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Phytochemical composition and antibacterial activities of *Azadirachta indica* (Neem): significance of traditional medicine in combating infectious diseases and antimicrobial resistance

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Abstract
Traditional medicines such as *Azadirachta indica* (*A. indica*) possess medicinal properties that should be explored in drug discovery to combat infectious diseases and antimicrobial resistance. This study provided insights into the antibacterial activities of *A. indica*. This narrative review utilised Google Scholar and PubMed to search for articles that were published from January 2000 to September 2023 on the antibacterial activities of *A. indica*. The study found that *A. indica* has activity against *Escherichia coli*, *Staphylococcus aureus*, *Enterococcus faecalis*, and other bacteria. The antibacterial activities of *A. indica* were reported to be due to its phytochemical content such as phenolics, tannins, saponins, flavonoids, terpenoids, fatty acids, and alkaloids. This review found that *A. indica* has antimicrobial activity against some Gram-negative and Gram-positive bacteria due to the presence of phytochemicals. There is a need to increase investment in drug discovery and the integration of traditional medicine into the healthcare system.

Keywords: Antibacterial activity, antimicrobial resistance, *azadirachta indica*, drug discovery, neem, traditional medicine

Introduction
Traditional medicines are essential in the treatment of various diseases [1–7]. There has been a rise in the burden of infectious diseases across the globe, including HIV, malaria, diarrhoea, tuberculosis, meningitis, respiratory tract infections, urinary tract infections, and soft tissue infections [8, 9]. This burden of infectious diseases has led to the overuse and misuse of antimicrobials [10–14]. Consequently, this has resulted in the emergence of antimicrobial resistance (AMR) [15–17]. AMR occurs when microorganisms stop responding to antimicrobials to which they were previously sensitive to [18]. AMR has fatal consequences as reported by many scholars [17–24], hence, the need to address it using various strategies [25–36], including the use of traditional medicine1 [7, 37].

The emergency of AMR demands a need to promote drug discovery and the development of newer antimicrobials and alternative therapies [2, 38–45]. This has led scientists to strengthen the isolation and discovery of novel bioactive compounds from plants to combat AMR [46–50]. Traditional medicines have the potential to cure many diseases and can be used as drug sources [1, 2, 45, 51–54]. The World Health Organization (WHO) has underscored the enduring significance of traditional medicines derived from medicinal plants in developing nations [55]. WHO states that a remarkable 80% of these developing countries continue to derive substantial benefits from the utilization of traditional medicines [55, 56]. Due to the evidence-based medicinal effects of traditional medicine, there have been many supports and recommendations to integrate it into the healthcare system, of which other countries have done so [55, 57–60].

Azadirachta indica (*A. indica*), also called the Neem tree, is a potential source of medicines and should be explored further in drug discovery [61–64]. This plant has been reported to have antimicrobial activity against many microorganisms [65–67]. *A. indica* has been found to have antibacterial activity against both Gram-positive and Gram-negative bacteria, including activity against *Escherichia coli* (*E. coli*) [68] and *Enterococcus* species [69]. Additionally, it has activity against drug-resistant infections, making it a potential antibiotic [70].

The active compounds (phytochemicals) present in *A. indica*, such as nimbin, nimbidin, azadirachtin, and gedunin, have been shown to exhibit remarkable bactericidal and bacteriostatic properties [71–74]. *A. indica* is also reported to possess many phytochemicals including phenolics, tannins, saponins, flavonoids, alkaloids [68], terpenoids and fatty acids [69]. Additionally, neem's antibacterial mechanism of action involves disrupting bacterial cell membranes, inhibiting crucial enzymes, and interfering with DNA replication, further enhancing its efficacy against microbial pathogens [75]. The antibacterial activities are reported at various minimum inhibitory concentration (MIC) and zones of inhibition.

There are various reports on the phytochemical contents and antimicrobial activities of *A. indica*. Therefore, this study reviewed articles on the phytochemical composition and antibacterial activities of *A. indica* against bacterial isolates. The findings of this review paper can be used to promote drug discovery from natural products.

Materials and methods

This was a systematic review that was conducted from March 2023 to September 2023. The literature search was done using Google Scholar and PubMed for all articles that were published on the antibacterial properties of *A. indica*. This study employed search words including '*Azadirachta indica*', '*A. indica'*, 'neem', 'antibacterial activity', 'antibacterial activities', 'traditional medicines', 'drug discovery', 'phytochemical (s)', and 'antimicrobial resistance'. This review was conducted in line with other narrative reviews conducted in traditional and herbal medicines [76–78]. This study included original articles that were written in English and published from January 2000 to September 2023. We excluded abstracts, grey literature, thesis, narrative reviews, systematic reviews, clinical trials, opinion papers, pre-prints, and publications on non-antibacterial medicinal uses of the plant.

From these studies that were included, the following data was extracted: author, year, origin of the plant material, part of the plant extracted, method of extraction, procedure, Phytochemicals present and findings.

Results

The phytochemical constituents and antibacterial activities of *A. indica* are shown in Table 1.

Table 1: Phytochemical composition and antibacterial activities of *Azadirachta indica*

Author names and year	Study Country	Findings on the tested pathogens	Tested part	Phytochemicals present
Hikaambo *et al*., 2022 [68]	Zambia	The extract had antibacterial activity against *E. coli* with a MIC of 10 mg/ml for the aqueous extract and 20 mg/ml for the ethanolic extract	Leaf extract	Phenolics, tannins, saponins, flavonoids, and alkaloids
Surhartono *et al*., 2023 [69]	Indonesia	The extract had antibacterial activity against *E. faecalis* at a concentration of 12.5%	Leaf extract	Terpenoids and fatty acids
Sahrawat *et al*., 2018 [79]	India	The extract had antibacterial activity against *E. coli* at a concentration of 58.77%	Leaf extract	Saponins, tannins, terpenoids, and alkaloids
Muhammad *et al*., 2019 [80]	Nigeria	The extract had antibacterial activity against *E. coli* and *K. pneumoniae* at a concentration of 50 mg/ml and 75 mg/ml respectively	Leaf extract	Phenols, tannins, and alkaloids
Momoh and Olaleye, 2022 [81]	Nigeria	The extract had minimum antibacterial activity against *E. coli* at a concentration of 100 mg/ml with a zone of inhibition of 9.33 mm, and highest activity at 500 mg/ml with a zone of inhibition of 21.36 mm	Root extract	Alkaloids, tannins, saponins, phenolics, and flavonoids
Singh, 2021 [82]	India	The extract had antibacterial activity against *E. coli* at a concentration of 1mg/ml with a zone of inhibition of 0.1 mm	Leaf extract	Flavonoids, resin, and phlobatannins
Ogidi *et al*., 2021 [83]	Nigeria	The extract had antibacterial activity against *Staphylococcus* spp, *Shigella* spp, *Vibrio* spp, and *E. coli* with zones of inhibition of 10mm and 7.0 mm, 7.0 mm and 26.0 mm, 7.0 mm and 10.6 mm, and 9.3 mm and 10 mm, respectively	Leaf extract	Saponins, tannins, steroids, glycosides, terpenoids, flavonoids, and alkaloids
Meressa, 2017 [84]	Ethiopia	The extract had antibacterial activity against *S. typhi* and *E. coli* with zones of inhibition of 20 mm and 16mm respectively	Stem bark extract	Tannins, saponins, phlobatannins, flavonoids, cardiac glycosides and alkaloids
Kulkarni, Pandey and Patil, 2017 [85]	India	The extract had antibacterial activity against *E. coli*, *S. aureus*, *K. pneumonia*, *E. aerogenes*, *P. aeruginosa*, *S. typhi*, *S. epidermidis* and *Proteus vulgaris*	Leaf extract	Alkaloids, glycosides, and tannins
Mohammed, Stephens and Sadiq, 2022 [86]	Nigeria	The extract had antibacterial activity against *E. coli* and *S. aureus* at a concentration of 200 mg/ml	Leaf extract	Glycosides, carbohydrates, phenols, alkaloids, and flavonoids
Fatima, 2020 [87]	Pakistan	The extract had antibacterial activity against *E. coli*, *S. aureus*, and *P. aeruginosa* with zones of inhibition of 25 mm, 26mm and 23mm respectively	Leaf extract	Tannins, glycosides, alkaloids, and saponins
Benisheikh, 2019 [88]	Nigeria	The extract had antibacterial activity against *S. aureus* with a zone of inhibition of 10 mm	Seed extract	Terpenoids
Mustafa, 2016 [89]	Saudi Arabia	The extract had antibacterial activity against *E. faecalis* a zone of inhibition of 17.70 mm	Leaf extract	Tannins
Faujdar, Bisht and Sharma, 2020 [90]	India	The extract had antibacterial activity against *E. coli*, *K. pneumoniae*, *Citrobacter* spp, *Proteus* spp, *Acinetobacter baumannii*, *P. aeruginosa*, and *Enterobacter* spp	Leaf extract	-
Maleki *et al*., 2017 [91]	Iran	The extract had antibacterial activity against *P. aeruginosa* at a concentration of 300 mg/ml. The extract also had activity against *S. aureus* and *E. faecalis*	Leaf extract	-

Melese *et al*., 2016 [92]	India	The extract had antibacterial activity against *S. aureus*, *P. aeruginosa*, and *E. coli* at the concentration of 400 mcg/ml	Seed and bark extracts	Alkaloids, carotenoids, saponins, anthraquinones, and cardiac glycosides
Sharma *et al*., 2014 [93]	India	The extract had antibacterial activities against *E. coli* and *Bacillus amyloliquefaciens* at the different MICs	Leaves, roots, seed and bark extracts	Tannin, saponins, alkaloids, phenolics, and flavonoids
Saleem *et al*., 2022 [94]	Pakistan	The extract had antibacterial activities against *S. aureus* and *E. coli* at the zone of inhibition of zone of 10 mm	Leaves extract	Saponins, tannins, reducing sugars, glycosides, alkaloids, and flavonoids
Dereje *et al*.,2021 [95]	Ethiopia	The extract had antibacterial activities against *Streptococcus pyogenes* at the concentration of 100 mg/ml	Leaves extract	Alkaloids, terpenoids, saponins, flavonoids, phenols, tannins, and steroids
Kumari *et al*., 2019 [96]	India	The extract had antibacterial activities against *Streptococcus mutans* with a zone of inhibition of 11.81 mm	Barks, leaves and twig pieces were the extracts	-
Mohammed, 2017 [97]	Sudan	The extract had antibacterial activities against *E. coli*, *P. aeruginosa*, *Proteus mirabilis*, *Klebsiella pneumoniae*, *S. aureus* and *E. faecalis* with the highest inhibition against *Proteus mirabilis* and *E. faecalis* at the concentration of 6.25 mg/ml and 12.5 mg/ml respectively	Leaf extract	Alkaloids, terpenoids, saponins, flavonoids, and phenols,
Altayb *et al*., 2022 [98]	Saudi Arabia	The extract had antibacterial activities against *S. aureus*, *P. aeruginosa*, *E. coli*, *Proteus spp.*, *S. epidermidis*, *Citrobacter* spp., *K. pneumoniae*, *E. faecalis*, *S. aureus* at the different zones of inhibition with the highest recorded with *P. aeruginosa* with 13 mm	Leaves extract	Fatty acids, hydrocarbons, pyridine derivatives, aldehydes, phenol group, aromatic substances, coumarins, monoterpenes and beta. d-Mannofuranoside O-geranyl
Ibrahim and Kebede, 2020 [99]	Ethiopia	The extract had antibacterial activities against *S. aureus*, *S. typhi*, *Streptococcus agalactiae*, and *Shigella boydii* with the methanol extract showing the highest inhibition zone at the concentrations of 125 mg/ml and 150 mg/ml	Leaves extract	-
Senthilkumar *et al*., 2018 [100]	India	The extract had antibacterial activities against *P. aeruginosa* at concentrations ranging from 2 to 16 µg/ml	Leaves extract	Silver nanoparticle
Singh *et al*., 2017 [101]	India	The extract had antibacterial activities against *S. aureus*, *Streptococcus pyogenes*, *E. coli*, *S. typhi*, and *K. pneumonia*	Leaves extract	-
Pandey, Verma and Singh, 2014 [102]	India	The extract had antibacterial activities against *E. coli* and *S. aureus* with the inhibition zone 13.23 ± 3.26 and 16.52 ± 3.28 at the concentration of 1.50 mg/ml respectively.	Leaves extract	Alkaloids, terpenoids, saponins, flavonoids, phenols, tannins, steroids, and aldehydes
Ali *et al*., 2021 [103]	Bangladesh	The extract had antibacterial activity against multidrug-resistant *E. coli* and *Salmonella* spp.	Leaf extract	-

Discussion

This study reviewed publications on the phytochemical composition and antibacterial activities of *A. indica* against various bacterial species. In reviewing the literature, our study found that the leaf, stem, root, and bark extract of *A. indica* has antibacterial activity due to the presence of phytochemicals including phenolics, tannins, saponins, flavonoids, alkaloids, terpenoids, fatty acids, glycosides, etc. Additionally, we found that *A. indica* has antibacterial activity against Gram-negative and Gram-positive bacteria including *E. coli*, *Enterococcus* species, *S. aureus*, *S. typhi*, *K. pneumoniae*, *Streptococcus pyogenes*, *Enterobacter* spp, and other bacteria.

This study revealed that the part of the plant that has antibacterial activities includes the leaf, stem, root, and bark extract, as reported by many publications [68, 79, 81, 83, 84, 86–88, 90, 91, 95, 96, 98, 102, 103]. The antibacterial activity of *A. indica* leaves is attributed to its bioactive compounds such as nimbin, nimbinin, and nimbidin, which act by disrupting the bacterial cell membrane and interfering with their metabolic processes [70]. The stem contains compounds like gedunin and mahmoodin, which have been identified as active antibacterial agents [104]. These compounds inhibit the growth and proliferation of bacteria by disrupting their cellular functions [105]. *A. indica* root extracts have been less studied compared to other parts of the plant, but evidence has shown its antibacterial efficacy [104]. Roots contain azadirachtin, a prominent bioactive compound responsible for many of *A. indica*'s medicinal properties, including its antibacterial activity. Azadirachtin has been found effective against bacterial strains like *Bacillus subtilis* and *K. pneumoniae* [106]. *A. indica* bark extract is another part of the plant that possesses antibacterial potential [104]. Some studies have shown that the bark extracts contain compounds such as quercetin and beta-sitosterol, which exhibit antibacterial activity against various pathogenic bacteria [107, 108]. It is important to note that while *A. indica* extracts have shown promising antibacterial properties *in vitro*, further research is required to explore their efficacy *in vivo* and their potential use in clinical settings. Furthermore, it's important to note that the levels of active compounds within these extracts can exhibit significant variations, contingent upon factors such as geographical location, prevailing climatic conditions, and the maturity of the plant from which they are derived. These variables can exert a profound impact on the collective antibacterial potential of the extracts, thereby necessitating a comprehensive understanding of the intricate interplay between environmental factors and the phytochemical composition of these natural remedies. Such insight is crucial for optimizing the antibacterial efficacy of these extracts and harnessing their therapeutic potential to combat microbial infections effectively.

The findings of this review provide compelling evidence for the antibacterial potential of *A. indica* against *E. coli*, *S.*

aureus, *Salmonella typhimurium*, *K. pneumoniae*, *P. aeruginosa*, *Enterococcus faecalis*, and *other bacteria*. The review, which encompassed a comprehensive analysis of literature from multiple studies, consistently demonstrated that *A. indica* exhibits significant antibacterial activity against this pathogenic bacterium. The consistent outcomes across these studies reinforce the validity and reliability of the observed antibacterial effects of *A. indica* against the various bacteria. However, the limited number of studies included in this review underscores the need for further research to strengthen the evidence base. The inclusion of more studies from diverse geographical locations and experimental setups would enhance the generalizability and robustness of the findings. Finally, the findings of this review demonstrated the importance of integrating traditional medicine into the modern healthcare system.

Conclusion

Azadirachta indica has antibacterial activities due to its rich composition of phytochemicals. These findings are critical and may be utilised in drug discovery. Therefore, there is a need for more investment in drug discovery, especially with the growing resistance of pathogens to conventional antimicrobials. There is a need to promote further research in traditional medicine that can inform policy and support the incorporation of traditional medicine in the healthcare system to be used together with conventional medicines.

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References

1. Keter LK, Mutiso PC. Ethnobotanical studies of medicinal plants used by Traditional Health Practitioners in the management of diabetes in Lower Eastern Province, Kenya. J Ethnopharmacol. 2012;139(1):74–80. DOI:10.1016/j.jep.2011.10.014
2. WHO. Antibacterial Agents in Clinical and Preclinical Development. World Health Organization 2021; c2021. Available: https://apps.who.int/iris/handle/10665/340694
3. Fialová SB, Rendeková K, Mučaji P, Nagy M, Slobodníková L. Antibacterial activity of medicinal plants and their constituents in the context of skin and wound infections, considering European legislation and folk medicine: A review. International Journal of Molecular Sciences. Multidisciplinary Digital Publishing Institute. 2021 Oct 4;22(19):10746. DOI:10.3390/ijms221910746
4. Kassa A, Tadele Y. Ficus sycomorus (Sycamore Fig or Shola) Leaf, A Potential Source of Protein for Ruminants: A Review. J Fish Livest Prod. 2015;3:152. DOI:10.4172/2332-2608.1000152
5. Hikaambo CN, Chisanga T, Kamapamba M, Akapelwa TM, Chimombe T, Chulu M, *et al*. Antibacterial Activity of Cassia abbreviata Oliv Bark Extract against *Escherichia coli* and Staphylococcus aureus. J Pharm Res Sci Technol. 2022;6:76–83. DOI:10.31531/jprst.1000161
6. Kabuka R, Mudenda S, Kampamba M, Chulu M, Chimombe T, Hikaambo CN. Phytochemical Analysis of Leaf, Stem Bark, and Root Extracts of Cassia abbreviata Grown in Zambia. Pharmacol & amp; Pharm. 2022;13(05):119-28. DOI:10.4236/pp.2022.135009
7. Hikaambo CN, Chilala P, Ndubi F, Mayoka G, Kampamba M, Kabuka R, *et al*. Antimicrobial Activities of Solanum aculeastrum Fruit Extract against *Escherichia coli*, Staphylococcus aureus and Candida albicans: Significance of African Traditional Medicine in Combating Infections and Attaining Universal Health Coverage. Pharmacol Pharm. 2023;14(5):176–188. DOI:10.4236/PP.2023.145013
8. Nyaruaba R, Okoye CO, Akan OD, Mwaliko C, Ebido CC, Ayoola A, *et al*. Socio-economic impacts of emerging infectious diseases in Africa. Infectious Diseases. Taylor & Francis; 2022 May 4;54(5):315-24. DOI:10.1080/23744235.2021.2022195
9. Houlihan CF, Whitworth JA. Outbreak science: recent progress in the detection and response to outbreaks of infectious diseases. Clin Med (Northfield Il). 2019;19(2):140. DOI:10.7861/CLINMEDICINE.19-2-140
10. Mancuso G, Midiri A, Gerace E, Biondo C. Bacterial antibiotic resistance: the most critical pathogens. Pathogens. MDPI; 2021;10(10):1310. DOI:10.3390/pathogens10101310
11. Paterson IK, Hoyle A, Ochoa G, Baker-Austin C, Taylor NGH. Optimising antibiotic usage to treat bacterial infections. Sci Rep. 2016;6(1):37853. DOI:10.1038/srep37853
12. Byrne MK, Miellet S, McGlinn A, Fish J, Meedya S, Reynolds N, *et al*. The drivers of antibiotic use and misuse: The development and investigation of a theory-driven community measure. BMC Public Health. 2019;19(1):1425. DOI:10.1186/s12889-019-7796-8
13. Manohar P, Loh B, Leptihn S. Will the Overuse of Antibiotics During the Coronavirus Pandemic Accelerate Antimicrobial Resistance of Bacteria? Infect Microbes Dis. 2020;2(3):87–88. DOI:10.1097/im9.0000000000000034
14. Brudzynski K, Abubaker K, St-Martin L, Castle A. Re-examining the role of hydrogen peroxide in bacteriostatic and bactericidal activities of honey. Front Microbiol. 2011;2:213. DOI:10.3389/fmicb.2011.00213
15. Goossens H. Antibiotic consumption and link to resistance. Clinical Microbiology and Infection. Clin Microbiol Infect; 2009, 12-15. DOI:10.1111/j.1469-0691.2009.02725.x
16. Goossens H, Lipsitch M. Global burden of antimicrobial resistance. Journal of Paediatrics and Child Health. 2022, 735-736. DOI:10.1111/jpc.15931
17. Murray CJ, Ikuta KS, Sharara F, Swetschinski L, Robles Aguilar G, Gray A, *et al*. Global burden of bacterial antimicrobial resistance in 2019: A systematic analysis. Lancet. 2022;399:629–655. DOI:10.1016/S0140-6736(21)02724-0
18. Prestinaci F, Pezzotti P, Pantosti A. Antimicrobial resistance: A global multifaceted phenomenon. Pathog Glob Health. 2015;109:309. DOI:10.1179/2047773215Y.0000000030
19. Ikuta KS, Swetschinski LR, Robles Aguilar G, Sharara F, Mestrovic T, Gray AP, *et al*. Global mortality associated with 33 bacterial pathogens in 2019: A systematic analysis for the Global Burden of Disease Study 2019. Lancet. 2022;400(10369):2221–2248. DOI:10.1016/S0140-6736(22)02185-7
20. Dadgostar P. Antimicrobial resistance: implications and costs. Infection and Drug Resistance. Dove Press; 2019, 3903-3910. DOI:10.2147/IDR.S234610

21. Ait Ouakrim D, Cassini A, Cecchini M, Plauchoras D. The health and economic burden of antimicrobial resistance. Eur J Public Health. 2020;30:23–44. DOI:10.1093/eurpub/ckaa165.1201
22. Hinchliffe S, Butcher A, Rahman MM. The AMR problem: demanding economies, biological margins, and co-producing alternative strategies. Palgrave Commun. 2018;4(1):1-12. DOI:10.1057/s41599-018-0195-4
23. de Kraker MEA, Stewardson AJ, Harbarth S. Will 10 Million People Die a Year due to Antimicrobial Resistance by 2050? PLoS Med. 2016;13(11):e1002184. DOI:10.1371/journal.pmed.1002184
24. Jonas OB, Irwin A, Berthe FCJ, Le Gall FG, Marquez P. Drug-resistant infections: A Threat to Our Economic Future. World Bank Rep. 2017;2:1-132. Available: https://www.worldbank.org/en/topic/health/publication/drug-resistant-infections-a-threat-to-our-economic-future
25. Mudenda S, Chabalenge B, Daka V, Mfune RL, Salachi KI, Mohamed S, *et al*. Global Strategies to Combat Antimicrobial Resistance: A One Health Perspective. Pharmacol Pharm. 2023 Aug 18;14(8):271-328. DOI:10.4236/PP.2023.148020
26. Godman B, Egwuenu A, Haque M, Malande OO, Schellack N, Kumar S, *et al*. Strategies to improve antimicrobial utilization with a special focus on developing countries. Life. 2021 Jun 7;11(6):528. DOI:10.3390/life11060528
27. Gahamanyi N, Umuhoza T, Saeed SI, Mayigane LN, Hakizimana JN. A Review of the Important Weapons against Antimicrobial Resistance in Sub-Saharan Africa. Appl Biosci. 2023 Apr 1;2(2):136-56.. DOI:10.3390/APPLBIOSCI2020011
28. Musoke D, Kitutu FE, Mugisha L, Amir S, Brandish C, Ikhile D, *et al*. A one health approach to strengthening antimicrobial stewardship in Wakiso District, Uganda. Antibiotics. 2020 Oct 31;9(11):764. DOI:10.3390/antibiotics9110764
29. Siachalinga L, Mufwambi W, Lee lH. Impact of antimicrobial stewardship interventions to improve antibiotic prescribing for hospital inpatients in Africa: a systematic review and meta-analysis. Journal of Hospital Infection. W.B. Saunders; 2022, 124–143. DOI:10.1016/j.jhin.2022.07.031
30. Saleem Z, Godman B, Cook A, Khan MA, Campbell SM, Seaton RA, *et al*. Ongoing Efforts to Improve Antimicrobial Utilization in Hospitals among African Countries and Implications for the Future. Antibiotics. 2022;11(12):1824. DOI:10.3390/antibiotics11121824
31. Mudenda S, Chabalenge B, Daka V, Mfune RL, Salachi KI, Mohamed S, *et al*. Global strategies to combat antimicrobial resistance: A one health perspective. Pharmacology & Pharmacy. 2023 Aug 18;14(8):271-328. DOI:10.11604/PAMJ.2023.45.152.41232
32. Espinosa-Gongora C, Jessen LR, Dyar OJ, Bousquet-Melou A, González-Zorn B, Pulcini C, *et al*. Towards a better and harmonized education in antimicrobial stewardship in European veterinary curricula. Antibiotics. 2021 Mar 30;10(4):364.. DOI:10.3390/antibiotics10040364
33. Dyar OJ, Huttner B, Schouten J, Pulcini C. What is antimicrobial stewardship? Clinical Microbiology and Infection. 2017 Nov 1;23(11):793-8.. DOI:10.1016/j.cmi.2017.08.026
34. Pokharel S, Raut S, Adhikari B. Tackling antimicrobial resistance in low-income and middle-income countries. BMJ Global Health. BMJ Specialist Journals; 2019 Nov 1;4(6):e002104.. DOI:10.1136/bmjgh-2019-002104
35. Calvo-Villamañán A, San Millán Á, Carrilero L. Tackling AMR from a multidisciplinary perspective: A primer from education and psychology. International Microbiology. Springer Science and Business Media Deutschland GmbH, 2023, 1-9. DOI:10.1007/s10123-022-00278-1
36. Annunziato G. Strategies to overcome antimicrobial resistance (AMR) making use of non-essential target inhibitors: A review. Int J Mol Sci. 2019 Nov 21;20(23):5844. DOI:10.3390/ijms20235844
37. Mudenda S, Hikaambo CN, Chabalenge B, Mfune RL, Mufwambi W, Ngazimbi M, *et al*. Antibacterial activities of honey against *Escherichia coli* and *Staphylococcus aureus*: A potential treatment for bacterial infections and alternative to antibiotics. J Pharmacogn Phytochem. 2023;12(3):06-13. DOI:10.22271/PHYTO.2023.V12.I3A.14655
38. Roca I, Akova M, Baquero F, Carlet J, Cavaleri M, Coenen S, *et al*. The global threat of antimicrobial resistance: Science for intervention. New Microbes and New Infections. 2015;6:22–29. DOI:10.1016/j.nmni.2015.02.007
39. Andrei G. Vaccines and Antivirals: Grand Challenges and Great Opportunities. Front Virol. 2021;1:666548. DOI:10.3389/fviro.2021.666548
40. Da Cunha BR, Fonseca LP, Calado CRC. Antibiotic discovery: Where have we come from, where do we go? Antibiotics. Multidisciplinary Digital Publishing Institute (MDPI), 2019 Apr 24;8(2):45. DOI:10.3390/antibiotics8020045
41. Boyd NK, Teng C, Frei CR. Brief Overview of Approaches and Challenges in New Antibiotic Development: A Focus on Drug Repurposing. Frontiers in Cellular and Infection Microbiology. Frontiers Media S.A. 2021 May 17;11:684515. DOI:10.3389/fcimb.2021.684515
42. Butler MS, Buss AD. Natural products - The future scaffolds for novel antibiotics? Biochem Pharmacol. 2006;71:919–929. DOI:10.1016/j.bcp.2005.10.012
43. Konaklieva MI. Addressing Antimicrobial Resistance through New Medicinal and Synthetic Chemistry Strategies. SLAS Discovery. Elsevier. 2019 Apr;24(4):419-39. DOI:10.1177/2472555218812657
44. Tacconelli E, Carrara E, Savoldi A, Harbarth S, Mendelson M, Monnet DL, *et al*. Discovery, research, and development of new antibiotics: the WHO priority list of antibiotic-resistant bacteria and tuberculosis. Lancet Infect Dis. 2018 Mar;18(3):318-27. DOI:10.1016/S1473-3099(17)30753-3
45. Khan S, Imran M, Imran M, Pindari N. Antimicrobial activity of various ethanolic plant extracts against pathogenic multi-drug resistant Candida spp. Bioinformation. 2017;13(3):67-72. DOI:10.6026/97320630013067
46. Khameneh B, Diab R, Ghazvini K, Fazly Bazzaz BS. Breakthroughs in bacterial resistance mechanisms and the potential ways to combat them. Microbial Pathogenesis. Academic Press; 2016 Jun 1;95:32-42. DOI:10.1016/j.micpath.2016.02.009
47. Oladunjoye I, Tajudeen Y, Oladipo H, El-Sherbini M. Planetary Health and Traditional Medicine: A Potential Synergistic Approach to Tackle Antimicrobial Resistance. Challenges. 2022 Jun 1;13(1):24.

DOI:10.3390/challe13010024
48. Gupta PD, Birdi TJ. Development of botanicals to combat antibiotic resistance. Journal of Ayurveda and Integrative Medicine. J Ayurveda Integr Med. 2017 Oct 1;8(4):266-75. DOI:10.1016/j.jaim.2017.05.004
49. Arip M, Selvaraja M, Mogana R, Tan LF, Leong MY, Tan PL, *et al*. Review on Plant-Based Management in Combating Antimicrobial Resistance - Mechanistic Perspective. Frontiers in Pharmacology. Frontiers Media S.A. 2022 Sep 29;13:879495. DOI:10.3389/fphar.2022.879495
50. Chandra H, Bishnoi P, Yadav A, Patni B, Mishra AP, Nautiyal AR. Antimicrobial resistance and the alternative resources with special emphasis on plant-based antimicrobials - A review. Plants. Plants (Basel); 2017, 457-462. DOI:10.3390/plants6020016
51. Silén H, Salih EYA, Mgbeahuruike EE, Fyhrqvist P. Ethnopharmacology, Antimicrobial Potency, and Phytochemistry of African Combretum and Pteleopsis Species (Combretaceae): A Review. Antibiotics. Multidisciplinary Digital Publishing Institute; 2023 Jan 28;12(2):264. DOI:10.3390/antibiotics12020264
52. Anand U, Jacobo-Herrera N, Altemimi A, Lakhssassi N. A comprehensive review on medicinal plants as antimicrobial therapeutics: Potential avenues of biocompatible drug discovery. Metabolites. Multidisciplinary Digital Publishing Institute; 2019 Nov 1;9(11):258. DOI:10.3390/metabo9110258
53. Yuan H, Ma Q, Ye L, Piao G. The traditional medicine and modern medicine from natural products. Molecules. 2016 Apr 29;21(5):559. DOI:10.3390/molecules21050559
54. Almuzaini AM. Phytochemicals: potential alternative strategy to fight Salmonella enterica serovar Typhimurium. Frontiers in Veterinary Science. Frontiers Media S.A. 2023 May 16;10:1188752. DOI:10.3389/fvets.2023.1188752
55. World Health Organization. Integrating Traditional Medicine in Health Care. In: World Health Organization [Internet]; c2023 [cited 17 Sep 2023]. Available: https://www.who.int/southeastasia/news/feature-stories/detail/integrating-traditional-medicine
56. Vaou N, Stavropoulou E, Voidarou C, Tsigalou C, Bezirtzoglou E. Towards advances in medicinal plant antimicrobial activity: A review study on challenges and future perspectives. Microorganisms. Multidisciplinary Digital Publishing Institute. 2021 Sep 27;9(10):2041. DOI:10.3390/microorganisms9102041
57. Chi C. Integrating traditional medicine into modern health care systems: Examining the role of Chinese medicine in Taiwan. Soc Sci Med. 1994 Aug 1;39(3):307-21. DOI:10.1016/0277-9536(94)90127-9
58. Park YL, Canaway R. Integrating traditional and complementary medicine with national healthcare systems for universal health coverage in Asia and the Western Pacific. Health Systems and Reform. Taylor & Francis. 2019 Jan 2;5(1):24-31. DOI:10.1080/23288604.2018.1539058
59. Fong HHS. Herbal Medicine in Modern Medical Practices. Integr Cancer Ther. 2002 Sep;1(3):287-93.
60. Ampomah IG, Malau-Aduli BS, Seidu A-A, Malau-Aduli AEO, Emeto TI. Integrating traditional medicine into the Ghanaian health system: perceptions and experiences of traditional medicine practitioners in the Ashanti region. Int Health. 2023 Jul;15(4):414-27. DOI:10.1093/inthealth/ihac059
61. Islas JF, Acosta E, G-Buentello Z, Delgado-Gallegos JL, Moreno-Treviño MG, Escalante B, *et al*. An overview of Neem (*Azadirachta indica*) and its potential impact on health. Journal of Functional Foods. Elsevier. 2023 Jul;15(4):414-27. DOI:10.1016/j.jff.2020.104171
62. Saleh Al-Hashemi ZS, Hossain MA. Biological activities of different neem leaf crude extracts used locally in Ayurvedic medicine. Pacific Sci Rev A Nat Sci Eng. 2016 Jul 1;18(2):128-31. DOI:10.1016/j.psra.2016.09.013
63. Herrera-Calderon O, Ejaz K, Wajid M, Shehzad M, Tinco-Jayo JA, Enciso-Roca E, *et al*. *Azadirachta indica*: Antibacterial activity of neem against different strains of bacteria and their active constituents as preventive in various diseases. Pharmacogn J. 2019;11(6s):1597-1604. DOI:10.5530/PJ.2019.11.244
64. Ahmed M, Marrez DA, Mohamed Abdelmoeen N, Abdelmoneem Mahmoud E, Ali MA-S, Decsi K, *et al*. Studying the Antioxidant and the Antimicrobial Activities of Leaf Successive Extracts Compared to the Green-Chemically Synthesized Silver Nanoparticles and the Crude Aqueous Extract from *Azadirachta indica*. Processes. 2023;11(6):1644. DOI:10.3390/pr11061644
65. Raja Ratna Reddy, Kumari K, Lokanatha, Mamatha, Damodar Reddy. Antimicrobial activity of *Azadirachta indica* (neem) leaf, bark and seed extracts. Int J Res Phytochem Pharmacol. 2013;3:1-4. Available: http://repo.lib.jfn.ac.lk/ujrr/handle/123456789/9557
66. Mahfuzul Hoque MD, Bari ML, Inatsu Y, Juneja VK, Kawamoto S. Anti-bacterial activity of guava (*Psidium guajava* L.) and neem (*Azadirachta indica* A. Juss.) extracts against foodborne pathogens and spoilage bacteria. Foodborne Pathog Dis. 2007;4(4):481–488. DOI:10.1089/fpd.2007.0040
67. Yaseen KH. Antibacterial activity of different part of Neem (Azadirchta indica) growing in Sharjah, United Arab Emirates. Iraqi J Sci. 2016;57(4B):2617–2626.
68. Hikaambo CN, Kaacha L, Mudenda S, Nyambe MN, Chabalenge B, Phiri M, *et al*. Phytochemical Analysis and Antibacterial Activity of *Azadirachta indica* Leaf Extracts against *Escherichia coli*. Pharmacol Pharm. 2022;13:1-10. DOI:10.4236/pp.2022.131001
69. Suhartono S, Soraya C, Shabira P. Antibiofilm activity of neem leaf (*Azadirachta indica* A. Juss) ethanolic extracts against Enterococcus faecalis *in vitro*. Dent J (Majalah Kedokt Gigi). 2023;56:98–103. DOI:10.20473/j.djmkg.v56.i2.p98-103
70. Wylie MR, Merrell DS. The Antimicrobial Potential of the Neem Tree *Azadirachta indica*. Frontiers in Pharmacology. Frontiers Media S.A.; 2022;13:891535. DOI:10.3389/fphar.2022.891535
71. Raut RR, Sawant AR, Jamge BB. Antimicrobial activity of *Azadirachta indica* (Neem) against Pathogenic Microorganisms. J Acad Ind Res. 2014;3(7):327–329.
72. Eid A, Jaradat N, Elmarzugi N. A Review of chemical constituents and traditional usage of Neem plant (*Azadirachta indica*). Palest Med Pharm J. 2017;2(2):75-81. DOI:10.59049/2790-0231.1060
73. Alzohairy MA. Therapeutics role of *Azadirachta indica* (Neem) and their active constituents in diseases prevention and treatment. Evidence-based Complementary and Alternative Medicine. Hindawi Limited, 2016, 7382506. DOI:10.1155/2016/7382506

74. Priyanka Dash S, Dixit S, Sahoo S. Phytochemical and Biochemical Characterizations from Leaf Extracts from *Azadirachta indica*: An Important Medicinal Plant. Biochem Anal Biochem. 2017;6(323):2161-1009. DOI:10.4172/2161-1009.1000323
75. Jerobin J, Makwana P, Suresh Kumar RS, Sundaramoorthy R, Mukherjee A, Chandrasekaran N. Antibacterial activity of neem nanoemulsion and its toxicity assessment on human lymphocytes *in vitro*. Int J Nanomedicine. 015 Oct 1;10(2):77-86. DOI:10.2147/IJN.S79983
76. Longden-Naufal C, Rolfe V, Mackonochie M. Narratives of Herbal Medicine Utilisation in the United Kingdom: Scoping Literature Review. Frontiers in Pharmacology. Frontiers Media S.A. 2022 Aug 25;13:886574.. DOI:10.3389/fphar.2022.886574
77. Mirzaie A, Halaji M, Dehkordi FS, Ranjbar R, Noorbazargan H. A narrative literature review on traditional medicine options for treatment of corona virus disease 2019 (COVID-19). Complementary Therapies in Clinical Practice. Elsevier; 2020, 101214. DOI:10.1016/j.ctcp.2020.101214
78. Ibáñez B, Melero A, Montoro A, Merino-Torres JF, Soriano JM, San Onofre N. A Narrative Review of the Herbal Preparation of Ayurvedic, Traditional Chinese, and Kampō Medicines Applied as Radioprotectors. Antioxidants. Multidisciplinary Digital Publishing Institute. 2023 Jul 17;12(7):1437. DOI:10.3390/antiox12071437
79. Sahrawat A, Sharma J, Rahul N S, Tiwari S, Joshi MD, Pundhir A, *et al*. Phytochemical analysis and Antibacterial properties of *Azadirachta indica*(Neem) leaves extract against *E. coli*. J Pharmacogn Phytochem. 2018;7(4):1368-71.
80. Muhammad A, Lawan IM, Abubakar A, Dangora II. Antimicrobial Activity of *Azadirachta indica* (Neem) Leave Extract against Some Clinical Isolates. Dutse J Pure Appl Sci. 2019;5(1b):97-104.
81. Momoh JO, Olaleye ON. Biochemical Characterization and Molecular Identification of *Escherichia coli* Isolate from Abattoir Wastes and Its Susceptibility to Ethanolic Root Extract of *Azadirachta indica* (neem). J Adv Microbiol. 2022;22(10):31–50. DOI:10.9734/jamb/2022/v22i1030502
82. Singh K. Phytochemical Analysis and Antimicrobial Activity of Various Indigenous Plant Species. Int J Res Appl Sci Eng Technol. 2021;9:241–245. DOI:10.22214/ijraset.2021.33067
83. Ogidi OI, NoahAyebabogha M, Eze PU, Okiemute O, Okafor CE. Determination of Phytoconstituents and Antimicrobial activities of aqueous and methanol extracts of Neem (*Azadirachta indica*) leaves. Int J Pharmacogn Chem. 2021;2:60-67. DOI:10.46796/ijpc.vi.155
84. Meressa MG. Phytochemical analysis and antibacterial evaluation of ethanol stem bark extract of *Azadirachta indica* Grown in Dibila Kebelle, Tigray-Ethiopia. Int J Eng Dev Res. 2017;5:1129–1132.
85. Kulkarni CP, Pandey A, Patil RC. Antibacterial and phytochemical analysis of Tinospora cordifolia, Azarchita indica and Ocimum santum leaves extract against common human pathogens: An *in vitro* study. J Pharmacogn Phytochem. 2017;6(5):702–706.
86. Mohammed CA, Stephens HM, Sadiq A. Phytochemical Analysis and Antimicrobial Activity of Neem Leaf, Lemon Grass and Mixture of Neem Leaf & Lemon Grass Essential Oil Extract (s) Against Some Selected Gram Positive and Gram Negative Organisms. Iconic Res Eng Journals. 2022;6:32–39.
87. Fatima A. Phytochemical screening and antibacterial activity of neem extracts on uropathogens. Pure Appl Biol. 2020;9(1):148–153. DOI:10.19045/bspab.2020.90018
88. Benisheikh AAG, Muhammad FM, Kelluri H, Aliyu ZM, Mallam UB, Jibrin MW. Phytochemical Extraction and Antimicrobial Studies on Crude Leaf Extract of *Azadirachta indica* (Neem) in Semi-Arid Region of Borno State, Nigeria. Int J Res Rev Vol. 2019;6(12):516–522. Available: www.ijrrjournal.com
89. Mustafa M. Antibacterial efficacy of neem (*Azadirachta indica*) extract against Enterococcus faecalis: An *in vitro* study. J Contemp Dent Pract. 2016;17(10):791–794. DOI:10.5005/jp-journals-10024-1932
90. Faujdar S, Bisht D, Sharma A. Antibacterial potential of neem (*Azadirachta indica*) against uropathogens producing beta-lactamase enzymes: A clue to future antibacterial agent? Biomed Biotechnol Res J. 2020;4(3):232–238. DOI:10.4103/bbrj.bbrj_38_20
91. Maleki L, Sadeghian-Rizi T, Ghannadian M, Sanati MH, Shafizadegan S, Sadeghi-Aliabadi H. Antibacterial Activity of *Azadirachta indica* Leaf Extracts Against Some Pathogenic Standards and Clinical Bacterial Isolates. Avicenna J Clin Microbiol Infect. 2018;5(1):12987. DOI:10.5812/ajcmi.12987
92. Sinaga M, Ganesan K, Kumar S, Nair P, Gani SB. Preliminary phytochemical analysis and *in vitro* antibacterial activity of bark and seeds of Ethiopian Neem (*Azadirachta indica* A. Juss). World J Pharm Pharm Sci. 2016;5(4):1714–1723. DOI:10.20959/wjpps20164-6479
93. Sharma Y, Dua D, Nupur S. Comparative study of different parts of *Azadirachta indica* (Neem) plant on the basis of antibacterial activity, phytochemical screening and its effect on rat PC-12 (pheochromocytoma) cell line. Int J Biotechnol Allied Fields. 2014;2(7):144–154.
94. Saleem W, Sarfraz B, Mazhar S. Combined Effect of Honey, Neem (*Azadirachta indica*), and Turmeric against Staphylococcus Aureus and *E. coli* Isolated from a Clinical Wound Sample. Biosci Rev. 2022;4(4):21–44. DOI:10.32350/bsr.44.01
95. Nigussie D, Davey G, Legesse BA, Fekadu A, Makonnen E. Antibacterial activity of methanol extracts of the leaves of three medicinal plants against selected bacteria isolated from wounds of lymphoedema patients. BMC Complement Med Ther. 2021;21:2. DOI:10.1186/s12906-020-03183-0
96. Divya Kumari P, Shenoy SM, Khijmatgar S, Chowdhury A, Lynch E, Chowdhury CR. Antibacterial activity of new atraumatic restorative treatment materials incorporated with *Azadirachta indica* (Neem) against Streptococcus mutans. J Oral Biol Craniofacial Res. 2019;9(4):321-325. DOI:10.1016/j.jobcr.2019.06.014
97. Mohammed HA, Omer AFA. Antibacterial Activity of *Azadirachta indica* (Neem) Leaf Extract against Bacterial Pathogens in Sudan. Am J Res Commun. 2015;3:246–251. Available: http://www.usa-journals.com/wp-content/uploads/2015/04/Mohammed_Vol35.pdf
98. Altayb HN, Yassin NF, Hosawi S, Kazmi I. *In-vitro* and in-silico antibacterial activity of *Azadirachta indica* (Neem), methanolic extract, and identification of Beta.d-Mannofuranoside as a promising antibacterial agent.

BMC Plant Biol. 2022;22(1):262. DOI:10.1186/s12870-022-03650-5
99. Ibrahim N, Kebede A. *In vitro* antibacterial activities of methanol and aqueous leave extracts of selected medicinal plants against human pathogenic bacteria. Saudi J Biol Sci. 2020;27(9):2261–2268. DOI:10.1016/j.sjbs.2020.06.047
100. Senthilkumar P, Rashmitha S, Veera P, Vijay Ignatious C, SaiPriya C, Samrot A V. Antibacterial activity of neem extract and its green synthesized silver nanoparticles against pseudomonas aeruginosa. J Pure Appl Microbiol. 2018 Jun 1;12(2):969-74. + DOI:10.22207/JPaM.12.2.60
101. Singh AK, Sharma RK, Sharma V, Singh T, Kumar R, Kumari D. Isolation, morphological identification and *in vitro* antibacterial activity of endophytic bacteria isolated from *Azadirachta indica* (neem) leaves. Vet World. 2017;10(5):510–516. DOI:10.14202/vetworld.2017.510-516
102. Pandey G, Verma K, Singh M. Evaluation of phytochemical, antibacterial and free radical scavenging properties of *Azadirachta indica* (neem) leaves. Int J Pharm Pharm Sci. 2014;6(2):444–447.
103. Ali E, Islam MS, Hossen MI, Khatun MM, Islam MA. Extract of neem (*Azadirachta indica*) leaf exhibits bactericidal effect against multidrug resistant pathogenic bacteria of poultry. Vet Med Sci. 2021;7(5):1921–1927. DOI:10.1002/vms3.511
104. Biswas K, Chattopadhyay I, Banerjee RK, Bandyopadhyay U. Biological activities and medicinal properties of neem (*Azadirachta indica*). Curr Sci. 2002;82:1336-1345. Available: https://www.jstor.org/stable/24106000
105. Sarah R, Tabassum B, Idrees N, Hussain MK. Bio-active compounds isolated from neem tree and their applications. Natural Bio-active Compounds. Springer Singapore; 2019, 509-528. DOI:10.1007/978-981-13-7154-7_17
106. Al-Sarraj FMB. A review on the impacts of *Azadirachta indica* on multi-drug resistant extended spectrum beta Lactamase-positive of *Escherichia coli* and Klebsiella pneumonia. Adv Life Sci. 2021;8:228-232. Available: https://www.als-journal.com/833-21/
107. Nguyen TL, Bhattacharya D. Antimicrobial activity of quercetin: an approach to its mechanistic principle. Molecules. 2022 Apr 12;27(8):2494. DOI:10.3390/molecules27082494
108. Azimkhanova BB, Ustenova GO, Sharipov KO, Rakhimov KD, Sayakova GM, Jumagaziyeva AB, *et al*. Chemical Composition and Antimicrobial Activity of Subcritical CO2Extract of *Lepidium latifolium* L. (Brassicaceae). Int J Biomater. 2021, 4389967. DOI:10.1155/2021/4389967