The Journal of Phytopharmacology 2026; 15(2):176-180
Online at: www.phytopharmajournal.com


Research Article

ISSN 2320-480X
JPHYTO 2026; 15(2): 176-180
March- April
Received: 18-11-2025
Accepted: 20-04-2026
Published: 27-04-2026

doi: 10.31254/phyto.2026.15208

**Daniel Akpe-Efiak Ambe**
Department of Pharmacognosy and Natural Medicine, Faculty of Pharmacy, University of Uyo, Uyo, Nigeria

**Joshua Charles Nworondah**
Department of Pharmacognosy and Natural Medicine, Faculty of Pharmacy, University of Uyo, Uyo, Nigeria

**Correspondence:**
**Dr. Daniel Akpe-efiak Ambe**
Department of Pharmacognosy and Natural Medicine, Faculty of Pharmacy, University of Uyo, Uyo, Akwa Ibom State, Nigeria
Email: danielaambe@uniuyo.edu.ng

# Reno-protective effects of *Canavalia ensiformis* (L.) DC. leaf extract in a gentamicin-induced nephrotoxicity model in mice

Daniel Akpe-Efiak Ambe, Joshua Charles Nworondah

## ABSTRACT

**Background:** Drug-induced nephrotoxicity is a leading cause of acute kidney injury, with gentamicin, a commonly prescribed aminoglycoside, known to cause dose-dependent renal impairment. *Canavalia ensiformis* (jack bean) is a leguminous plant rich in phytochemicals with reported antioxidant and cytoprotective properties. **Objective:** This study evaluated the nephroprotective effect of the methanolic leaf extract of *C. ensiformis* against gentamicin-induced renal damage in mice. **Materials and Methods:** Twenty-five male albino mice (25–30 g) were randomised into five groups (n = 5). Group 1 received feed and water (standard control); Group 2 received gentamicin (100 mg/kg/day, i.p.); Groups 3–5 received gentamicin (100 mg/kg/day, i.p.) plus *C. ensiformis* extract (50, 100, or 200 mg/kg/day, p.o.) for 12 days. Serum creatinine, urea, albumin, and total protein were measured, and the kidneys were subjected to histopathological analysis. **Results:** Gentamicin significantly increased serum creatinine (45.80 ± 4.00 µmol/L) and urea (6.62 ± 0.18 mmol/L) compared with the standard control (28.10 ± 2.70 µmol/L; 4.60 ± 0.60 mmol/L, $P < 0.05$). Co-administration of the extract dose-dependently restored renal function, with the 200 mg/kg group showing the most significant improvement (creatinine: 30.20 ± 0.42 µmol/L; urea: 3.45 ± 0.11 mmol/L). Histological findings corroborated biochemical results: gentamicin caused glomerular hypertrophy and tubular degeneration, while extract-treated groups showed progressive restoration of renal cytoarchitecture, with near-normal morphology at 200 mg/kg. **Conclusion:** Methanol leaf extract of *C. ensiformis* protected against gentamicin-induced nephrotoxicity in mice, preserving renal function and architecture in a dose-dependent manner. These findings highlight its potential as a nephroprotective agent and warrant further investigation into its phytochemical and mechanistic properties.

**Keywords:** *C. ensiformis*, Gentamicin, Acute Kidney Injury, Nephrotoxicity, Renal Histopathology, Albino Mice.

## INTRODUCTION

Acute kidney injury (AKI) is a significant global health burden, contributing substantially to morbidity and mortality, particularly in hospitalised patients [1]. Its multifactorial aetiology includes sepsis, ischemia, and drug-induced nephrotoxicity, with the latter accounting for up to one-quarter of all cases [2]. Nephrotoxic drugs often impair renal tubular and glomerular function, leading to disturbances in fluid balance, electrolyte homeostasis, and nitrogenous waste excretion [3].

Gentamicin, a widely used aminoglycoside antibiotic, is effective against a broad spectrum of Gram-negative bacteria but is well recognised for its nephrotoxic potential [4]. Gentamicin accumulates in the renal proximal tubules, where it triggers oxidative stress, mitochondrial dysfunction, lipid peroxidation, and inflammatory responses, culminating in tubular necrosis and impaired glomerular filtration [5]. Despite advances in supportive therapy, there is currently no established pharmacological intervention to prevent gentamicin-induced nephrotoxicity, making the search for novel protective agents' imperative.

Medicinal plants have long been explored as sources of nephroprotective compounds [6] due to their rich phytochemical profiles, including flavonoids, phenolics, saponins, and alkaloids, which exhibit antioxidant and anti-inflammatory properties [7]. *C. ensiformis* (Fabaceae), commonly called

jack bean, a leguminous plant traditionally used in African ethnomedicine, has been reported to contain bioactive constituents such as flavonoids, tannins, and saponins [8]. While the plant has been studied for nutritional and antimicrobial activities, its nephroprotective potential has not been systematically evaluated.

Given the central role of oxidative stress in gentamicin-induced kidney injury and the phytochemical richness of *C. ensiformis*, its extract may possess renoprotective properties. Therefore, the present study investigated the protective effects of the methanol leaf extract of *C. ensiformis* on gentamicin-induced nephrotoxicity in mice, using biochemical and histopathological endpoints to assess renal function and architecture.

## MATERIAL AND METHODS

### Chemicals, Reagents and Equipment

All chemicals and reagents used were of analytical grade and obtained from reputable suppliers in India. Methanol (Lobachem, India; 99.5%, AR grade), 10% buffered formalin prepared from formaldehyde solution (37–40%), gentian violet (96%), hematoxylin (95%), and eosin (90%) were used for the study. The equipment used included a light microscope (Primostar 415550-1501-000, Zeiss, USA), microtome (AM-2268 ARI), analytical balance (Ohaus PR423/E-420GX), automated tissue processor (Leica ASP300/ASP300 S), and pH meter (Hellog PH-3C).

### Plant collection and identification

Fresh leaves of *C. ensiformis* (1 kg) were collected in February 2023 from Uyo Local Government Area, Akwa Ibom State, Nigeria. Botanical identification was performed by Prof. Henry Akinibosun, Department of Plant Biology and Biotechnology, Faculty of Life Sciences, University of Benin, Benin City, Edo State. A voucher specimen (UBH-C448.3.2) was deposited at the departmental herbarium. The leaves were shade-dried for 12 days, foreign matter removed, and pulverised using a mortar and pestle.

### Extraction

A 500 g portion of the powdered plant was extracted with absolute methanol in a Soxhlet extractor at 67 °C. The methanolic extract of *C. ensiformis* leaves (CEME) was concentrated under reduced pressure using a rotary evaporator, weighed, stored in a labelled bottle, and refrigerated at 4 °C until use [9].

### Ethical approval

Ethical approval was obtained from the Akwa Ibom State Ministry of Health Ethics Committee (Approval no. AKHREC/11/07/25/350). All experimental procedures adhered to the National Institutes of Health Guide for the Care and Use of Laboratory Animals (NIH Publication No. 85–23, revised 1996).

### Experimental design

The animal experiment was conducted in the Animal House, Faculty of Basic Medical Sciences, University of Uyo, Nigeria. The experimental protocol was carried out according to the method described by Rizwana *et al*. [10], with slight modifications to suit the conditions of the present study. Twenty-five inbred male albino mice (25–30 g) were procured from the Animal House, Faculty of Basic Medical Sciences, University of Uyo, Nigeria. Animals were housed under standard conditions (12 h light/dark cycle, 25 ± 2 °C) with free access to feed and water.

Mice were randomly assigned to five groups (n = 5):

- **Group 1 (Normal control):** Received feed and water only.
- **Group 2 (Sham control):** Received gentamicin (100 mg/kg/day, i.p.).
- **Group 3:** Received gentamicin (100 mg/kg/day, i.p.) + extract (50 mg/kg/day, p.o.).
- **Group 4:** Received gentamicin (100 mg/kg/day, i.p.) + extract (100 mg/kg/day, p.o.).
- **Group 5:** Received gentamicin (100 mg/kg/day, i.p.) + extract (200 mg/kg/day, p.o.).

All treatments were administered daily for 12 days. Body weights were recorded at baseline and before sacrifice. On day 13, animals were anaesthetised, sacrificed, and blood samples collected via cardiac puncture. Kidneys were excised and processed for histological analysis [10].

### Blood collection and serum separation

Blood samples were allowed to clot and centrifuged at 3000 rpm for 5 min. The serum was collected with Pasteur pipettes and stored at 4 °C for biochemical analysis [11].

### Biochemical analysis

Commercial diagnostic kits were used to assess renal function biomarkers. Standard reagent kits were used to measure the amounts of potassium and sodium in the serum. The alkaline picrate method was used to quantify creatinine, and the diacetyl monoxime method was used to determine urea concentration in accordance with the Randox Laboratories assay procedure [12]. The concentrations of total protein and albumin in the serum were quantified following the standard procedures outlined by Tietz. [13].

### Statistical analysis

Data were expressed as mean ± SEM. Statistical significance was determined using one-way ANOVA followed by Student's t-test for pairwise comparisons at a 95% confidence interval (GraphPad Prism version 6.01). A value of $P < 0.05$ was considered statistically significant.

## RESULTS

### The effects of CEME on kidney function

The effects of *C. ensiformis* extract on renal biochemical indices in mice are shown in Table 1. Serum creatinine levels in the treated groups (50–200 mg/kg) displayed non-significant dose reduction levels, Urea levels were notably lower at 100 and 200 mg/kg compared to the standard control, with the sham control having the highest value. Albumin levels remained relatively consistent across all groups with no significant changes. Total protein levels were significantly reduced at 200 mg/kg, whereas the sham control had the highest level. Overall, the administration of CEME reduced the changes observed in the sham group, particularly regarding urea, suggesting a possible nephroprotective effect.

### Effects of CEME on the renal histo-architecture

Histological evaluation of the kidney sections demonstrated distinct differences across treatment groups (Figures 1–5). The control group exhibited normal renal cytoarchitecture with well-defined glomeruli, Bowman's capsule, and renal tubules (Figure 1). Administration of gentamicin alone (100 mg/kg) produced marked renal injury characterized by hypertrophied glomeruli, obliteration of Bowman's space, and widespread glomerular degeneration (Figure 2). Co-treatment with gentamicin and the plant extract showed dose-dependent nephroprotective effects. At 50 mg/kg of extract (Figure 3), partial improvements in the renal structure were observed, including the presence of intact glomeruli and Bowman's capsules, although some glomeruli remained degenerated. Mice treated with 100 mg/kg of extract (Figure 4) displayed further histological improvements,

with most renal corpuscles appearing normal and only a few showings evidence of atrophy. Notably, the highest extract dose (200 mg/kg) exhibited the highest protection, restoring renal histo-architecture close to normal with distinct glomeruli, Bowman's capsule, and Bowman's space (Figure 5).

**Table 1:** Effects of CEME on the kidney biochemical parameters in mice

| Treatment group | Creatinine (umol/L) | Urea (mmol/L) | Albumin (µmol/L) | Total protein (g/L) |
|---|---|---|---|---|
| Standard control | 24.00 ± 2.30 | 4.60 ± 0.60 | 12.70 ± 0.85 | 28.10 ± 2.70 |
| 50 mg/kg | 25.60 ± 2.11 | 4.88 ± 0.63 | 12.70 ± 0.59 | 25.20 ± 5.33 |
| 100 mg/kg | 23.00 ± 1.19 | 3.85 ± 0.26 | 13.40 ± 0.50 | 30.20 ± 0.54 |
| 200 mg/Kg | 22.70 ± 0.96 | 3.45 ± 0.11 | 12.50 ± 0.75 | 26.90 ± 0.28 |
| Sham control | 28.30 ± 2.30 | 6.62 ± 0.18 | 13.70 ± 0.28 | 39.90 ± 3.28 |

Data are expressed as mean ± SEM, P < 0.05, n=5

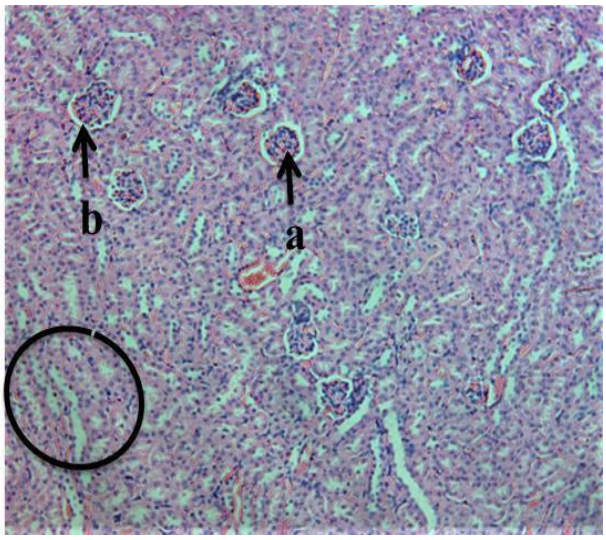


**Figure 1:** Micrograph of the kidney of control mice given feed and water showing well-structured and distinct renal histo-structure, such as glomeruli (a), Bowman's capsule (b), and renal tubules (circle). H&E, x100 magnification.

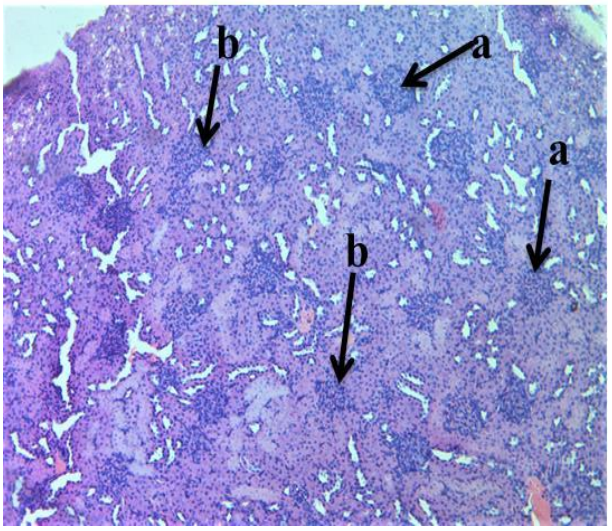


**Figure 2:** Micrograph of the kidney of group 4 mice given 100 mg/kg of gentamicin alone, showing hypertrophied glomeruli occluding the Bowman's space (a) and degenerated glomeruli in the majority of the renal corpuscles (b). H&E, x100 magnification

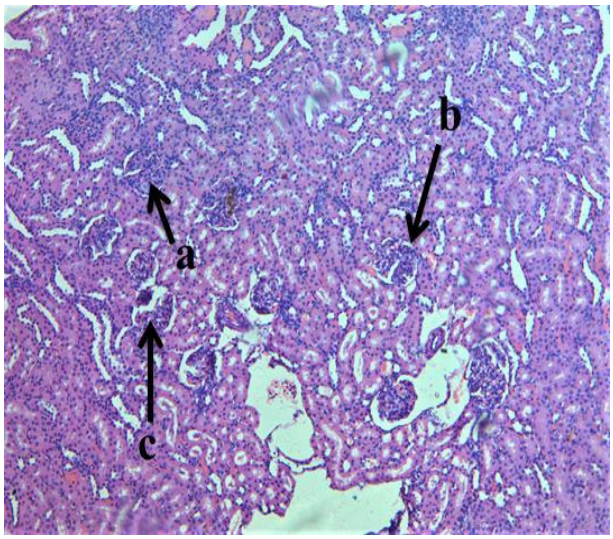


**Figure 3:** Micrograph of the kidney of group 3 mice given 100 mg/kg of gentamicin plus 50 mg/kg of extract, showing some improvements in the renal cytoarchitecture, such as glomerulus (a), Bowman's space and capsule (b), though with the presence of a degenerated glomerulus (c). H&E, x100 magnification

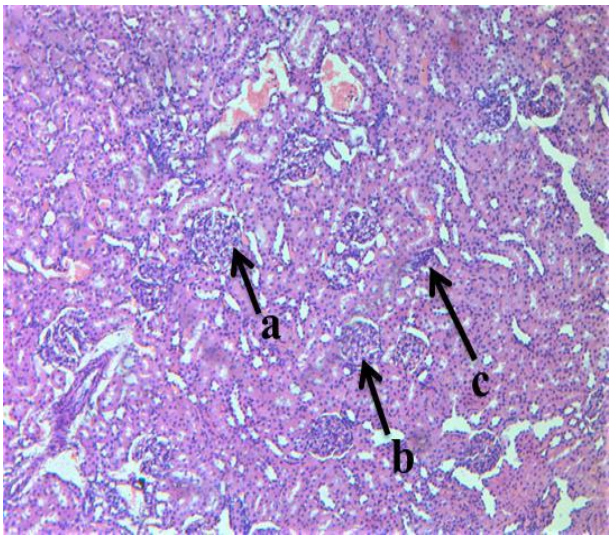


**Figure 4**: Micrograph of the kidney of group 2 mice given 100 mg/kg of gentamicin plus 100 mg/kg of extract, showing some improvements in most of the renal corpuscles, such as glomerulus (a), Bowman's capsule (b), though with the presence of a few atrophied glomeruli (**c**). H&E, x100 magnification

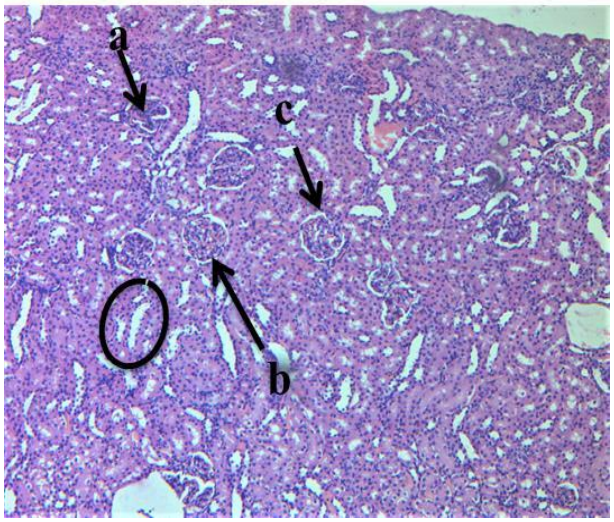


**Figure 5:** Micrograph of the kidney of group 1 mice given 100 mg/kg of gentamicin plus 200 mg/kg of extract showing improvement of the renal cyto-structure, such as glomeruli (a), Bowman's capsule (b), and Bowman's space (**c**). H&E, x100 magnification

## DISCUSSION

Acute kidney injury remains a significant clinical challenge, with drug-induced nephrotoxicity accounting for approximately 10-25% of cases [14]. Aminoglycosides, particularly gentamicin, are effective antimicrobials but are limited by their nephrotoxic potential. Gentamicin primarily targets renal tubular epithelial cells, leading to oxidative stress, lipid peroxidation, mitochondrial dysfunction, and disruption of glomerular filtration.

Administration of CEME attenuated gentamicin-induced renal dysfunction in a dose-dependent manner. The sham control group exhibited significantly elevated ($p < 0.05$) serum urea level, confirming renal impairment. In contrast, co-administration of CEME preserved renal biochemical indices, with the 200 mg/kg dose producing the most pronounced effect, as reflected by reduced serum urea and creatinine levels. Albumin and total protein concentrations remained relatively stable across all groups, suggesting preserved plasma protein homeostasis (Table 1).

These findings are consistent with earlier studies that demonstrated the nephroprotective effects of phytochemicals in plant extracts against gentamicin-induced renal injury. For instance, Abo-Elmaaty *et al*. reported that *Moringa oleifera* extract ameliorated gentamicin nephrotoxicity by lowering serum creatinine and urea levels [15], while Agu *et al*. observed similar protective effects with *Vernonia amygdalina* [16]. The biochemical preservation observed in the present study suggests that CEME may exert comparable cytoprotective actions.

Histopathological analysis supported these biochemical findings. Renal tissues from the control group (Figure 1) displayed intact and well-organised cytoarchitecture, characterised by distinct glomeruli, Bowman's capsules, and renal tubules, indicative of normal renal physiology. Conversely, kidneys from mice treated with gentamicin alone (Figure 2) exhibited marked pathological alterations, including glomerular hypertrophy, obliteration of Bowman's space, and widespread degeneration of glomerular structures, features consistent with reports by Gamaan *et al*., who described glomerular atrophy and tubular necrosis as hallmarks of gentamicin-induced nephrotoxicity [17].

Co-treatment with CEME conferred a dose-dependent protective effect. At 50 mg/kg (Figure 3), partial recovery was evident, with reappearance of Bowman's space and reduced glomerular degeneration, although some pathological changes persisted. At 100 mg/kg (Figure 4), a more substantial restoration of renal architecture was observed, with most renal corpuscles displaying normal glomeruli and Bowman's capsules, and only a few showing residual atrophy. Notably, treatment with 200 mg/kg CEME (Figure 5) produced marked histological improvement, characterised by well-formed glomeruli, intact Bowman's capsules, and preserved Bowman's space, closely resembling the control group architecture. Similar dose-dependent histological recovery has been documented with other plant-based nephroprotective agents, such as *Curcuma longa* (curcumin) [18], further supporting the role of phytochemicals in renal tissue repair. Together, the current findings support existing research on plant-based nephroprotectants and offer new evidence for the protective potential of *C. ensiformis* methanol extract against drug-induced kidney damage.

## CONCLUSION

The methanol extract of *C. ensiformis* leaves demonstrated nephroprotective activity against gentamicin-induced kidney injury in mice. The extract preserved renal biochemical parameters and histological architecture, with 200 mg/kg showing higher activity. These findings suggest that *C. ensiformis* may possesses nephroprotective potential.

**Acknowledgements**

The authors acknowledge Professor Henry Akinnibosun's contribution to the identification of the plant sample used in the study.

**Conflict of interest**

The authors declared no conflict of interest.

**Financial Support**

None declared.

**ORCID ID**

Daniel Akpe-Efiak Ambe: https://orcid.org/0000-0002-5809-1596

Joshua Charles Nworondah: https://orcid.org/0009-0003-3930-5362

## REFERENCES

1. Ostermann M, Lumlertgul N, Jeong R, See E, Joannidis M, James M. Acute kidney injury. Lancet. 2025;405(10474):241-56.
2. Wolf U. A drug safety briefing (II) in transplantation from real-world individual pharmacotherapy management to prevent patient and graft from polypharmacy risks at the very earliest stage. Pharmaceuticals. 2024;17(3):294.
3. Dobrek L. A synopsis of current theories on drug-induced nephrotoxicity. Life. 2023;13(2):325.
4. Galal HM, Abd El-Rady NM. Aqueous garlic extract suppresses experimental gentamicin-induced renal pathophysiology mediated by oxidative stress, inflammation and Kim-1. Pathophysiology. 2019;26(3-4):271-79.
5. Gamaan MA, Zaky HS, Ahmed HI. Gentamicin-induced nephrotoxicity: a mechanistic approach. Azhar Int J Pharm Med Sci. 2023;3(2):11-9.
6. Yadav RN, Ganeshpurkar A, Dubey N. Protective effects of Tinospora cordifolia on cyclophosphamide-induced nephrotoxicity in Wistar mice. Free Radic Antioxid. 2024;14(1):21-6.
7. Rahmawati E, Waangsir FWF, Selasa P. Alkaloids, flavonoids, tannins and saponins contents in *Moringa oleifera* leaves. Indones J Glob Health Res. 2022;4(1):139-44.

8. Nkwocha G, Anukam K, Adumanya O. Evaluation of the nutritional potentials of three species of Canavalia for use in livestock diets. J Sustain Technol. 2021;11(2):86-92.
9. Ambe DA, Okon EI, Udobre AS, Udoh AE. *C. ensiformis* L. (Fabaceae) attenuates benign prostatic hyperplasia: a potential phytotherapeutic agent for managing BPH. J Pharmacogn Phytochem. 2025;14(4):323- 8.
10. Rizwana K, Mohammad A, Shah CS, Fahamiya N, Shiffa M. Nephroprotective activity of the hydro-alcoholic, ethanolic extract and its fractions of tukhme soya (*Anethum sowa* Roxb.) against gentamicin-induced nephrotoxicity in albino mice. J Young Pharm. 2022;14(1):50-5.
11. Musa TY, Lawal SB, Mansur L, Musbau AA. Evaluation of selected parameters of rat liver and kidney function following repeated administration of yohimbine. Biokemistri. 2003;15(2):50-6.
12. Harrison AO, Anthonet NE, Chinna NO, Awolayeofori D, Sam JO, Anthony FU, *et al*. Hepatorenal protection of food supplement (*Prosopis africana*) in rat following metal mixture exposure via alterations in oxido-inflammatory markers. Discov Food. 2025;5:154.
13. Burtis CA, Ashwood ER, editors. Tietz textbook of clinical chemistry. London: W.B. Saunders Company; 1994.
14. Perazella MA, Rosner MH. Drug-induced acute kidney injury. Clin J Am Soc Nephrol. 2022;17(8):1220-33.
15. Abo-Elmaaty AM, Al-Shahat DS, Mohamed SA, Kamel MA. Prevention of hepato-renal toxicity with *Moringa oleifera* in gentamicin-treated mice. J Adv Vet Res. 2023;13(7):1338-46.
16. Agu, Orji P, Chime S, Duru K, Nwannadi V. Protective effect of aqueous leaf extract of *Vernonia amygdalina* (bitter leaf) on gentamicin-induced nephrotoxicity in adult Wistar rats. J Med Pharm Allied Sci. 2023;12(3):5790-95.
17. Gamaan MA, Zaky HS, Ahmed HI. Gentamicin-induced nephrotoxicity: a mechanistic approach. Azhar Int J Pharm Med Sci. 2023;3(2):11-9.
18. Jogdand SD, Shinde R, Sinha V, Chandrakar N. Nephroprotective effect of turmeric on oxidative stress, renal histopathology and toxicity induced by gentamicin. Int J Basic Clin Pharmacol. 2017;6(6):1282-86.



**HOW TO CITE THIS ARTICLE**

Ambe DAE, Nworondah JC. Reno-protective effects of *C. ensiformis* (L.) DC. leaf extract in a gentamicin-induced nephrotoxicity model in mice. J Phytopharmacol 2026; 15(2):176-180. doi: 10.31254/phyto.2026.15208