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ORIGINAL RESEARCH

Phytochemical Screening and Antimicrobial Properties of *Parkia biglobosa* (African Locust Bean) (Jacq) Leaf Extracts on Selected Microbial Isolates

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ABSTRACT

Globally, antimicrobial resistance poses a significant threat to the effectiveness of existing antibiotics and remains a major public health concern. This challenge necessitates the exploration of alternative antimicrobial agents from medicinal plants. This study evaluated the phytochemical, constituents and antimicrobial activity of *Parkia biglobosa* (locust bean) ethanol and aqueous leaf extracts against selected bacterial and fungal isolates. Bioactive compounds were extracted using the maceration method, followed by phytochemical screening. Microbial isolates (*Rhizopus stolonifer, Aspergillus niger, Escherichia coli,* and *Staphylococcus aureus*) were obtained from the Federal Teaching Hospital, Lokoja, and cultured using standard microbiological techniques. Antimicrobial activity was assessed using the agar well diffusion method, while Minimum Inhibitory Concentration (MIC) was determined by the tube dilution technique. Data were analyzed using SPSS version 23.0. Phytochemical analysis revealed the presence of saponins, phenols, tannins, terpenoids, and flavonoids in both extracts, while alkaloids was detected only in the ethanolic extract. Antimicrobial activity increased with conentration, with the highest inhibition observed at 100 mg/mL. *Aspergillus niger* showed the highest inhibition (14.00 ± 1.00 mm), followed by *E. coli* (13.00 ± 1.00 mm), *S. aureus* (11.00 ± 1.00 mm), and *R. stolonifer* (11.50 ± 1.50 mm). Furthermore, The Minimum Bactericidal Concentration (MBC) of aqueous and ethanol extracts against *S. aureus* were 75 mg/mL and 50 mg/mL respectively. For *E. coli*, the aqueous extract's MBC was 50 mg/mL while the ethanol extract's was 75 mg/mL. The Minimum Fungicidal Concentrations (MFC) for *Rhizopus stolonifer* were 75 mg/mL and 50 mg/mL, while for *Aspergillus niger*, values were 50 mg/mL and 75 mg/mL for aqueous and ethanolic extracts. Overall, the study demonstrates that *P. biglobosa* leaf extracts exhibit notable antimicrobial activity, supporting their traditional use and highlighting their potential as a source of novel antimicrobial agents against resistant pathogens, but was limited to some selected microbes.

Keywords: Extracts, Isolates, *Parkia biglobosa*, phytochemical activity, antimicrobial resistance

INTRODUCTION

Parkia biglobosa (Jacq.) commonly known as the African locust bean, is a perennial leguminous tree belonging to the Fabaceae family (Audu et al., 2025). It is native to West Africa, including northern and southwestern Nigeria, and typically grows to a height of 7–20 m. The plant is highly valued for its nutrient-dense seeds, which are rich in protein, essential amino acids, fatty acids, vitamins, dietary fiber, and possess high caloric content (Ilesanmi et al., 2024). Traditionally, the seeds are fermented to produce local condiments such as *iru* (Yoruba), *dawadawa* (Hausa), and *ogiri* (Igbo). This fermentation process enhances digestibility and flavor while involving beneficial microorganisms that can inhibit pathogenic organisms such as *Bacillus subtilis*, *Staphylococcus aureus*, and *Escherichia coli* (Aliyu et al., 2025).

In addition to its nutritional importance, *P. biglobosa* has been widely utilized in traditional medicine for the treatment of various infectious diseases, including its use as an antimalarial and antibacterial agent. However, despite these ethnomedicinal applications, there remains limited comprehensive scientific validation of its antibacterial properties (Kabir et al., 2024).

The global rise in antimicrobial resistance (AMR), particularly among pathogens such as *Staphylococcus aureus* and *Escherichia coli*, largely driven by antibiotic misuse and environmental contamination, has resulted in increasing treatment failures (ME et al*.,* 2025). This growing public health challenge underscores the urgent need to explore novel antimicrobial agents from natural sources. Therefore, this study aims to evaluate the phytochemical constituents and antimicrobial activities of *P. biglobosa* leaf extracts against selected microbial strains

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MATERIALS AND METHODS

Area of Study

The study was carried out in Lokoja, Kogi State (figure 1). It is an educational and administrative settlement, which brought about the establishment of new settlement for non-residents of Lokoja city. Lokoja town is located on the west bank of the Niger River. Besides being an important commercial settlement, the site originally ceded to the British in 1841 by the King of Idah, 50miles selected for the first British consulate. The original site of Lokoja, is a 1,349-foot- (411-metre) high mass of oolitic iron ore. Lokoja is situated on the local highway between Kabba and Anyagba and has ferry service across the Niger River.

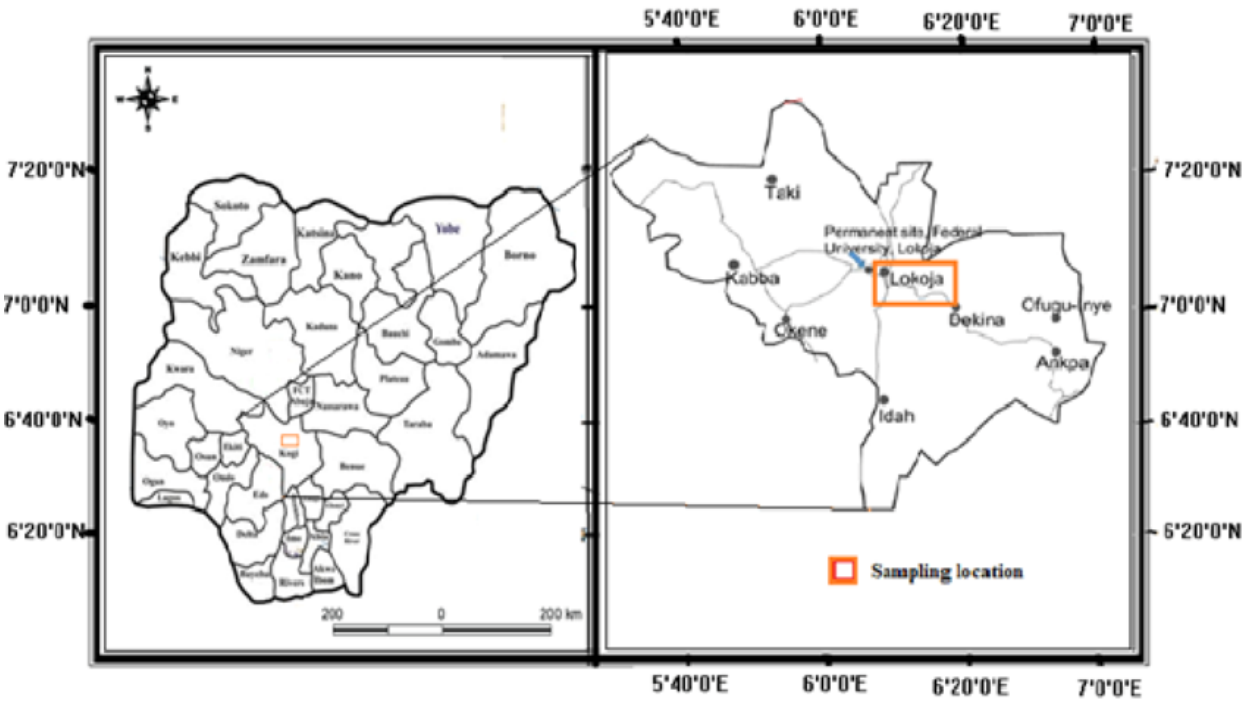

Figure 1: Map showing the study location (Source: Ojiego et al., 2022)

Sample Collection

Thirty (30) fresh leaves of *Parkia biglobosa* were collected from Adankolo campus of Federal University Lokoja during the dry season and transported to the herbarium for identification. The plant material was confirmed by Mr Adebowale, a plant taxonomist at the Herbarium, Department of Botany, Federal University Lokoja. Microbial isolates (*Rhizopus stolonifer, Aspergillus niger, E. coli* and *S. aureus*) were gotten from the Federal Teaching Hospital with the help of a medical laboratory scientist. The samples were stored in an insulated box and transported to the Biological Sciences laboratory of Federal University Lokoja where they were subcultured for authentication and stored on agar plates at 4ºC in a refrigerator for further processing and analysis.Two isolates were used which are fungi and bacteria.

Extraction of Plant Material

The fresh leaves were rinsed thoroughly two to three times with running tap water followed by a wash with sterile water to remove impurities. They were air-dried at room temperature to constant weight for two weeks, then homogenized to fine powder using an electric blender. The plant sample was shielded from sunlight to ensure that the active compounds remain intact (Ihuma et al., 2022). The powdered leaves were stored in labeled, sealed reagent bottles for further processing. Bioactive compounds were extracted via maceration. (Bothon et al., 2023)

100g powdered leaves were weighed, placed in two separate containers and mixed with 500ml absolute ethanol (99.5%) and distilled water for ethanol and aqueous extraction respectively. Each mixture was shaken thoroughly and left to macerate for 72 hours at room temperature. After filtering the solution, the extract was heated at 50°C to evaporate solvents completely with the use

of water bath for two to three hours for ethanol and aqueous extract respectively (Musa et. al., 2025). The resulting crude extract was collected and weighed at different concentrations.

Phytochemical Analysis

The extract of *Parkia biglobosa* leaves were examined for the presence of Alkaloids, flavonoid, saponins, tannins, phenols and terpenoids following the method of Yadav et al. (2014) and Entonu et al. (2023) as follows:

Alkaloids

The alkaloid assay was carried out by dissolving the extract in dilute hydrochloric acid, filtered, and tested with Wagner's reagent (iodine in potassium iodide). A brown or reddish precipitate indicated the presence of alkaloids (Ishaya et al., 2025).

Flavonoids

A few drop of sodium hydroxide solution were added to each extract. The yellow color that developed was removed upon addition of dilute acid, indicating the presence of flavonoids (Ishaya et al., 2025).

Saponins

Two grams of the powdered sample were boiled in 20 mL of distilled water, filtered, and cooled. The filtrate was then tested for saponins using frothing and emulsification methods (Ishaya et al., 2025), where the formation of stable froth and emulsion indicated their presence.

Phenols

0.2g samples treated with 5% ferric chloride solution; development of a deep blue color indicated the presence of phenols (Ishaya et al., 2025).

Tannins

Two milliliters of a 2% ferric chloride solution were added to the extracts, the appearance of a blue-green or blue-black coloration signified tannins (Ishaya et al., 2025).

Terpenoids (Salkowski Test)

Five ml sample mixed with 2ml chloroform, layered with 3 ml concentrated sulfuric acid; development of reddish-brown precipitate indicated terpenoids (Ishaya et al., 2025).

Culture Media

Microbial isolates were inoculated onto sterilized agar plates, allowed to solidify, and incubated at 37 °C for 24 hours for bacterial strains and 25 °C for 48 hours for fungal pathogens (Khedr et al., 2024; Akanni et al., 2026). Pure cultures with distinct colonies were examined microscopically. Fungal isolates were prepared on clean slides with a drop of water, covered with a cover slip, and observed at ×40 magnification, with identification based on mycelial structure, fruiting bodies, and spore characteristics using a mycological atlas and morphological references (Sarah et al., 2016). Bacterial isolates were identified using morphological features from Bergey's Manual of Determinative Bacteriology, followed by Gram staining and observation under ×100 oil immersion.

Antimicrobial Sensitivity Test

Antimicrobial activities were assessed in triplicate for each extract using the Agar Well Diffusion method. Culture plates were inoculated with isolates; wells (14 millimeters diameter) were bored on the agar surface with a cork borer. 60 microliters of extracts were introduced into each well using sterile syringes at different concentrations. Plates were labeled and incubated at 37 ºC for 24 hours for bacterial strain and 25 ºC for 48 hours for fungal pathogens (Khedr et al*.,* 2024; Akanni et al., 2026), then observed for zones of inhibition. Inhibition zones were measured in centimeters using a ruler and recorded.

Determination of Minimum Inhibitory Concentration (MIC)

Minimum Inhibitory Concentration (MIC) for *Escherichia coli* and *Staphylococcus aureus* was determined using the tube dilution method with serial dilutions in nutrient broth (Aliyu et al., 2025). An extract solution (200 mg/mL) was serially diluted in double- and single-strength media, with a control tube containing no extract. Each tube received 0.2 mL of the test organism inoculum, was incubated at 37 °C overnight, and examined for turbidity. The lowest concentration showing no visible growth was recorded as the MIC (Oladeji et al., 2025).

Determination of Minimum Fungicidal Concentration (MFC)

Fungicidal activity was assessed by subculturing samples from MIC assay tubes showing no visible fungal growth onto Potato Dextrose Agar (PDA) plates, followed by incubation at 37 °C for 48 hours. The Minimum Fungicidal Concentration (MFC) was defined as the lowest extract concentration that produced no fungal colonies on the medium. The procedure involved pouring melted, cooled PDA into Petri dishes and aseptically spreading 0.1 mL aliquots from the selected

tubes onto the agar surface, with plates incubated and examined for growth. The lowest concentration with no detectable fungal growth was recorded as the MFC (Entonu et al., 2023).

Data Analysis

Each zone of inhibition was recorded and presented in tabular form showing extract concentrations and mean inhibition zones. One-way analysis of variance (ANOVA) test was used to determine significant differences among treatments. Results are presented as mean ± standard deviation, with all analyses performed using SPSS version 23.0.

RESULTS

Tannins, alkaloids, flavonoids, saponins, terpenoids, and phenols, were detected in the ethanoic extract (Table 1) whereas in the aqueous extract all the phytochemicals were present except for alkaloids which was absent.

Table 1: Phytochemical constituents of *Parkia biglobosa* Ethanoic/Aqueous Leave Extract

Phytochemicals	Status	
	Ethanoic	Aqueous
Saponins	+	+
Phenols	+	+
Tannins	+	+
Terpenoids	+	+
Alkaloid	+	-
Flavonoids	+	+

+ Present; -Absent

The antimicrobial effectiveness of *P. biglobosa* leaf extract was evaluated against several microbial isolates, including *Escherichia coli*, *Staphylococcus aureus*, *Aspergillus niger*, and *Rhizopus stolonifer* at concentrations of 25 mg/mL, 50 mg/mL, 75 mg/mL, and 100 mg/mL.

Results showed that the extract's effectiveness increased with concentration, with the highest antimicrobial activity observed at 100 mg/mL across all tested isolates. Table 2 displays the mean zones of inhibition for the ethanol extract of *P. biglobosa* leaf. The inhibition zone for *Rhizopus stolonifer* was greatest at 100 mg/ml (11.50±1.50 mm) and nonexistent at 25 mg/ml. *Aspergillus niger* showed maximum inhibition at 100 mg/mL (14.00±1.00 mm) and least inhibition (6.00±1.00 mm) at the lowest concentration. For *E. coli*, the highest inhibition was 13.00±1.00 mm at 100 mg/mL, while *S. aureus* recorded 11.00±1.00 mm.

Table 2: Antimicrobial Activity of *P. biglobosa* Against Selected Microbial Isolates

Isolate	Concentration			
	25 mg/mL	50 mg/mL	75 mg/mL	100 mg/mL
Bacteria				
S. aureus	0.00	7.00±3.00	10.00±2.00	11.00±1.00
E. coli	6.50±0.50	10.00±0.00	11.00±1.00	13.00±1.00
Fungi				
A. niger	6.00±1.00	8.00±2.00	12.50±0.50	14.00±1.00
R. stolonifer	0.00	7.00±1.00	9.00±1.00	11.50±1.50

Values represent mean ± SEM from duplicate readings.

The Minimum Bactericidal Concentration (MBC) of aqueous and ethanol extracts against *S. aureus* were at 75 mg/mL and 50 mg/mL respectively. For *E. coli*, the aqueous extract's MBC was observed at 50 mg/mL while the ethanol extract's was at 75 mg/mL. The Minimum Fungicidal Concentrations (MFC) for *Rhizopus stolonifer* were observed at 75 mg/mL and 50 mg/mL, while for *Aspergillus niger*, values were at 50 mg/mL and 75 mg/mL for aqueous and ethanolic extracts respectively (Table 3).

Table 3: MIC of *Parkia biglobosa* Against Selected Microbial Isolates

	Concentration			
Isolates	**ETH (mg/mL)**	**AQS (mg/mL)**	**ETH (mg/mL)**	**AQS (mg/mL)**
Bacteria (MBC)	100	75	-	50
S. aureus				
E. coli	50	75	50	50
Fungi (MFC)				
A. niger	75	75	50	75
R. stolonifer	100	-	-	50

ETH = ethanol extract; AQS = aqueous extract.

DISCUSSION

Saponins, tannins, flavonoids, terpenoids, and phenols, were identified in the plant extract similarly reported by Kabir et al. (2024) and Audu et al., (2025). These phytochemicals are found naturally in many plants and are recognized for their biological activity, including bactericidal and fungicidal properties (Ihuma et al., 2022), which must have contributed to the antimicrobial effects observed in the present study. The absence of alkaloids in aqueous extract of leaves of *P. biglobosa* is in conformity with that of Ajaiyeoba (2002) and Iheukwumere (2025), an indication that some alkaloids are poorly soluble in water, especially since alkaloids were present in the ethanoic extract of the same stem bark.

The medicinal properties of *P. biglobosa* are probably due to the presence of some of these phytochemicals because they have been attributed to antimicrobial activity in several medicinal plants (Damter et al., 2024; Kabir et al., 2024; Aliyu et al**.,** 2025). Our findings indicated that the leaves of *P. biglobosa,* when macerated at an appropriate concentration can effectively inhibit the

growth of microbial organisms as described by Ihuma et al. (2022), who also reported the growth inhibition of *S. aureus* and *E. coli* at different concentration using the stem bark of *P. biglobosa.* The increase in antimicrobial activity with extract concentration observed in this study aligns with Kabir et al. (2024), who reported a direct relationship between concentration and antimicrobial potency.

By and large, the leaves of *P. biglobosa* demonstrate considerable antimicrobial effectiveness against the isolates, especially at concentrations of 100% and 75%. The zone of inhibition expanded as the extract concentration increased, indicating a positive correlation. For instance, no inhibition zones were observed against *Rhizopus stolonifer* and *Staphylococcus aureus* at 25 mg/ml; however, at higher extract concentrations inhibition was observed and this is consistent with the report of Akanni et al. (2024). Notably, *E. coli* showed significant increases in inhibition with increase in concentration of the leaf extract.

The Inhibitory effects of the leaf extracts may be attributed to their phytochemical constituents which is consistent with ME et al. (2025), who reported antimicrobial activity of stem bark extracts against similar organisms. Overall, *P. biglobosa* leaf extracts contain active antimicrobial compounds that, upon purification, could be developed into effective agents against microbial resistance which aligns with the findings of Kabir et al. (2024).

CONCLUSION

Phytochemical analysis revealed the presence of saponins, tannins, flavonoids, phenols, and alkaloids, while terpenoids were detected only in the ethanolic extract, highlighting the effect of solvent type on compound extraction. This study also showed that *Parkia biglobosa* leaf extracts

possess significant antimicrobial activity against *Escherichia coli*, *Staphylococcus aureus*, *Aspergillus niger*, and *Rhizopus stolonifer*.

The antimicrobial effect was concentration-dependent, with maximum inhibition at 100 mg/mL and minimal activity at lower concentrations. The ethanolic extract demonstrated greater efficacy than the aqueous extract. MBC and MFC results further confirmed both bactericidal and fungicidal properties.

These findings support the traditional use of *P. biglobosa* in treating microbial infections and indicate its potential as a source of new antimicrobial agents against resistance. Further research should focus on isolating active compounds, evaluating safety, conducting in vivo studies, understanding mechanisms of action, testing resistant strains, and developing standardized pharmaceutical formulations.

AUTHOR'S CONTRIBUTION

Maimuna, I: Conceptualization, Writing - review & editing; Kayode, A: Writing - original draft, Data curation; Tanko, D.: Review and editing. All authors contributed to the development of the final manuscript and approved its submission.

CONFLICT OF INTEREST

The authors declare they have no competing interest.

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REFERENCES

Ajaiyeoba, E. O. (2002). Phytochemical and Antimicrobial Properties of *Parkia biglobosa* and *Parkia* bicolar leaf extracts. *African journal of Biomedical Research.* 5(3): 125-129

Akanni, G. B., Njobeh, P., Adeyanju, A. A., Mahlanza, Z., Maleke, M., Olatunde, S. J. & Adebo, O. A. (2026). Legume-based Fermented Foods and Beverages: Nutritional Aspects and Health Implications. In *Fermented Foods and Beverages: Functionalities and Nutritional Aspects*. GB: CABI. pp. 36-64

Akanni, I. O., Onaolapo, J. A., Bolaji, R. O., Danmalam, H. U. & Udobi, C. E. (2024). Antibacterial and antibiofilm activities of aqueous fraction of methanol extract from stem-bark of Parkia biglobosa against chronic wounds' pathogens. *Nigerian Journal of Pharmaceutical and Applied Science Research*, 13(2): 11-20.

Aliyu, A. H., Dawaki, A. M., Abubakar, B. A., Idowu, A., Musa, M. M. & Mohammed, S. (2025). Phytochemical Screening and Anti-Microbial Activity of Parkia biglobosa Seed'Oil Extract. *BIMA Journal of Science and Technology Gombe*, 9(1B): 267-273.

Audu, H., Musa, A. & Bello, Y. (2025). Comparative study of phytochemicals and proximate composition of *Parkia biglobosa*, *Xylopia aethiopica*, and *Aframomum melegqueta* for nutraceutical applications. *Sahel Journal of Life Sciences FUDMA,* 3(1): 87-91.

Bothon, F. T. D., Atindéhou, M. M., Koudoro, Y. A., Lagnika, L. & Avlessi, F. (2023). Parkia biglobosa Fruit Husks: Phytochemistry, Antibacterial, and Free Radical Scavenging Activities. *American Journal of Plant Sciences*, 14(2): 150-161.

Damter, S. A., Dastu, A. J., Guluwa, L. Y., Gofwan, G. P., Womnokol, D. P. & Gyang, I. (2024). Phytochemical Screening of *Parkia biglobosa* Stem Bark and Leaves Aqueous Extract. *Nigerian Journal of Animal Production,* 7(3): 659-661. https://doi.org/10.51791/njap.vi.5496.

Entonu, E. E., Ngene, A. C., Odu, C. E., Ogaji, A. O., Nguhemen, H. W., Ochiheche, E. G. & Egbere, J. O. (2023). Evaluation of antifungal effect of Parkia biglobosa and Vitellaria paradoxa against selected pathogenic fungi. *Mycopath*, 21(1): 25-32.

Iheukwumere, I. H., Ajeh, J. C., Iheukwumere, C. M., Ike, V. E., Obianom, A. O., Ihenatuoha, U. A. & Ilechukwu, C. C. (2025). Exploring the Phytochemical and Antimicrobial Properties of Fruit Vinegar: A Study on Phoenix Dactylifera and Malus Sylvestris. *IPS Journal of Applied Microbiology and Biotechnology*, 4(1): 115-122.

Ihuma, J. O., Kure, S. & Famojuro. (2022). Antimicrobial Effect of the Stem Bark Extracts of *Parkia biglobosa* (Jacq.) G. Don on *Escherichia coli* and *Staphylococcus aureus. Asian Journal of Biology,* 116(3): 1-9.

Ilesanmi, V. O., Adegbehingbe, K. T., Oyeniyi, D. O., Oke, O. G. & Dada, A. D. (2024). In-vitro antibacterial activities of fermented and unfermented *Parkia biglobosa* seeds against selected entero-pathogens. *Vegetos*, 37(2): 566-577.

Ishaya, S. G., Khan, N., Ali, A. B., Edo, G. I., Mafe, A. N., Yousif, E. & Umar, H. (2025). Evaluation of Cymbopogon citratus (lemongrass) leaf extract: phytochemical screening, antimicrobial activities, cytotoxicity, antioxidant properties, and toxicological assessment. *Drug and Chemical Toxicology*, 16(2): 1-13.

Kabir, D., Yusuf, D. & Lawan, I. (2024). Correlation between Phytochemicals Content, Antioxidant and Antimicrobial Activities of the Leaf, Seed, Bark, and Root Extracts of Parkia biglobosa. *Journal of Applied Science and Environmental Studies*, *7*(4): 327-342.

Khedr, N. M., A. A. Ibrahim, M. El-Metwally, M., Eldakroory, S. & Soliman, M. I. (2024). Phytochemical analysis, antioxidant activity, antimicrobial evaluation, and cytotoxicity effects of wild medicinal plants. *SABRAO J. Breed. Genet,* 56(4): 1552-1562.

ME, S., El-Sherbiny, G. M., Sharaf, M. H., Kalaba, M. H. & Shaban, A. S. (2025). Phytochemical analysis, antimicrobial, antioxidant, and cytotoxicity activities of *Schinus molle* (L.) extracts. *Biomass Conversion* and *Biorefinery*, 15(3): 3753-3770.

Musa, M. & Aji, A. (2025). Antimicrobial activities of selected Fabaceae plants found in Northern Nigeria: a systematic and critical review. *Journal of Natural Products and Drug Development*, 1(1): 8-20.

Ojiego, B. O., Ogu, G. I., Dantanko, F., Okolo, J. C. & Audu, K. E. (2022). Health risk assessment for selected heavy metals in Telfairia occidentalis (fluted pumpkin) leaf retailed in markets within Lokoja metropolis. SVU International Journal of Agricultural Science. 4(3): 125-134.

Oladeji, O. A., Taiwo, K. A., Ogidi, C. O. & Faturoti, A. O. (2025). Production, nutritional benefits, limitations and strategies for enhancing the national value of fermented native condiments from selected legumes and wild seeds in Nigeria. *Discover Food*, *5*(1): 142.

Sarah, K., Catriona, H., Helen, A. & David, E. (2016). Description of Medical Fungi. Third Edition (Revised November, 2016). Australia. pp, 279.

Udobi, C. E., Abdulrahman, F. & Hassan, M. (2025). Nutritional Values and Antimicrobial Potential of African Locust bean (*Parkia biglobosa*) (Mimosaceae). Sahel *Journal of Applied Chemistry.* 7(1): 49-55.

Yadav, M., Chatterji, S., Gupta, S. K. & Watal, G. (2014). Preliminary Phytochemical Screening of Six Medicinal Plants used in Traditional Medicine. *International Journal of Pharmaceutical Science,* 6: 539-542.