International Journal of Forensic Medicine

**E-ISSN:** 2707-4455
**P-ISSN:** 2707-4447
IJFM 2022; 4(2): 19-22
Received: 16-06-2022
Accepted: 21-07-2022

**Abu OD**
Department of Biochemistry, Faculty of Life Sciences, University of Benin, Benin City, Nigeria

**Onoagbe IO**
Department of Biochemistry, Faculty of Life Sciences, University of Benin, Benin City, Nigeria

**Ohikhuare F**
Department of Chemistry and Biochemistry, College of Letters and Science, University of Wisconsin, Milwaukee, USA

**Corresponding Author:**
**Abu OD**
Department of Biochemistry, Faculty of Life Sciences, University of Benin, Benin City, Nigeria

# Nephrotoxic evaluation of ethanol stem bark extract of *Dialium guineense* in normal wistar rats

**Abu OD, Onoagbe IO and Ohikhuare F**

**DOI:** https://doi.org/10.33545/27074447.2022.v4.i2a.51

**Abstract**
The present study was undertaken to investigate the nephrotoxic effect of ethanol extract of *Dialium guineense* stem bark in normal Wistar albino rats. Thirty-five male rats were assigned to seven (7) groups (5 rats per group). One group served as control, while graded doses of the extract (200 - 5000 mg/kg body weight, bwt) were administered to the test rats daily for twenty-eight days. The control rats received distilled water. Thereafter plasma concentrations of creatinine, urea, electrolytes and urease activity were measured. There were no significant differences in the concentrations of the measured renal parameters before and after treatment, among the groups ($p > 0.05$). These results indicate that ethanol extract of the medicinal plant stem bark may be relatively safe. However, further studies spanning several months may be necessary.

**Keywords:** *Dialium guineense*, electrolytes, medicinal plant, nephrotoxicity, renal function

## Introduction
As an organ that participates in whole-body homeostasis, the kidney regulates acid-base balance, electrolyte concentrations, extracellular fluid volume and blood pressure. The kidney accomplishes these homeostatic functions both independently and in concert with other organs, particularly those of the endocrine system [1]. Different endocrine hormones coordinate these endocrine functions. Many of the kidney's functions are accomplished by relatively simple mechanisms of filtration, reabsorption, and secretion, which take place in the nephron [2, 3]. The kidneys are responsible for elimination of unmodified drugs and metabolites. Kidney dysfunction is often found in severe liver disease and once liver function falls below a certain threshold, sodium retention occurs followed by ascites [4, 5]. Nephrotoxicity refers to injury to the kidneys or impairment of kidney function caused by exposure to xenobiotics such as drugs, food additives, alcohol, chlorinated solvents, per oxidized fatty acids, fungal toxins, radioactive isotopes, environmental toxicants, and even some medicinal plants [6]. Nephrotoxicity is one of the main toxicities of herbal medicines [7]. Plants are at the center of Traditional Medicine. Their use in disease management is as old as man [8, 9]. Medicinal plants serve as cheap alternative to orthodox medicine since they are readily available [10-12]. *Dialium guineense* is a medicinal plant that is used in parts of Africa for the treatment of various ailments [13]. In Nigeria, it is known by different names: *Icheku* (Igbo), *Awin* (Yoruba), *Tsamiyarkurm* (Hausa) and *Amughen* (Benin) [14]. The plant contains bioactive substances such as alkaloids, tannins, saponins and phenolics [15, 16]. Some of these bioactive compounds may have adverse effects on the kidney. Till date, not much is known about the subchronic toxicity of extracts of *Dialium guineense* stem bark. This study was undertaken to investigate the nephrotoxic effect of ethanol extract of *D. Guineense* stem bark in normal Wistar rats.

## Materials and Methods
### Chemicals and Kits
Reagents used in this study were of analytical grade. Kidney function tests kits were obtained from Randox Laboratories Limited (UK). All other chemicals were bought from British Drug House (BDH) (England), Merck (Germany) and Sigma-Aldrich Ltd. (USA). Fresh stem barks of *D. Guineense* were obtained from Auchi, Edo State, Nigeria and authenticated at the herbarium of the Department of Plant Biology and Biotechnology, University of Benin, Benin City, Nigeria (No. $UBH_D330$).

**Plant Extraction**

Extraction of the pulverized plant material was by maceration over a 72 h period [17]. A portion (500 g) of the powdered stem bark was soaked in 5000 mL of absolute ethanol. The resultant extract was filtered with a muslin cloth and freeze dried using a lyophilizer.

**Experimental Rats**

A total of 35 adult male wistar rats, which weighed between 160 and 180 g (mean weight = 170 ± 10 g) were procured from the Department of Anatomy, University of Benin, Benin City, Nigeria. The rats were housed in metal cages under standard laboratory conditions: room temperature, 55 – 65 % humidity and 12-h light/12-h dark cycle. They were allowed free access to pelletized growers mash and clean drinking water. Prior to commencement of the study, the rats were acclimatized to the laboratory environment for seven days. Standard experimental protocol was followed for this study.

**Experimental Design**

The rats were assigned to 7 groups (5 rats per group). One group served as control, while graded doses of the extract (200 - 5000 mg/kg body weight, bwt) were administered to the test rats daily for twenty-eight days. Blood samples were collected before treatment (basal samples) and at the end of the 28th day. Blood sample collected in plain or heparin containers was centrifuged at 3000 rpm for 10 min to obtain plasma which was used for kidney function tests.

**Kidney Function Tests**

Kidney function tests (KFTs) such as urea, creatinine, sodium ion, potassium ion, chloride ion and bicarbonate ion were performed in plasma [18-20]. Urease activity was determined as shown below:

$$\text{Urease activity (U/L)} = \frac{\Delta [\text{urea}]}{\text{time}}$$

**Statistical Analysis**

Data are expressed as mean ± SEM (n = 5). The statistical analysis was performed using SPSS (version 20). Groups were compared using Duncan multiple range test. Statistical significance was assumed at $p < 0.05$.

**Results**

**Effect of Graded Doses of Ethanol Extract of *D. guineense* Stem Bark on Weight Parameters**

Percentage increases in body weights of rats treated with ethanol extract of *D. Guineense* stem bark were significantly reduced, when compared with control group ($p < 0.05$), but there were no significant differences in the corresponding relative organ weights among the groups ($p > 0.05$; Table 1).

Data are percentage weight increase and relative kidney weight, and are expressed as mean ± SEM (n = 3). $^{a}p < 0.05$, when compared with control group; $^{b}$ $p < 0.05$, when compared with the 200 mg/kg bwt group.

**Table 1:** Percentage body weight increase and relative kidney weight of rats treated with ethanol extract of *D. Guineense* stem bark

| Groups | % Increase in weight | Relative kidney weight ($\times 10^{-3}$) |
|---|---|---|
| Control | 61.35 ± 4.11 | 3.34 ± 0.03 |
| 200 mg/kg bwt | 52.60 ± 2.92$^{a}$ | 3.60 ± 0.08 |
| 500 mg/kg bwt | 22.63 ± 1.56$^{ab}$ | 3.87 ± 0.31 |
| 1000 mg/kg bwt | 21.00 ± 1.00$^{ab}$ | 3.30 ± 0.15 |
| 2000 mg/kg bwt | 18.30 ± 1.06$^{ab}$ | 3.47 ± 0.11 |
| 3500 mg/kg bwt | 17.73 ± 0.92$^{ab}$ | 3.84 ± 0.21 |
| 5000 mg/kg bwt | 16.80 ± 1.10$^{ab}$ | 3.50 ± 0.20 |

**Indices of Kidney Function**

There were no significant differences in the plasma concentrations of creatinine, urea, electrolytes and urea/creatinine in rats treated with ethanol extract, when compared with the control group ($p > 0.05$). These results are shown in Tables 2 and 3.

**Table 2:** Concentrations of creatinine and urea in rats treated with ethanol extract of *D. Guineense* stem bark

| Groups | | Creatinine (mg/dL) | Urea (mg/dL) | Urea / Creatinine |
|---|---|---|---|---|
| Control | | 1.70±0.03 | 17.52±6.50 | 10.31±2.91 |
| 200 mg/kg bwt | B | 1.59±0.00 | 15.34±1.29 | 9.65±0.00 |
| | T | 1.73±0.03 | 16.94±1.69 | 9.79±1.03 |
| 500 mg/kg bwt | B | 1.73±0.05 | 15.56±0.32 | 8.99±0.73 |
| | T | 2.02±0.20 | 18.35±4.13 | 9.79±0.84 |
| 1000 mg/kg bwt | B | 1.50±0.03 | 17.78±1.94 | 11.85±0.47 |
| | T | 1.75±0.30 | 22.88±7.31 | 9.79±0.91 |
| 2000 mg/kg bwt | B | 1.51±0.00 | 16.25±0.00 | 10.76±0.07 |
| | T | 1.70±0.20 | 13.50±2.75 | 7.94±0.53 |
| 3500 mg/kg bwt | B | 1.02±0.34 | 23.92±0.16 | 23.45±5.02 |
| | T | 1.70±0.20 | 22.58±1.34 | 13.28±2.03 |
| 5000 mg/kg bwt | B | 2.70±0.02 | 17.85±0.64 | 6.61±0.03 |
| | T | 3.05±0.85 | 19.27±0.74 | 6.32±0.16 |

Data are indices of kidney function and are expressed as mean ± SEM (n = 5). B = Basal means; and T = Test means.

**Table 3:** Concentrations of plasma electrolytes in rats treated with ethanol extract of *D. Guineense* stem bark

| Groups | $Na^{+}$ (mmol/L) | $K^{+}$ (mmol/L) | $HCO_3^{-}$ (mmol/L) | $Cl^{-}$ (mmol/L) |
|---|---|---|---|---|
| Control | 138.50±5.50 | 7.90±0.20 | 18.00±0.40 | 105.50±6.50 |
| 200 mg/kg bwt | 136.50±1.50 | 6.55±0.15 | 17.00±2.00 | 105.50±3.50 |
| 500 mg/kg bwt | 132.50±8.50 | 5.65±1.25 | 15.50±1.50 | 101.50±5.00 |
| 1000 mg/kg bwt | 143.50±0.50 | 8.85±0.85 | 16.00±1.00 | 108.00±0.00 |
| 2000 mg/kg bwt | 143.50±0.50 | 5.95±0.05 | 19.00±2.00 | 109.50±0.50 |
| 3500 mg/kg bwt | 141.00±1.00 | 6.15±1.95 | 17.00±1.00 | 106.50±0.50 |
| 5000 mg/kg bwt | 140.00±2.00 | 7.90±0.60 | 20.50±3.50 | 109.00±1.00 |

Data are concentrations of plasma electrolytes and are expressed as mean ± SEM (n = 5).

**Effect of Graded Doses of Ethanol Extract of *D. Guineense* Stem Bark on Urease Activity**

There were no significant differences in urease activity before and after treatment, among the groups ($p > 0.05$; Figure 1).

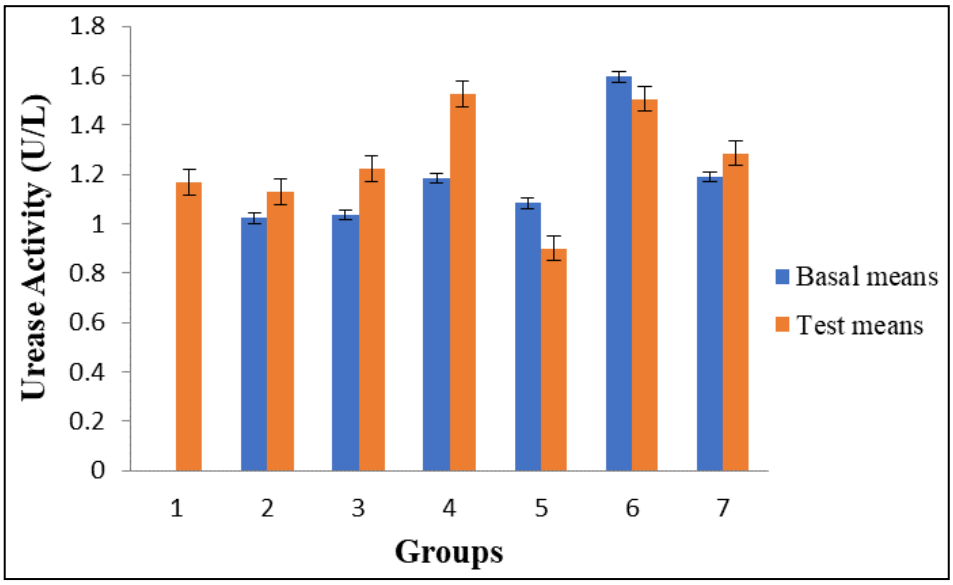


**Fig 1:** Urease activity in the different groups

## Discussion

Bioactive agents from natural sources have gained wide acceptability in modern medicine, as they are known to reduce the risk of diseases by either scavenging free radicals or halting their formation [21]. Most of the pharmacologically important drugs are derived from plants. Plant derivatives used as drugs play crucial role in health-care systems around the globe. They are not only used for the management of disease conditions but also for maintenance of proper health [22]. As good as medicinal plants are, they can be toxic at certain doses and prolonged exposure. In the 1960s, some scholars first reported cases of acute renal failure caused by the administration of *Aristolochia manshuriensis* Kom in China, after which relevant reports also appeared abroad [23]. Nephrotoxicity is one of the main toxicities of herbal medicines [7]. In 1993, Belgian scholars reported that nine European women suffered from renal failure after taking weight-loss capsules containing Chinese herbal medicine *A. Obliqua S.M. Hwang*; this condition is called "Chinese herb nephropathy (CHN)" [7]. Besides aristolochic acid, many other herbal medicine components cause renal toxicity. Severe renal tubular lesions have been found in renal puncture examinations of patients with kidney disease caused by *Tripterygium regelii Sprague ET. Takeda*. The lesions are accompanied by obvious inflammatory cell infiltration, degeneration, and necrosis of renal tubular epithelial cells [24, 25]. The administration of a large amount of *Glycyrrhiza uralensis* Fisch. Caused rhabdomyolysis and acute renal injury. Reports on the beneficial effects of *D. guineense* stem bark abound, but little or nothing is known about its nephrotoxic effect. This study investigated the nephrotoxic effect of ethanol extract of *D. guineense* stem bark in normal Wistar rats. The results showed that graded doses of ethanol extract *of D. guineense* stem bark did not significantly alter the levels of electrolytes, urea and creatinine, an indication that it may not be toxic at the subchronic level. The safety of the plant extracts had been reported in previous studies [26-31].

## Conclusion

The results obtained in this study indicate that ethanol extract *of D. Guineense* stem bark is not nephrotoxic and may be used in traditional system of medicine at doses not exceeding 5000 mg/kg bwt. However, further studies spanning several months may be necessary.

**Acknowledgement**
Not available

**Author's Contribution**
Not available

**Conflict of Interest**
Not available

**Financial Support**
Not available

## References

1. Walter F, Boron PHD. Medical Physiology: A Cellular and molecular approach. Elsevier/Saunders; c2004.
2. Clapp WL. (2009) "Renal Anatomy". In: Zhou XJ, Laszik Cotra RS, Kumar V, Fausto N, Nelso F, Robbins SL, Abbas AK. Robbins and Cotran pathologic basis of disease (7th Ed.). St. Louis, MO: Elsevier Saunders; c2005. p. 878.
3. Bard J, Vize PD, Woolf AS. The kidney: from normal development to congenital disease. Boston: Academic Press; c2003. p. 154.
4. Cotran RS, Kumar V, Fausto N, Nelso F, Robbins SL, Abbas AK. Robbins and Cotran pathologic basis of disease (7th Ed.). St. Louis, MO: Elsevier Saunders; c2005. p. 878.
5. Le T. First Aid for the USMLE Step 1. New York: McGraw-Hill Medical; c2013.
6. Abu OD, Imafidon KE, Obayuwana HO. Nephrotoxic and in vivo antioxidant effects of citrullus lanatus seed extract. Biomedical Journal of Science and Technical Research. 2021;33(5):26281-26286.
7. Vanherweghem JL, Depierreux M, Tielemans C, Abramowicz D, Dratwa M, Jadoul M. Rapidly progressive interstitial renal fibrosis in young women: association with slimming regimen including Chinese herbs. Lancet. 1993 Feb 13;341(8842):387-391.

8. Brattin WJ, Glende EA, Recknagel RO. Pathological mechanisms in carbon tetracholoride hepatotoxicity. J Free Radic Biol Med. 1985 Jan 1;1(1):27-38.
9. Grabley T, Akuodor CW. Medicinal Plants and Traditional Medicine. Watermelon Crop Information. Cucurbit Breeding Horticultural Science; c2010.
10. Abu OD, Onoagbe IO. Biochemical effect of aqueous extract of Dialium Guineense stem bark on oxidative status of normal Wistar rats. International Journal of Clinical Biology and Biochemistry. 2019;1(2):15-18.
11. Sofowora A. Medicinal Plants and Traditional Medicine in Africa. 2nd Edition. Spectrum Books Ltd., Ibadan, Nigeria; c1993. p. 289.
12. Akah PA, Nwabie AI. Evaluation of Nigerian traditional medicinal plants used for rheumatic inflammatory disorders. Journal of Ethnopharmarcology. 1994;42:179-182.
13. Dalziel JM, Hutchison J. Flora of West Tropical Africa. Vol.1 (2nd Ed). The White friars Press Ltd. London; c1973. p. 561.
14. Arogba SS, Ajiboro A, Odukwe I. A physicochemical study of Nigerian velvet tamarind (Dialium guineense L.) fruit. Journal of the Science of Food and Agriculture. 2006;66(4**):**533-534.
15. Abu OD, Onoagbe IO, Obahiagbon O. Qualitative phytochemical screening and proximate analysis of Dialium guineense stem bark. IAR Journal of Agriculture Research and Life Sciences. 2020a;1(4):108-112.
16. Abu OD, Onoagbe IO, Obahiagbon O. Analyses of metal and amino acid compositions of aqueous and ethanol stem bark extracts of Dialium guineense. Journal of Biogeneric Science and Research. 2020b;6(4):1-3.
17. Abu OD, Imafidon KE, Iribhogbe ME. Biochemical effect of aqueous leaf extract of Icacina trichanta Olivon urea, creatinine and kidney oxidative status in $CCl_4$-induced Wistar rats. Nigerian Journal of Life Sciences. 2015;5(1):85-89.
18. Chaney AL, Marbach AP. 'Modified reagents for determination of urea and ammonia'. Clin. Chem. 1962 Apr 1;8(2):130-132.
19. Blass KG, Thiebert RJ, Lam LK. Study of mechanism of JAFE reactions. Clin Chem. 1974;12(7):336-343.
20. Forrester RL, Wataji LJ, Silverman DA, Pierre KJ. Enzymatic method for determination of $CO_2$ in serum. Clin Chem. 1976;22(2):243-245.
21. Wang CZ, Mehendale SR, Yuan CS. Commonly used antioxidant botanicals: active constituents and their potential role in cardiovascular illness. Am. J. Chin. Med. 2007;35(04):543-558.
22. Dec GW. Digoxin remains useful in the management of chronic heart failure. Med. Clin. 2003 Mar 1;87(2):317-337.
23. Wu SH. Acute renal failure caused by Aristolochia manshuriensis Kom: a report of 2 cases. Jiangsu J. Tradit. Chin. Med. 1964;5:12-13.
24. Luo HM, Gu CY, Liu CX, Wang YM, Wang H, Li YB. Plasma metabolic profiling analysis of Strychnos nux-vomica Linn. And Tripterygium wilfordii Hook F-induced renal toxicity using metabolomics coupled with UPLC/Q-TOF-MS. Toxicol. Res. (Camb). 2018b Nov 1;7(6):1153-1163.
25. Luo Y, Liu HY, Zhou BH, Song JC. Research of toxicity of Herba Leonuri. Jilin J. Tradit. Chin. Med. 2018c;38:1082-1086.
26. Abu OD, Onoagbe IO, Ekugum E. Hepatotoxicity of Graded Doses of Ethanol Extract of Dialium guineense Stem Bark in Wistar Rats. Journal of Pharmaceutical and Bio-Medical Sciences. 2022a Sep 13;2(9):347-352.
27. Abu OD, Onoagbe IO, Ekugum E. Nephrotoxic Evaluation of Aqueous Stem Bark Extract of Dialium guineense in Normal Wistar Rats. Journal of Pharmaceutical and Bio-Medical Sciences. 2022b Sep 17;2(9):353-357.
28. Abu OD, Alegun O, Ojo AU. Pancreatotoxicity of Ethanol Extract of Dialium guineense Stem Bark in Rats. World Journal of Pharmaceutical and Life Sciences. 2022;8(11):40-45.
29. Abu OD, Okuo AV, Ayele PE. Pancreatotoxicity Effect of Aqueous Extract of Dialium guineense Stem Bark in Wistar Rats. International Journal of Novel Research in Life Sciences. 2022;9(5):31-37.
30. Abu OD, Odagwe UB, Ojo AU. Cardio toxicity of Ethanol Extract of Dialium guineense Stem Bark in Rats. World Journal of Pharmaceutical and Life Sciences. 2022;8(11):34-39.
31. Abu OD, Umar AB, Adekanle E. Cardiotoxic Effect of Aqueous Extract of Dialium guineense Stem Bark in Wistar Rats. East African Scholars Journal of Agriculture and Life Sciences. 2022;5(9):167-172.



**How to Cite This Article**
Abu OD, Onoagbe IO and Ohikhuare F. Nephrotoxic evaluation of ethanol stem bark extract *of Dialium guineense* in normal wistar rats. International Journal of Forensic Medicine. 2022;4(2):19-22. DOI: https://doi.org/https://doi.org/10.33545/27074447.2022.v4.i2a.51