococcus aureus, Klebsiella pneumoniae*, and *Proteus mirabilis*, with zones ranging from 13–15 mm (Mohammed et al., 2015), (Jamge *et al.*, 2014). In contrast,

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lower activity is observed against *Streptococcus mutans* and *Candida albicans*, with inhibition zones of 10 mm and 6 mm, respectively (Bansal *et al.*, 2019). These findings highlight *Neem*'s selective antimicrobial potency, particularly against certain Gram-positive and Gram-negative bacteria, while demonstrating reduced efficacy against fungal strains. This underscores *Neem*'s potential as a natural antimicrobial agent and the necessity for further studies to optimize its application against less susceptible pathogens.

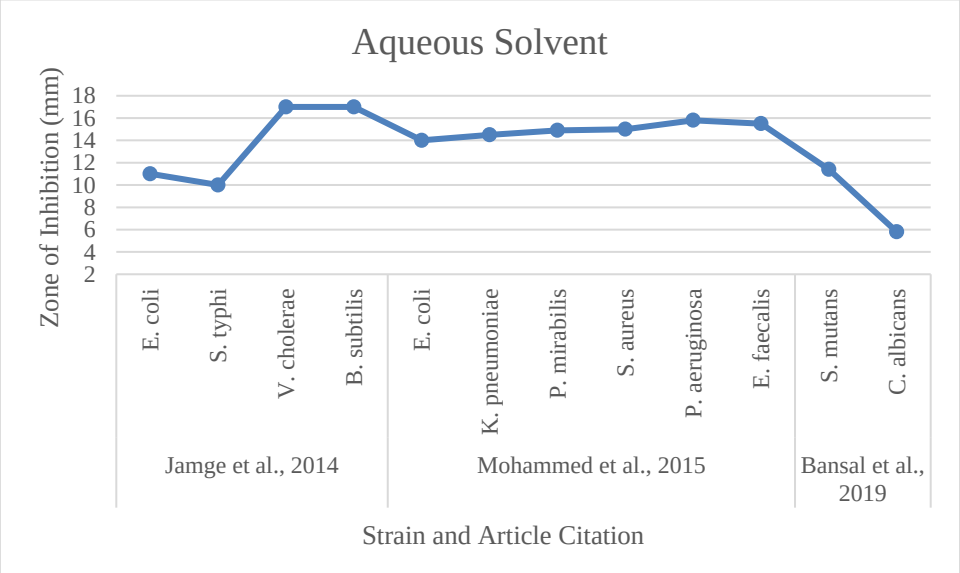

Figure 2. Antimicrobial activity of Neem extract in Aqueous solvent

Figure 3 illustrates the antimicrobial activity of Azadirachta *indica (Neem)* extracts in various organic solvents (ethanol, methanol, ethyl acetate, chloroform, and acetone) against different microbial strains, as measured by the zone of inhibition (in mm). *Neem* extracts prepared with ethanol generally exhibit the highest antimicrobial activity, with notable inhibition against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Escherichia coli*, achieving zones of inhibition up to 30 mm (Mohammed, Fadhil and Omer, 2015; Maleki *et al.*, 2017; Altayb *et al.*, 2022). Methanol and ethyl acetate extracts also demonstrate strong antimicrobial effects, particularly against *P. aeruginosa and S. aureus*, though their efficacy varies slightly (Krishnan, Y. & Kui.W., 2015; Maleki *et al.*, 2017).

Chloroform and acetone extracts show moderate antimicrobial activity, with inhibition zones ranging between 15–20 mm against strains like *Bacillus subtilis* and *Staphylococcus aureus* (Krishnan, Y. & Kui.W., 2015), (Mohapatra, Ranjan and Sandra Horta Monica, 2014). In contrast, some strains, such as *Enterobacter* and *Citrobacter species*, exhibit lower susceptibility to these solvents, with inhibition zones below 10 mm (Faujdar, Bisht and Sharma, 2020). These findings suggest that organic solvents, particularly ethanol and methanol, enhance the extraction of bioactive compounds from *Neem*, leading to higher antimicrobial activity against a broad spectrum of pathogens Further research is necessary to explore the mechanisms underlying solvent-specific differences and optimize their therapeutic applications.

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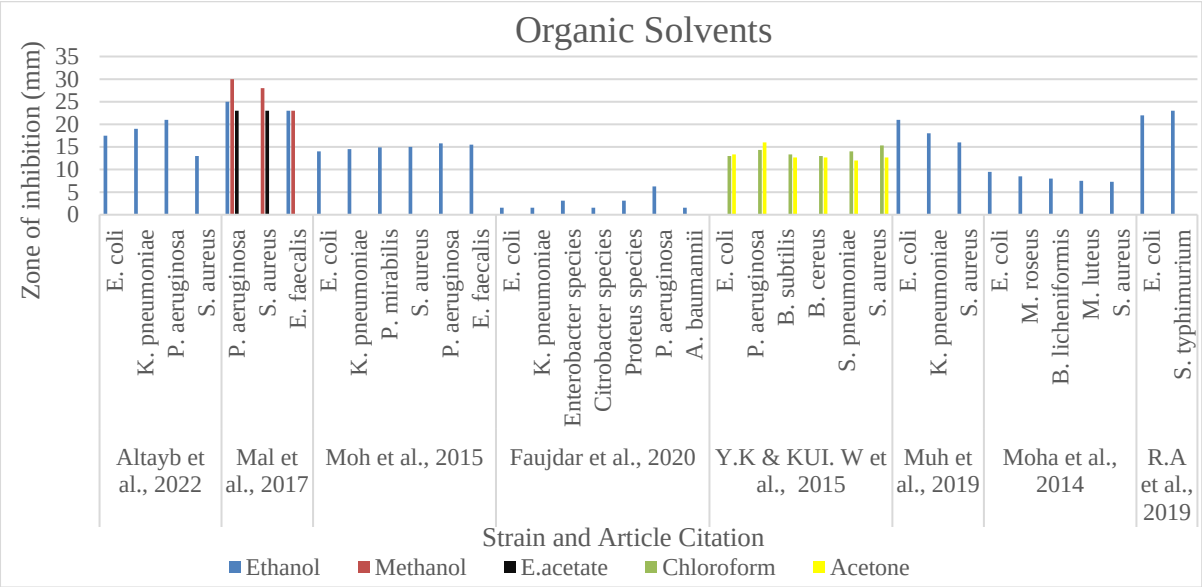

Figure 3. Antimicrobial activity of Neem extract in various organic solvents.

Figure 4 illustrates the Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) values of *Azadirachta indica (Neem)* extracts against various microbial strains, measured in mg/mL. MIC values, shown in blue, represent the lowest concentration of extract required to inhibit microbial growth, while MBC values, shown in red, indicate the lowest concentration required to kill the microorganisms. The MIC and MBC values vary significantly across the tested strains. For instance, *Escherichia coli*, *Staphylococcus aureus*, and *Klebsiella pneumoniae* demonstrate relatively high MIC and MBC values (ranging from 60 to over 100 mg/mL), indicating lower sensitivity to the extract (Muhammad *et al.*, 2019), (Ali *et al.*, 2021). In contrast, strains such as *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and certain *Proteus* and *Citrobacter species* exhibit lower MIC and MBC values (around 10–20 mg/mL), signifying greater sensitivity (Faujdar, Bisht and Sharma, 2020). The extract's antifungal activity is also notable, with *Candida albicans* showing MIC and MBC values of approximately 30 mg/mL (Bansal *et al.*, 2019). Similarly, *Streptococcus mutans* demonstrates moderate susceptibility, with MIC and MBC values below 50 mg/mL (Bansal *et al.*, 2019). The findings underscore the variability in the antimicrobial potency of *Neem* extracts, depending on the microbial strain, and highlight its potential application as a broad-spectrum antimicrobial agent. This data also emphasizes the necessity for further optimization of extraction methods to enhance *Neem's* effectiveness against resistant strains.

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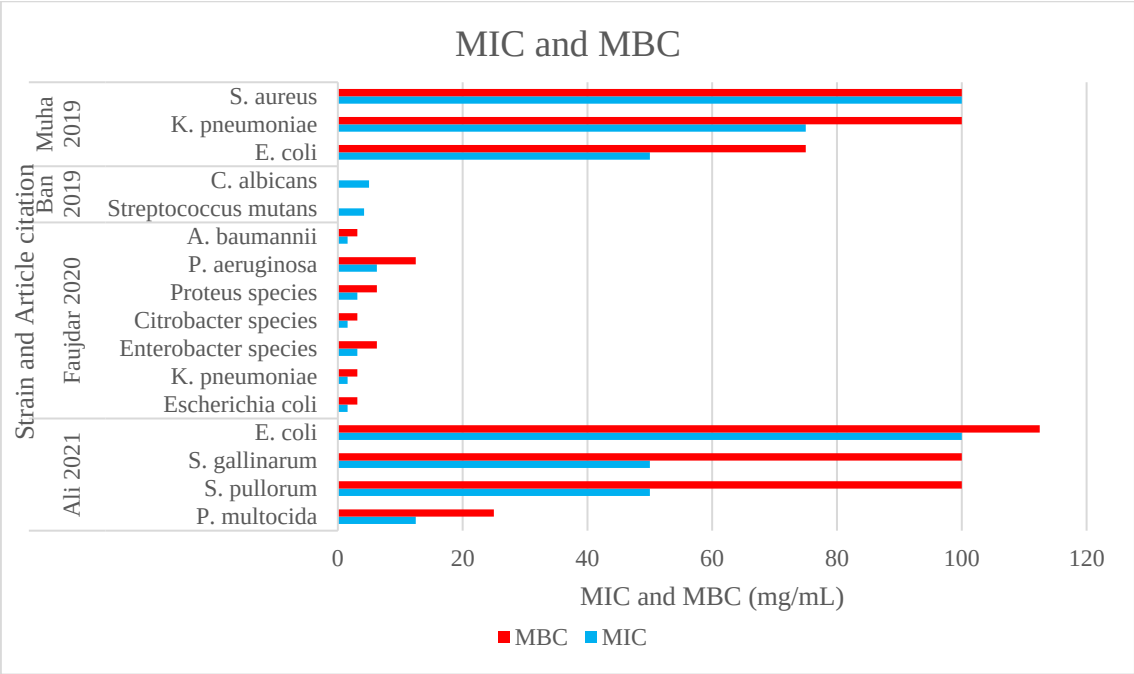

Figure 1. Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) of Neem extract, the values are measured in mg/mL.

The antimicrobial efficacy of *Neem* varies significantly depending on the extraction method, solvent type, and microbial strain. Studies have shown that ethanol and methanol extracts demonstrate stronger antimicrobial activity compared to aqueous extracts, likely due to the higher solubility of bioactive compounds like azadirachtin and nimbolide in organic solvents (Alzohairy, 2016; Herrera-Calderon *et al.*, 2019). This variation underscores the importance of selecting appropriate extraction technique for optimal efficacy in microbiology experiments.

Additionally, the mode of antimicrobial action in *Neem* extract is multifaceted. Compounds such as gredunin and nimbin disrupt bacterial cell membranes, while azadirachtin interferes with microbial metabolic pathways (Wylie and Merrell, 2022). This suggests that *Neem* could serve an effective natural antimicrobial alternative, particularly in regions with limited access to commercial antibiotics. Comparative studies between *Neem* and conventional antibiotics reveal promising results, with *Neem* extracts exhibiting inhibitory effects against *E. Coli, S. aureus, and P. aeruginosa*, comparable to certain synthetic antimicrobial agents (Mohapatra, Ranjan and Sandra Horta Monica, 2014; Ali *et al.*, 2021). Such findings support its potential applications in microbiology education, where students can explore alternative antimicrobial mechanisms beyond conventional drug-based approaches.

Table 2. Results from studies which integrated the knowledge of medicinal plants into biology learning

Title and citation	Medicinal Plants used	Educational context	Methodology	Findings
Developing LKPD Medicinal Plant Diversity with an Investigation Model Group on Learning comprehension	Various medicinal plants	High school biology education (Grade X, SMA Negeri 1 Cangkringan)	R&D approach Using a modified 4D model (Define, Design, Develop).	The LKPD increased comprehension in the experimental group compared to the control group, with post-test significance of 0.007 (<0.05). Improvements

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Title and citation	Medicinal Plants used	Educational context	Methodology	Findings
(Prayitno and Suryadarma, 2019)				include better understanding, interest, and critical thinking.
Ethnobotanical Study of Medicinal Plants Utilized by the Baduy Tribe used as a learning resource (Khastini *et al.*, 2019)	67 Species primarily Spermatophytes	High school biology education in Banten Province, Indonesia	R&D approach using the Thiagarajan model (Define, Design, Develop).	Developed a contextual-based module. Students gave positive feedback. Particularly appreciating the presentation aspect with an 89.5% satisfaction rating.
Ethnobotany of Dayak Medicinal Plants in Kayu Ara Village, Landak Regency as a Learning Resource (Sunarya, Rahayu and Sunandar, 2024)	21 species from 16 families	High school biology education, particularly in biodiversity lessons	Qualitative research using interviews and observations. Snowball sampling for plants identification, and teacher questionnaire using Guttman scale was used to assess their potential as learning resources.	Ethnobotany of Dayak medicinal plants was highly effective as a biology learning resource, achieving an average potential score of 92.86%. The study enhanced understanding of biodiversity and local wisdom while linking learning to real-life applications.
Human Pregnancy and Birth: The Enrichment of a Booklet with Medicinal Plants for Postpartum Wellness (Hartika, Panjaitan and Tenriawaru, 2024)	15 species of postnatal medicinal plants from Pasti Jaya Village	High school biology education focusing on reproductive system topics, including fertilization, pregnancy, and childbirth	R&D approach based on the Borg and Gall model.	Developed a validated booklet (CVI and CVR = 1.00) suitable for teaching the reproductive system.
Identifying Medicinal Plants in Local Custom Nasinoah Forest to Develop Local Wisdom-Based Learning Material (Fonny Pit, Ulung Anggraito and Ngabekti, 2019)	33 species from 25 families	High school biology education in Timor Tengah Selatan Regency (biodiversity-focused contextual learning)	Qualitative research using interviews, surveys, and literature review. Data were used to design learning instruments, including lesson plans, syllabus, and worksheets validated by three experts.	Medicinal plants were documented and integrated into contextualized learning materials. Validated learning materials were highly effective for understanding biodiversity and preserving local wisdom, with teacher and student responses rated as highly reliable (86.72%).

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As summarized in **Table 2**, several studies have integrated medicinal plants into biology education, effectively enhancing student understanding of biodiversity, ecology, and local wisdom. These studies utilized diverse approaches, including the development of contextual learning materials, booklets, and worksheets, which were validated and shown to improve comprehension, engagement, and appreciation of traditional ecological knowledge. Integrating medicinal properties of *Neem* into microbiology education can employe the development of LKPD (Sudent worksheet), which will be guided by successful approaches detailed in Table 2 above. Drawing on the modified 4D model applied in developing LKPDs for medicinal plant diversity at SMA Negeri 1 Cangkringan (Prayitno & Suryadarma, 2019), the *Neem*-based worksheet will incorporate theoretical and practical components to ensure comprehensive learning. The theoretical content will explore *Neem*'s bioactive compounds, such as azadirachtin and nimbin, and their antimicrobial mechanisms, linking these to microbiological principles. Practical activities, inspired by contextualized learning materials validated in studies from Banten Province (Khastini *et al.*, 2019) and Timor Tengah Selatan Regency (Fonny Pit *et al*., 2019), will include preparing *Neem* extracts and conducting microbial inhibition assays, such as agar diffusion tests. Ethnobotanical perspectives, as effectively integrated in studies of the Dayak (Sunarya et al., 2024) and Baduy tribes (Khastini *et al.*, 2019), will contextualize *Neem*'s traditional applications, enriching students' understanding of biodiversity and cultural heritage. To promote critical thinking, the LKPD will feature exercises for analyzing experimental results and discussing *Neem*'s potential in addressing antibiotic resistance and environmental sustainability. Following the methodologies in the cited studies, the LKPD will undergo rigorous validation through expert reviews and classroom trials, ensuring its efficacy in enhancing student comprehension, engagement, and awareness of microbiology.

Integrating *Neem* into microbiology curricula provides a unique opportunity to bridge ethnopharmacology and microbiological studies, promoting both scientific and cultural literacy. Existing studies on medicinal plant-based learning, such as those by (Fonny Pit *et al*., 2019; Prayitno & Suryadarma, 2019), highlight the effectiveness of incorporating indigenous knowledge into formal science education. By adopting a similar approach, microbiology educators can develop contextual learning activities where students analyze *Neem's* antimicrobial properties through experimental assays, such as agar diffusion test and minimum inhibitory concentration (MIC) measurements.

Furthermore, Neem's potential role in tackling antimicrobial resistance (AMR) offers a relevant theme for inquiry-based learning. Students can compare the effectiveness of *Neem* extracts against antibiotic-resistant strains, fostering a deeper understanding of natural antimicrobials in combating bacterial resistance. This aligns with modern educational trends emphasizing hands-on research experience and sustainability-focused learning (Straus & Chudler, 2016). Integrating *Neem* into microbiology education can therefore enhance critical thinking skills while reinforcing the importance of naturl product research in public health.

Incorporating *Neem*'s antimicrobial properties into microbiology curricula offers significant educational value, particularly by enhancing student engagement with alternative antimicrobials and providing hands-on lab experience. Practical activities with *Neem* allow students to develop skills in sample preparation and microbial inhibition analysis, bridging theory with practice. Additionally, comparing *Neem*'s efficacy with synthetic antimicrobials fosters critical thinking, as students explore antimicrobial resistance and phytomedicine's potential in public health. *Neem* also promotes sustainability awareness by encouraging students to appreciate traditional medicinal knowledge and ecological impacts. However, while *Neem*'s applications are promising, further research is needed to assess its long-term educational impact, optimize integration methods, and compare its effectiveness to synthetic agents in various settings (Straus & Chudler, 2016). While *Neem*'s potencies are promising to be used in education, particularly for Microbiology learning, current research is limited by small sample sizes, diverse methodologies, and regional availability, which

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may affect its generalizability and relevance across different educational contexts. Additionally, a potential selection bias toward studies published in English or reporting positive results may skew findings, highlighting the need for more inclusive and in depth research. To strengthen the evidence for *Neem*'s educational value, future research should include randomized controlled trials to compare *Neem* with synthetic antimicrobials, longitudinal studies to assess lasting impacts on knowledge retention and attitudes, and comparative efficacy studies that examine both educational outcomes and microbiological effectiveness.

4. CONCLUSION

This systematic review highlights the significant antimicrobial properties of Azadirachta indica (*Neem*) and its potential as a valuable educational resource in microbiology learning. The bioactive compounds of *Neem*, such as azadirachtin, nimbolide, and gedunin, have demonstrated efficacy against a broad spectrum of pathogens, including antibiotic-resistant strains, making it a promising natural alternative to synthetic antimicrobials. Integrating *Neem* into microbiology curricula offers a dual benefit: fostering hands-on student engagement with natural antimicrobial studies and promoting awareness of sustainable health practices. By bridging traditional pharmacological knowledge with modern scientific education, *Neem*-based learning worksheet can enhance student comprehension of microbiological concepts, critical thinking, and experimental skills. Furthermore, the incorporation of ethnobotanical perspectives supports environmental sustainability and cultural appreciation, aligning with global education goals. Despite these promising findings, variability in study methodologies and limited sample sizes underscore the need for further research to standardize experimental protocols and evaluate long-term educational impacts. Future studies should explore the comparative efficacy of *Neem* against synthetic antimicrobials in various educational and clinical contexts, alongside randomized trials to validate its broader applications.

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