

Biochemical Investigations, Antimicrobial Activity, and Metallic Nanoparticle Synthesis Using Aqueous Extract of *Alchornea laxiflora* Leaf

Loveth Olawamide Adebayo^{1*} , Margaret O Oseni² , Olatunde Abass Oseni³ 

¹Department of Science Laboratory Technology, Faculty of Science, Ekiti State University, Ado-Ekiti, Nigeria

²Department of Chemistry, Federal University, Oye-Ekiti, Nigeria

³Department of Medical Biochemistry, College of Medicine, Ekiti State University, Ado-Ekiti, Nigeria

ARTICLE INFO

Article Type:
Research

Article History:

Received: 11 Oct 2024

Revised: 29 Nov 2024

Accepted: 15 Dec 2024

Available online: 23 Sep 2025

Keywords:

Alchornea laxiflora,
Phytochemicals,
Antioxidants,
Toxicity tests,
Anti-infective agents,
Green chemistry technology

*** Corresponding author:**

E-mail: adebayoloveth04@gmail.com

ABSTRACT

Introduction: Medicinal plants represent valuable sources for novel therapeutic compounds, with *Alchornea laxiflora* being traditionally used in West African medicine. This study aimed to characterize the biochemical composition, antioxidant activity, toxicological safety, antimicrobial efficacy, and green synthesis potential of *A. laxiflora* aqueous extract.

Methods: Fresh *A. laxiflora* leaves were collected from Akure, Nigeria and subjected to aqueous extraction following standard protocols. Phytoconstituents were identified through qualitative screening. Antioxidant activity was assessed through DPPH scavenging and FRAP assays. Antimicrobial activity was evaluated against bacterial and fungal pathogens using agar well diffusion. Metallic nanoparticles were synthesized and characterized by UV-Vis and FTIR spectroscopy. Acute toxicity was studied in Wistar rats (200 and 400 mg/kg doses) with monitoring of liver enzymes.

Results: The extract contained alkaloids, flavonoids, tannins, saponins, and phenolic compounds. Significant antioxidant activity was observed in DPPH and FRAP assays ($p < 0.05$). No adverse effects were noted in toxicity studies ($p > 0.05$). The extract showed antimicrobial activity against all tested pathogens. Synthesized nanoparticles demonstrated enhanced antimicrobial properties.

Conclusion: This study validates the traditional use of *A. laxiflora* and supports its potential for pharmaceutical development in antimicrobial and nanomedicine applications.

Please cite this paper as:

Adebayo LO, Oseni MO, Oseni OA. Biochemical investigations, antimicrobial activity, and metallic nanoparticle synthesis using aqueous extract of *Alchornea laxiflora* Leaf. Journal of Biochemicals and Phytomedicine. 2025; 4(2): 50-59. doi: 10.34172/jbp.2025.17.

Introduction

Alchornea laxiflora, commonly known as Lowveld Bead String in English and Ewe Pepe in Yoruba, is a member of the Euphorbiaceae family found widely across tropical Africa, particularly in forested areas and along riverbanks (Siwe-Noundou, 2019). It typically grows as a shrub or small tree, reaching heights between 3 to 10 meters, with glossy, dark green leaves that are ovate to elliptic in shape, serrated at the margins, and arranged alternately. The plant is deciduous, producing small, greenish-yellow flowers in lax inflorescences, which develop into three-lobed capsules containing black seeds.

Medicinally, various parts of *A. laxiflora*, including its leaves, bark, and roots, are utilized in traditional African medicine (Siwe-Noundou et al., 2019). It is employed to treat a diverse array of ailments such as malaria, respiratory issues, gastrointestinal disorders, and skin infections due to its rich phytochemical content. The key bioactive compounds identified include flavonoids, tannins, saponins, alkaloids, and phenolic acids, which contribute to its antimicrobial, anti-inflammatory, antioxidant, and analgesic properties. Beyond its medicinal uses, *A. laxiflora* plays ecological roles by providing habitat and food for wildlife and contributing to soil improvement and erosion control in agroforestry systems. Culturally, it holds significance in African communities for its medicinal and practical applications (Jain et al., 2022; Olajire et al., 2017).

A. laxiflora has a diverse phytochemical composition, including alkaloids, flavonoids, phenolic compounds, terpenoids, fatty acids, and other chemicals. Flavonoids like quercetin derivatives and phenolic chemicals like ellagic acid are especially plentiful, which contribute to its antioxidant, anti-inflammatory, and antibacterial effects. The plant exhibits diverse pharmacological activities including antidiabetic effects through insulin sensitization, anticancer properties via apoptosis induction, antimicrobial actions against a range of pathogens, and antioxidant capabilities against oxidative stress. Additionally, it shows promise in treating anemia by improving hematological indices, supports reproductive health by regulating uterine contractions, and possesses anticonvulsant and anti-inflammatory activities. These findings underscore *A. laxiflora* significance in traditional medicine across Africa and its potential as a source for developing therapeutic agents against various health (Olasehinde et al., 2015, Oloyede et al., 2011).

Research highlights diabetes, microbial infections, cancer, and inflammatory diseases among others as the ailments for which it has been shown to be therapeutically effective. Nanotechnology applications have potential opportunities to improve the efficacy of *Alchornea*-derived pharmaceuticals by increasing solubility,

bioavailability, and targeted administration. However, toxicity studies have revealed possible dangers associated with high dosages, such as hepatotoxicity, nephrotoxicity, and gastrointestinal discomfort, underlining the significance of regulated use to limit adverse effects while maximizing therapeutic benefits (Jain et al., 2022). The aims of this study were to determine the biochemical composition, antioxidant activity, toxicological safety, antibacterial effectiveness, and green synthesis of an aqueous extract of.

Materials and Methods

A. laxiflora plant matter (the leaves) were collected in January 2024 in Akure, Ondo State, Nigeria. This was done in the early morning to prevent moisture loss and maximize phytochemical content. The plant sample specimen was authenticated and stored in the herbarium of the Department of Plant Science, Faculty of Science, Ekiti State University, Ado Ekiti, Nigeria with number (UHAE 2420049).

Preparation of Plant Leaf

The leaves of *A. laxiflora* were carefully removed from the plant and rinsed under running tap water to remove any surface dust or contaminants. The cleaned leaves were then air-dried for two weeks at room temperature (25°C). The dried leaves were ground into a fine powder using an electric grinding machine.

Aqueous Extraction

A. laxiflora leaves were extracted according to WHO guidelines using a 20:100 (w/v) ratio of powdered leaves to distilled water. The mixture was shaken continuously at 150 rpm for 24 hours, filtered, and stored at 4°C for stability. The filtrate was then used for analysis within a week of extraction (Olajire et al., 2018).

Phytochemical Analysis

Phytochemical analysis of fresh *A. laxiflora* samples utilized various chemicals and reagents to detect the presence of flavonoids, alkaloids, saponins, phenolic compounds and steroids through specific tests and various colour observation qualitatively using (Akinpelu et al., 2015) method.

Some In-vitro Antioxidant Analyses of the Leaf Sample

The DPPH free radical scavenging activity was assessed at various doses using absorbance measurements at 516 nm. The ferric reducing antioxidant power (FRAP) test consisted of combining the extract with reagents and measuring absorbance at 700 nm. The ferric reducing antioxidant power (FRAP) test consisted of combining the extract with reagents and measuring absorbance at 700 nm. Total phenolic content was

determined using the Folin-Ciocalteu reagent and sodium carbonate, with absorbance measured at 700 nm, as reported by Gupta et al. (2016) and Marinova and Batchvarov (2011). Vitamin C concentration was evaluated by converting ascorbic acid to dehydroascorbic acid and measuring absorbance at 520 nm, as described by Abeysuriya et al. (2020). Olatunde Farombi et al. (2003) proposed a technique for analyzing flavonoids, vitamin C, and phenolic compounds.

Green Synthesis and Anti-microbial Analyses

The green production of nanoparticles and antimicrobial analyses utilizing *A. laxiflora* aqueous extract were determined and characterized using UV-visible, FTIR spectroscopies, and antimicrobial assays, as described by (Jadoun et al., 2021).

Toxicological Analysis

Wistar rats were used for the acute toxicity test with an aqueous extract of *A. laxiflora*. To assess the extract's impact on the rats' behavior, 200 mg/kg and 400 mg/kg doses were administered. The liver marker enzymes, alkaline phosphatase (ALP), aspartate aminotransferase (AST), and alanine aminotransferase (ALT), were measured and examined from the blood using the procedures outlined by Reitman and Frankel (1957) and Wright et al. (1972).

Antimicrobial analysis: The antimicrobial analysis was carried out on the extract as well as the synthesized metallic nanoparticles. The bacteria (*Alcaligen odorance*, *Pseudomonas syringiae*, *Streptococcus faecalis*, *Salmonella typhi* and *Enterobacter aerogenes* fungus (*Fusarium*) and *oxysporium*) were used to determine the antimicrobial properties. The procedures involved preparing and standardizing bacterial inoculum, followed by testing the extract and nanoparticles efficacy against bacterial isolates using agar well diffusion assays to measure zones of inhibition.

Results

Proximate and Antinutrient Composition

The proximate analysis of *A. laxiflora* leaf powder revealed a substantial nutritional profile. The moisture content was determined to be $9.26 \pm 0.01\%$. The ash content was notably high at $13.04 \pm 0.01\%$, indicative of a rich mineral composition. The plant material contained considerable amounts of crude fat ($11.77 \pm 0.01\%$), crude fiber ($17.65 \pm 0.005\%$), and crude protein ($24.06 \pm 0.01\%$). Carbohydrates, calculated by difference, accounted for $24.22 \pm 0.01\%$ of the composition.

The antinutrient analysis demonstrated the presence of various compounds at moderate concentrations (Table 1).

Table 1: Antinutrient composition of *Alchornea laxiflora*

Parameters	Values
Phytate (%)	0.49±0.03
Alkaloids (mg/g)	6.00±0.00
Oxalate (mg/g)	1.09±0.01
Tannin (mg/L)	17.90±0.01
Trypsin inhibitor (%)	36.16±0.89

Mineral and Phytochemical Composition

The mineral composition analysis confirmed the presence of essential macro and trace elements (Table 2). Magnesium was the most abundant mineral ($38.45 \pm 0.05\%$), followed by phosphorus, calcium, and potassium. Trace amounts of manganese, chromium, and lead were present.

Table 2: Mineral composition of Lowveld bead string (*Alchornea laxiflora*)

Parameters	Values
P	9.98±0.03
Fe	0.94±0.001
Ca	5.16±0.01
Cu	0.31±0.001
Cr	0.09±0.001
Mg	38.45±0.05
K	4.07±0.01
Na	2.46±0.01
Mn	0.10±0.00
Pb	0.06±0.00
Zn	1.93±0.002

Qualitative phytochemical screening of the aqueous leaf extract confirmed the presence of a diverse array of secondary metabolites, including saponins, phenols, tannins, flavonoids, alkaloids, terpenoids, steroids, glycosides, and phlobatannins.

Quantitative assessment of key antioxidant compounds revealed high levels of vitamin C (200.00 ± 0.00 mg/100g), flavonoids ($14.00 \pm 0.00\%$), and phenolic compounds (10.05 ± 0.11 mg GAE/g).

Antioxidant Activity Assays

The aqueous extract exhibited significant in vitro antioxidant potential across multiple assays. It demonstrated strong free radical scavenging activity, with a DPPH inhibition rate of $79.03 \pm 0.12\%$. The Ferric Reducing Antioxidant Power (FRAP) was equivalent to 25.53 ± 0.09 mg of Vitamin C per gram of sample. Furthermore, the extract showed nitric oxide (NO) scavenging activity of $42.17 \pm 0.30\%$ and a low Thiobarbituric Acid Reactive Substances (TBARS) value of 0.04 ± 0.00 mg MDA/g, indicating potent inhibition of lipid peroxidation.

Antimicrobial Activity

The aqueous leaf extract and its metallic nanoparticles (AgNP, CuNP, ZnNP) displayed selective antimicrobial activity (Table 3). The plain leaf extract showed inhibition only against *Pseudomonas syringae* (3.0 mm zone of inhibition). In contrast, the nanoparticle formulations exhibited enhanced and broader activity. AgNP was effective against *Streptococcus faecalis* and *Enterobacter aerogenes*. CuNP and ZnNP showed activity against all tested bacterial strains to varying degrees, with ZnNP demonstrating the broadest spectrum of inhibition.

The samples also exhibited antifungal activity against *Fusarium oxysporium*. The leaf extract and CuNP showed the highest mycelial growth inhibition (55.56%), followed by ZnNP (48.15%) and AgNP (44.44%).

Table 3: Antibacterial potential properties of Lowveld bead string (*Alchornea laxiflora*) aqueous leaf extract and its nanoparticles

Samples	Antibacterial potential (Zone of inhibition; mm)				
	<i>Alcaligen odorance</i>	<i>Pseudomonas syringiae</i>	<i>Streptococcus faecalis</i>	<i>Salmonella typhi</i>	<i>Enterobacter aerogenes</i>
<i>A. laxiflora</i> leaf extract	0	3	0	0	0
<i>A. laxiflora</i> AgNP	2	0	14	0	5
CuNPs of <i>A. laxiflora</i>	4	0	2	0	2
ZnNP of <i>A. laxiflora</i>	2	10	5	7	3

Toxicological and Hematological Assessment

Sub-acute administration of the aqueous extract at 200 mg and 400 mg doses resulted in a dose-dependent influence on liver enzyme markers (Table 4). Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) levels showed a slight increase, while alkaline phosphatase (ALP) activity rose more markedly from 50.75 ± 0.58 U/L to 68.06 ± 0.00 U/L at the higher dose.

Table 4: Sub-acute toxicity of liver maker enzymes of *Alchornea laxiflora*

Parameters	Aqueous extract at 200 mg	Aqueous extract at 400 mg
AST (U/L)	43.48 ± 0.16	45.71 ± 1.04
ALT (U/L)	44.70 ± 1.06	47.20 ± 0.18
ALP (U/L)	50.75 ± 0.58	68.06 ± 0.00

Hematological analysis revealed modest, dose-dependent changes in blood parameters. Hemoglobin (HB) concentration increased from 5.90 ± 1.30 U/L to 9.10 ± 0.80 U/L. White blood cell (WBC) and red blood cell (RBC) counts remained relatively stable. Differential white blood cell counts indicated decreases in monocytes, eosinophils, and basophils at the higher 400 mg dose (Table 5).

Table 5: White blood count on aqueous extract of lowveld bead string (*Alchornea laxiflora*)

Parameters	Aqueous extract at 200 mg	Aqueous extract at 400 mg
N (U/L)	61.50 ± 1.50	62.00 ± 1.00
L (U/L)	36.50 ± 4.00	32.00 ± 2.00
M (U/L)	8.00 ± 4.00	3.50 ± 0.50
E (U/L)	3.00 ± 0.00	1.00 ± 0.00
B (U/L)	3.00 ± 2.00	2.00 ± 1.00

UV-Vis and FTIR Spectroscopy Characterization

UV-Vis spectroscopic analysis was employed to confirm the formation of metallic nanoparticles (NPs) and characterize the phytochemical profile of the aqueous leaf extract. The extract exhibited characteristic absorption peaks at approximately 264.5, 285.0, and 328.5 nm (Figure 1), which are typical for phenolic compounds, flavonoids, and other aromatic phytoconstituents.

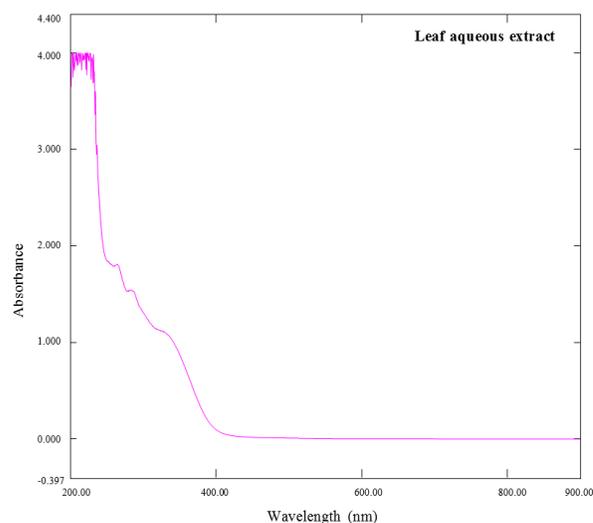


Figure 1: UV-Vis spectrum of the aqueous leaf extract of *Alchornea laxiflora*.

Characteristic absorption maxima at 264.5, 285.0, and 328.5 nm are indicative of polyphenolic compounds, flavonoids, and other aromatic constituents present in the extract.

The successful biosynthesis of nanoparticles was evidenced by noticeable shifts in the absorption profiles of the metal NPs compared to the plain extract. The silver nanoparticles (AgNPs) showed a prominent absorption maximum at 327 nm, a hallmark of the surface plasmon resonance (SPR) characteristic of metallic AgNPs. The copper (CuNPs) and zinc (ZnNPs) nanoparticles also displayed distinct spectral patterns, confirming their formation (Figure 2).

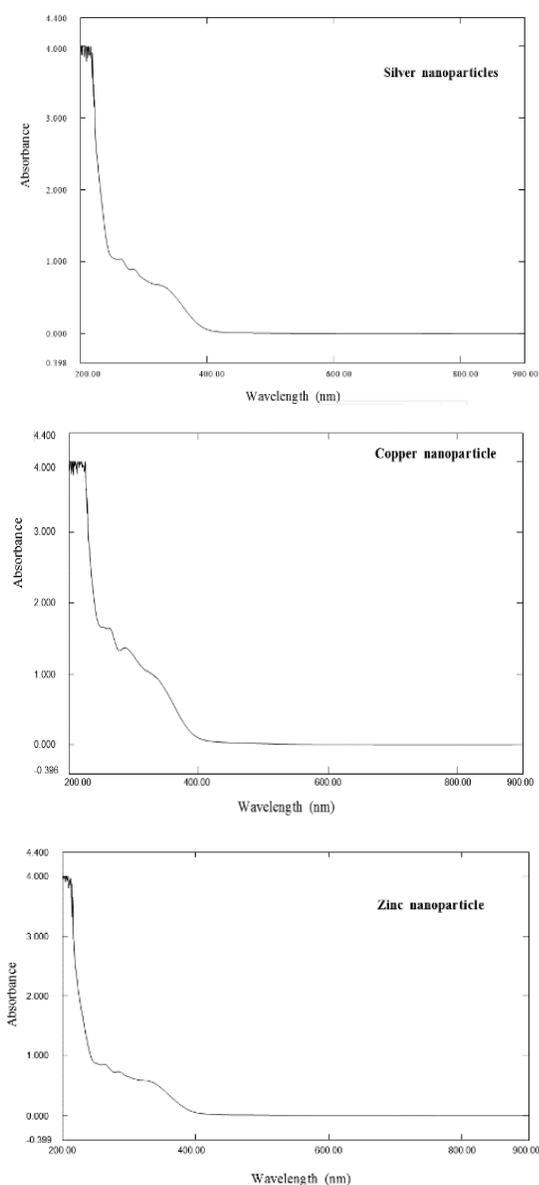


Figure 2: UV-Vis spectrum of the biosynthesized silver nanoparticles (AgNPs).

The prominent absorption peak observed at 327 nm is a definitive signature of the surface plasmon resonance (SPR) phenomenon, confirming the successful formation of spherical silver nanoparticles.

FTIR spectroscopy was used to identify the functional groups in the plant extract responsible for the reduction and stabilization of the nanoparticles. The spectrum of the pure extract (Figure 5a) revealed major absorption bands indicative of O-H stretching ($\sim 3250\text{ cm}^{-1}$), C-H stretching ($\sim 2832\text{ cm}^{-1}$), amide I C=O stretching ($\sim 1636\text{ cm}^{-1}$), and C-O stretching ($\sim 1207\text{ cm}^{-1}$), confirming the presence of phenols, proteins, and other organic compounds. Critical shifts and changes in the intensity of these peaks in the spectra of the nanoparticles (e.g., AgNPs, Figure 6a) clearly demonstrated the involvement of these biomolecules in the capping and stabilization of the synthesized nanoparticles.

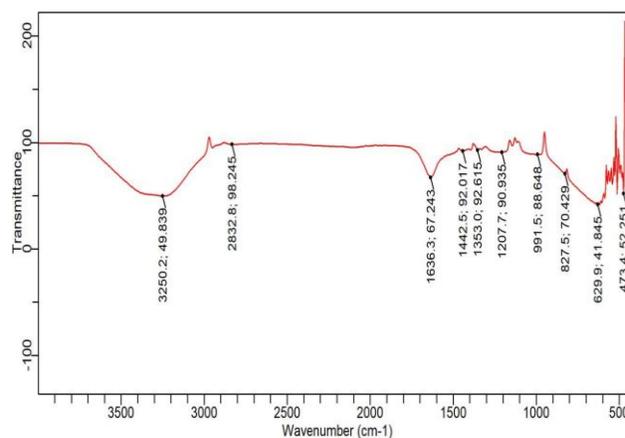


Figure 3: FTIR spectrum of the aqueous leaf extract of *Achornea laxiflora*. Key functional group assignments include: a broad band at $\sim 3250\text{ cm}^{-1}$ (O-H stretching of phenols/alcohols), a peak at $\sim 1636\text{ cm}^{-1}$ (C=O stretching of amide I in proteins), and a band at $\sim 1207\text{ cm}^{-1}$ (C-O stretching), identifying biomolecules responsible for metal ion reduction and nanoparticle capping.

Discussion

The comprehensive phytochemical profiling conducted in this study establishes *A. laxiflora* as a promising botanical species with significant nutritional and therapeutic potential. The findings are discussed in the context of existing literature to elucidate their implications.

The proximate composition analysis reveals a moisture content of 9.26%, which is higher than the 5.7% reported for *Aspilia kotschyi* (Adamu et al., 2017), suggesting potentially enhanced shelf stability and reduced susceptibility to microbial spoilage. The notably high ash content (13.04%), exceeding the 10.32% found in *Acalypha hispida* (Iniaghe et al., 2009), signifies a rich reservoir of inorganic minerals, a claim robustly supported by the mineral composition data. The substantial crude protein content (24.06%) surpasses that of renowned nutritional sources like *Moringa oleifera*

(17.01%) as reported by Ogbe and Affiku (2021), highlighting its value as a plant-based protein source. Furthermore, the favorable crude fat content (11.77%), significantly higher than the 0.83% in *Aspilia kotschyi* (Adamu et al., 2017), indicates a valuable source of energy and essential fatty acids. This nutritional profile is complemented by a considerable crude fiber content (17.65%), known to promote digestive health, and a carbohydrate content (24.22%) comparable to that of *Carica papaya* (23.34%) documented by Kanadi et al. (2021).

The presence of antinutritional factors must be considered for safe consumption. The oxalate content (1.09 mg/g) is relatively low for leafy plants, especially when compared to high-oxalate plants like spinach (*Spinacia oleracea*), which is known to bind calcium and potentially contribute to nephrolithiasis (Nayagam and Rajan, 2021). Similarly, the tannin level (17.9 mg/L) is lower than the 43 mg/g reported for lotus leaves (Häring et al., 1999), which may aid in controlling inflammation and improving digestibility. The phytate content (0.49%) is moderate relative to cereal grains like maize (*Zea mays*), which can contain up to 1.2% (Coulibaly et al., 2011). While the alkaloid content (6 mg/g) is higher than that reported for pre-flowering *Celosia argentea* (Adegbaju et al., 2019) and the trypsin inhibitor activity (36.16%) is considerably higher than levels in sweet potato (Zhang and Corke, 2001), these concentrations are unlikely to pose significant health risks if the plant is consumed in a balanced diet or processed appropriately to mitigate their impact on mineral absorption and protein digestibility.

The mineral analysis demonstrates that magnesium is the most abundant element (38.45 mg/100g), followed by phosphorus (9.98 mg/100g). These values are substantially higher than those reported for *Solanum melongena* (2.80 mg/100g for both) by Edeke et al. (2021). Magnesium is crucial for numerous biochemical reactions, including energy production and neuromuscular regulation, while phosphorus is vital for bone formation and energy metabolism. The calcium content (5.16 mg/100g), though lower than the 28.9 mg/100g found in *Brassica oleracea* (Ogbede et al., 2015), remains essential for bone health. The levels of potassium (4.07 mg/100g) and sodium (2.46 mg/100g), while lower than those in *Alchornea cordifolia* (Nwaoguikpe et al., 2013), are significant for maintaining fluid balance, nerve function, and muscle contraction. Essential trace elements such as zinc (1.93 mg/100g), iron (0.94 mg/100g), and copper (0.31 mg/100g), present in amounts relative to *Solanum aethiopicum* (Edeke et al., 2021), play critical roles in enzymatic catalysis and immune function. The presence of chromium and manganese, comparable to levels in *Brassica*

oleracea (Ogbede et al., 2015), is important for metabolism and bone formation. The detection of lead (0.06 mg/100g), albeit minimal, warrants attention due to its inherent toxicity.

The qualitative phytochemical screening confirmed the presence of a diverse spectrum of bioactive compounds, including saponins, phenols, tannins, flavonoids, alkaloids, terpenoids, steroids, glycosides, and phlobatannins, consistent with previous findings for this species (Akinpelu et al., 2015). These compounds are associated with a wide range of pharmacological properties, such as antioxidant, anti-inflammatory, antimicrobial, and cholesterol-lowering effects. Quantitative analysis revealed an exceptionally high vitamin C content (200 mg/100g), a potent antioxidant, and significant levels of phenolic compounds (10.05 mg GAE/g), albeit lower than those reported for *Salvia officinalis* by Atanassova et al. (2011). These compounds are directly responsible for the observed robust antioxidant activity.

The antioxidant capacity of *A. laxiflora*, as evaluated through multiple assays, is remarkable. The DPPH radical scavenging activity (79.03%) is comparable to the potent activities documented for green tea (*Camellia sinensis*) (Novilla et al., 2022) and *Rosmarinus officinalis* (Rašković et al., 2014), indicating a strong ability to neutralize free radicals. The low TBARS value (0.04 mg MDA/g), similar to that of turmeric (*Curcuma longa*) (Aminuddin et al., 2023), denotes a powerful capacity to inhibit lipid peroxidation, thereby protecting cellular membranes. The significant FRAP value (25.53 mg Vit. C/g) and nitric oxide scavenging activity (42.17%) are on par with standard antioxidants like vitamins C and E (Ahmed et al., 2013), underscoring a multifaceted antioxidant mechanism capable of mitigating oxidative and nitrosative stress.

A particularly fascinating finding is the dramatic enhancement of antimicrobial activity upon the green synthesis of metallic nanoparticles (AgNP, CuNP, ZnNP) using the plant extract. While the plain aqueous extract showed limited antibacterial efficacy, the nanoparticles exhibited broad-spectrum activity against Gram-negative bacteria, including *Pseudomonas syringae*, *Streptococcus faecalis*, and *Enterobacter aerogenes*. This synergy can be attributed to the well-documented mechanisms of nanoparticle action, such as reactive oxygen species (ROS) generation, disruption of microbial cell membranes, and release of toxic metal ions, to which Gram-negative bacteria are particularly susceptible. The significant antifungal inhibition against *Fusarium oxysporium*, particularly by AgNP (55.56%), aligns with previous studies (Akinpelu et al., 2015; Tchinda et al., 2017) and highlights its potential as a natural antifungal agent. The variation in efficacy between different metal nanoparticles underscores the importance of the core metal in

determining antimicrobial specificity and potency. The sub-acute toxicity assessment indicates a dose-dependent influence on biochemical parameters. The elevation in liver marker enzymes, particularly the notable increase in ALP (from 50.75 U/L to 68.06 U/L) at the higher dose, suggests a potential hepatotropic effect, warranting caution and further investigation to establish a safe dosage threshold, a concern similarly reported for *Annona senegalensis* (Okoye et al., 2012). The hematological analysis revealed bioactive effects on the hematopoietic system. The significant increase in hemoglobin concentration and alterations in differential white blood cell counts, including decreases in monocytes, eosinophils, and basophils at the higher dose, are indicative of physiological modulation. These changes, showing trends similar to those reported by Alelign et al. (2020), while not severely adverse at the tested doses, emphasize the necessity for detailed long-term toxicological studies to fully ascertain the plant's safety profile for therapeutic or nutritional applications.

The UV-Vis and FTIR spectroscopic analyses provide critical evidence for the successful formation and stabilization of nanoparticles. The distinct absorption profiles in the UV-Vis spectra (Figures 1-4) are characteristic of surface plasmon resonance, confirming the synthesis of metallic nanoparticles (Ag, Cu, Zn), a phenomenon well-documented in the literature (Suresh et al., 2023; Baishya et al., 2023; Neamah et al., 2023; Donmez and Keyvan, 2023). The FTIR spectra (Figures 5-8) identified key functional groups (e.g., from polyphenols, proteins) involved in the reduction of metal ions and capping of the nanoparticles, thereby preventing aggregation and enhancing stability (Joseph et al., 2024). These modifications, facilitated by the inherent phytochemicals of *A. laxiflora*, are consistent with mechanisms that enhance antibacterial efficacy by increasing the effective surface area for microbial interaction and disruption (Hembram et al., 2018, 2023; Sabapathi et al., 2023). This green synthesis approach proves to be an efficient and eco-friendly method for producing bioactive nanoparticles.

Conclusion

This study systematically characterizes *A. laxiflora* as a rich source of nutrients, essential minerals, and diverse bioactive compounds with potent antioxidant properties. The significant enhancement of its inherent antimicrobial activity through the green synthesis of metallic nanoparticles presents a promising avenue for developing novel therapeutic agents. However, the observed dose-dependent effects on liver enzymes and hematological parameters highlight the importance of conducting further comprehensive toxicological studies to ensure its safe utilization in

dietary formulations and phytomedicine. Future work should focus on isolating specific active compounds, elucidating their exact mechanisms of action, and conducting in vivo studies to validate these promising in vitro findings.

Declarations

Conflict of interest

The authors declare there is no competing interests.

Acknowledgement

A lot of thanks are given to all member staff of department of Pharmaceutics, faculty of pharmacy, National Ribat University and Medicinal and Aromatic Plants & Traditional Medicine Research Institute, National Center for Research, Sudan.

Consent for publications

The authors gave approval for the publication of the manuscript.

Funding support

This study was not funded by any organization.

Authors' contributions

Conceptualization: Oseni Olatunde A. and Adebayo Lovet O.

Data curation: All authors were involved in data curation.

Formal analyses: All authors were involved in analyses of the sample.

Funding acquisition: All authors were involved in funding of the research.

Investigation: All Authors. Methodology: All Authors involved.

Project administration: Oseni Olatunde A., Adewumi Funmilayo. A. and Adebayo Lovet O.

Resources: All Authors involved.

Software: Oseni Olatunde A., Adewumi Funmilayo. A. and Adebayo Lovet O.

Supervision: Oseni Olatunde A.

Validation: All Authors validated.

Visualization: Adebayo Lovet O.

Writing-original draft: Oseni Olatunde A., Adewumi Funmilayo. A. and Adebayo Lovet O.

Writing-review & editing: All Authors.

Ethical considerations

Ethical issues (including plagiarism, misconduct, data fabrication, falsification, double publication or submission, redundancy) have been completely observed by the authors.

Reference

- Abeyasuriya HI, Bulugahapitiya VP, Loku Pulukkuttige J. Total vitamin C, ascorbic acid, dehydroascorbic acid, antioxidant properties, and iron content of underutilized and commonly consumed fruits in Sri Lanka. *International Journal of Food Science*. 2020; 2020(1):4783029. DOI: 10.1155/2020/4783029.
- Adamu AU, Paul ED, Gimba CE, Ndukwe IG. Phytochemical and proximate analysis of *Aspilia kotschyi* (Sch. bipex, hochst) Oliv. *Nigerian Journal of Technology*. 2017; 36(4):1135-7. DOI: 10.4314/njt.364.1482.
- Adegbaolu OD, Otunola GA, Afolayan AJ. Proximate, mineral, vitamin and anti-nutrient content of *Celosia argentea* at three stages of maturity. *South African Journal of Botany*. 2019; (1):124:372-9. DOI: 10.1016/j.sajb.2019.05.036.
- Ahmed D, Zara S, Baig H. In vitro analysis of antioxidant activities of *Oxalis corniculata* Linn. fractions in various solvents. *African Journal of Traditional, Complementary and Alternative Medicines*. 2013; 10(1):158-65.
- Akinpelu DA, Abioye EO, Aiyegoro OA, Akinpelu OF, Okoh AI. Evaluation of antibacterial and antifungal properties of *Alchornea laxiflora* (Benth.) Pax. & Hoffman. *Evidence-Based Complementary and Alternative Medicine*. 2015; (1): 684839. DOI: 10.1155/2015/684839.
- Alelign T, Chalchisa D, Fekadu N, Solomon D, Sisay T, Debella A, Petros B. Evaluation of acute and sub-acute toxicity of selected traditional antiurolithiatic medicinal plant extracts in Wistar albino rats. *Toxicology Reports*. 2020; (7):1356-65. DOI: 10.1016/j.toxrep.2020.10.001.
- Aminuddin M, Sargowo D, Sardjono TW., Widjiati W. *Curcuma longa* supplementation reduces MDA, TNF- α , and IL-6 levels in a rat model exposed to soot particulates. *Open Veterinary Journal*. 2023; 13(1): 11–19. DOI: 10.5455/OVJ.2023.v13.i1.2.
- Arina N, Wikan M, Dwi D. Antioxidant activities of green tea (*Camellia Sinensis* L.) leaves from Ciwidey, West Java. in *The 4th International Seminar on Global Health, KnE Medicine*, 2022; DOI: 10.18502/kme.v2i2.11077.
- Atanassova M, Georgieva S, Ivancheva K. Total phenolic and total flavonoid contents, antioxidant capacity and biological contaminants in medicinal herbs. *Journal of the University of Chemical Technology & Metallurgy*. 2011;46(1).
- Baishya T, Das P, Ashraf GJ, Dua TK, Paul P, Nandi G, et al. Preparation of silver nanoparticles by *Osbeckia stellata* aqueous extract via green synthesis approach: Characterization and assessment of their antioxidant, antidiabetic, cytotoxicity, and antibacterial properties. *Biotechnology and Applied Biochemistry*. 2023; 70(6):2097-107. DOI: 10.1007/s13399-023-03743-7.
- Coulibaly A, Kouakou, B, Chen, J. Phytic acid in cereal grains: structure, healthy or harmful ways to reduce phytic acid in cereal grains and their effects on nutritional quality. *American Journal of Plant Nutrition and Fertilization Technology*. 2011; 1(1): 1–22. DOI: 10.3923/ajpnft.2011.1.22.
- Donmez S, Keyvan E. Green synthesis of zinc oxide nanoparticles using grape seed extract and evaluation of their antibacterial and antioxidant activities. *Inorganic and Nano-Metal Chemistry*. 2023; 4:1-8.
- Edeke A, Uchendu N, Omeje K, Odiba AS. Nutritional and pharmacological potentials of *solanum melongena* and *solanum aethiopicum* fruits. *The Journal of Phytopharmacology*. 2021; 10(1):61-7. DOI: 10.31254/phyto.2021.10112.
- Gupta M, Karmakar N, Sasmal S, Chowdhury S, Biswas S. Free radical scavenging activity of aqueous and alcoholic extracts of *Glycyrrhiza glabra* Linn. measured by ferric reducing antioxidant power (FRAP), ABTS bleaching assay (α TEAC), DPPH assay and peroxy radical antioxidant assay. *International Journal of Pharmacology and Toxicology*. 2016; 4:235-40. DOI: 10.14419/ijpt.v4i2.6578.
- Häring DA, Suter D, Amrhein N, Lüscher A. Biomass allocation is an important determinant of the tannin concentration in growing plants. *Ann Bot*. 2007; 99(1): 111-120. DOI:10.1093/aob/mcl227.
- Hembram KC, Kumar R, Kandha L, Parhi, PK, Kundu CN, Bindhani BK. Therapeutic prospective of plant-induced silver nanoparticles: application as antimicrobial and anticancer agent. *Artificial Cells, Nanomedicine, and Biotechnology*. 2018; 46(sup3): S38–S51. DOI: 10.1080/21691401.2018.1489262.
- Hembram M, Sahu JR, Swain J, Soren S, Kumari N, Panda J, et al. Synthesis, characterisation and structural determination of pyridyl based Ni (II) complex for microbial study and biosensing of DNA. In *AIP Conference Proceedings*. 2023.
- Iniaghe OM, Malomo, SO, and Adebayo, JO. Proximate composition and phytochemical constituents of leaves of some *Acalypha species*. *Pakistan Journal of Nutrition*. 2009; 8: 256–8. DOI: 10.3923/pjn.2009.256.258.
- Jadoun S, Arif R, Jangid NK, Meena RK. Green synthesis of nanoparticles using plant extracts: A review. *Environmental Chemistry Letters*. 2021;19(1):355-74. DOI: 10.1007/s10311-020-01074-x.

- Jain NK, Tailang M, Kumar S. Appraising the therapeutical potentials of *Alchornea laxiflora* (Benth.) Pax & K. Hoffm., an underexplored medicinal herb: A systematic review. *Frontiers in Pharmacology*. 2022; 13: 958453. DOI: 10.3389/fphar.2022.958453.
- Joseph DA, Oseni MO, Oseni OA. Biochemical investigations and green synthesis characterization using aqueous extract of *Ageratum conyzoides* L. leaf. *Journal of Biochemicals and Phytomedicine*. 2024; 3(2): 9-19. DOI: 10.34172/jbp.2024.15.
- Kanadi MA, Yila R, Ibrahim MP, Yaradua AI, Nasir A. Proximate composition and phytochemical constituents of matured *Carica papaya* seed extracts. *Asian Journal of Research in Biochemistry*. 2021; 9(1): 28-33. DOI: 10.9734/ajrb/2021/v9i130193.
- Marinova G, Batchvarov V. Evaluation of the methods for determination of the free radical scavenging activity by DPPH. *Bulgarian Journal of Agricultural Science*. 2011; 17(1):11-24.
- Nayagam JR, Rajan R. Calcium oxalate crystals as raw food antinutrient: a review. *Journal of Pharmaceutical Research International*. 2021; 33:295-301. DOI: 10.9734/jpri/2021/v33i41B32368.
- Neamah SA, Albukhaty S, Falih IQ, Dewir YH, Mahood HB. Biosynthesis of zinc oxide nanoparticles using *Capparis spinosa* L. fruit extract: Characterization, biocompatibility, and antioxidant activity. *Applied Sciences*. 2023; 9;13(11):6604. DOI: 10.3390/app13116604.
- Nwaoguikpe RN, Braide W, Okwu GN, Anyanwu BO, Ujowundu CO. The effects of extracts of christmas bush (*Alchornea cordifolia*) on sickle cell hemoglobin. *Journal of Pharmaceutical Research International*. 2013; 4(1): 158-71. DOI: 10.9734/BJPR/2014/4467.
- Ogbe A, Affiku JP. Proximate study, mineral and anti-nutrient composition of moringa oleifera leaves harvested from lafia, nigeria: potential benefits in poultry nutrition and health. *Journal of Microbiology, Biotechnology and Food Sciences*. 2021; 1(3): 296-308.
- Ogbede SC, Saidu AN, Kabiru AY, Busari MB. Nutrient and anti-nutrient compositions of *Brassica Oleraceae* Var. Capitata L. *IOSR Journal of Pharmacy*. 2015; 5(3):19-25.
- Okoye TC, Akah PA, Ezike AC, Okoye MO, Onyeto CA, Ndukwu F, et al. Evaluation of the acute and sub acute toxicity of *Annona senegalensis* root bark extracts. *Asian Pacific Journal of Tropical Medicine*. 2012; 1;5(4):277-82. DOI: 10.1016/S1995-7645(12)60039-X
- Olajire AA, Adeyeye GO, Yusuf RA. *Alchornea laxiflora* bark extract assisted green synthesis of platinum nanoparticles for oxidative desulphurization of model oil. *Journal of Cluster Science*. 2017; 28(5): 1565-78. DOI:10.1007/s10876-017-1167-3.
- Olajire AA, Ifediora NF, Bello MD, Benson NU. Green synthesis of copper nanoparticles using *Alchornea laxiflora* leaf extract and their catalytic application for oxidative desulphurization of model oil. *Iranian Journal of Science and Technology, Transactions A: Science*. 2018;42:1935-4. DOI: 10.1007/s40995-017-0404-9.
- Olasehinde EF, Ogunjobi JK, Akinlosotu OM. Investigation of the inhibitive properties of *Alchornea laxiflora* leaves on the corrosion of mild steel in HCl: Thermodynamics and kinetic study. *Journal of American Science*. 2015;11(1 S): 32-9.
- Olatunde Farombi E, Ogundipe OO, Samuel Uhunwangho E, Adeyanju MA, Olarenwaju Moody J. Antioxidant properties of extracts from *Alchornea laxiflora* (Benth) Pax and Hoffman. *Phytotherapy research*. 2003;17(7):713-6. DOI: 10.1002/ptr.1050.
- Oloyede GK, Onocha PA, Adaramoye OA, Thonda SE. Hepatoprotective activity and flavonoids of *Alchornea laxiflora* leaf extract. *Research Journal of Phytochemistry*. 2011; 5(3): 190-200. DOI:10.3923/rjphyto.2011.190.200.
- Rašković A, Milanović I, Pavlović N, Čebović T, Vukmirović S, Mikov M. Antioxidant activity of rosemary (*Rosmarinus officinalis* L.) essential oil and its hepatoprotective potential. *BMC Complementary and Alternative Medicine*. 2014; 14:225. DOI: 10.1186/1472-6882-14-225.
- Reitman S, Frankel S. A colorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. *American Journal of Clinical Pathology*. 1957; 28(1): 56-63. DOI: 10.1093/ajcp/28.1.56.
- Sabapathi N, Ramalingam S, Aruljothi KN, Lee J, Barathi S. Characterization and therapeutic applications of biosynthesized silver nanoparticles using *Cassia auriculata* flower extract. *Plants*. 2023;12(4):707. DOI: 10.3390/plants12040707.
- Siwe-Noundou X, Ndinteh DT, Olivier DK, Mnkandhla D, Isaacs M, Muganza FM, et al. Biological activity of plant extracts and isolated compounds from *Alchornea laxiflora*: Anti-HIV, antibacterial and cytotoxicity evaluation. *South African Journal of Botany*. 2019;122:498-503. DOI: 10.1016/j.sajb.2018.08.010.
- Suresh P, Doss A, Praveen Pole RP, Devika M. Green synthesis, characterization and antioxidant activity of bimetallic (Ag-ZnO) nanoparticles using *Capparis zeylanica* leaf extract. *Biomass Conversion and Biorefinery*. 2023; 11:1-9.

Tchinda CF, Voukeng IK, Beng VP, Kuete V. Antibacterial activities of the methanol extracts of *Albizia adianthifolia*, *Alchornea laxiflora*, *Laportea ovalifolia* and three other Cameroonian plants against multi-drug resistant Gram-negative bacteria. Saudi Journal of Biological Sciences. 2017; 24(4): 950-5. doi: 10.1016/j.sjbs.2016.01.0

Wright PJ, Leatherwood PD, Plummer DT. Enzymes in rats: Alkaline phosphatase. Enzymologia. 1972; 42: 317-27. Available from: <https://pubmed.ncbi.nlm.nih.gov/4337755/>.

Zhang Z, Corke H. Trypsin inhibitor activity in vegetative tissue of sweet potato plants and its response to heat treatment. Journal of the Science of Food and Agriculture. 2001; 81(14): 1358-63. doi: 10.1002/jsfa.945.

Copyright © 2024 The Author(s). This is an open-access article distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.