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# Serum Biochemical And Histopathological Studies On Hepatic And Renal Effects Of Aqueous Leaves Extract Of *Olea Hochstetteri* Bak. In Wistar Albino Rats

**Aji S.B.**

Department of Animal Health and Production Technology, Adamawa State College of Agriculture Ganye, Nigeria

**Egbe-Nwiyi T.N.C.**

Department of Veterinary Pathology, Faculty of Veterinary Medicine, University of Maiduguri, Nigeria

**Sandabe U.K.**

Department of Veterinary Physiology and Biochemistry, Nigeria

***Abstract:*** ***The use of Olea hochstetteri Bak. in traditional medicine in Nigeria to treat various illnesses necessitated the need to examine its effect following prolonged use. In this research, the effects of orally administered aqueous leaves extract of Olea hochstetteri Bak. on some serum biochemical parameters and the on liver and kidney of rats were studied. Sixty Wistar albino rats randomly separated into 4 groups (A to D) of 15 rats each were used in the experiment. Rats in groups A, B, C and D were given daily oral dose of 250, 500 and 1000 mg/kg of extract and distilled water (control) respectively for 3 weeks. At the end of each week, 5 rats from each group were humanely sacrificed and their sera were collected for biochemical analyses for aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), total and conjugated bilirubin, total protein, albumin, urea, and creatinine and their livers and kidneys were collected for histopathological studies using standard techniques and procedures. The result of the study showed that the extract caused very significant (P<0.05) increases in AST, ALT and ALP in groups A, B and C. Significant (P<0.05) increase in serum total and conjugated bilirubin, urea and creatinine and decrease in serum total protein and albumin were observed in rats in groups B and C. Histopathological studies revealed only mild degenerative changes and diffused interstitial mononuclear cellular infiltration of the hepatic parenchyma with evidence of hydrophic changes, with degeneration of renal tubular epithelia and necrosis at day 21 in rats in group A. Various types and degrees of histopathological changes were observed in rats in groups B and C throughout the treatment period. These include severe hepatic periportal mononuclear cellular infiltration, moderate to severe congestion of the liver and necrosis of hepatocytes in the liver. Also, severe degeneration of the renal tubular epithelia, renal interstitial haemorrhages, marked necrosis of the renal glomerular and tubular epithelial cells, evidence of amyloidosis within the renal glomerular corpuscles and the presence of protein casts deposits within the renal tubular lumen were observed in the kidneys. The severity of these lesions appeared to be dose and time-dependant. It was concluded that the extract is toxic to rats causing damage to the liver and kidney especially when given at 500 and 1000 mg/kg but could be given at a dosage not exceeding 250 mg/kg and for a period not more than 1 week. Further toxicological studies on the extract, with the view to understanding its effects on other organs of the body, such as the hematopoietic, cardiovascular, respiratory, nervous, reproductive and digestive organs was recommended.***

***Keywords: Olea hochstetteri Bak., Leaves Extract, Serum Biochemistry, Histopathology, Hepatic/renal Effects, Albino Rats***

## I. INTRODUCTION

*Olea hochstetteri* Bak. (English – East African Olive) is a tree of the Montane forest that is widely distributed in Africa and belongs to the family Oleaceae (Hutchingson and Dalziel, 1958). It is a small tree with dark green leathery leaves and thick, smooth, grayish bark (Keay, *et al*., 1964). In Nigeria, it is used in traditional medicine for the treatment of febrile

illnesses, severe headache, psychiatric illness, abdominal pain, relief from witchcraft and evil spirits, in wound dressing and against diseases of unknown etiologies (Mahre *et al*. 2009; Aji *et al*., 2010a). Studies revealed that the intra-peritoneal $LD_{50}$ of the aqueous leaves extract of *O. hochstetteri* Bak. in rats is 1280 mg/kg with the extract causing decrease in PCV, Hb, MCV, MCHC, total WBC and lymphocyte counts. Phytochemical analysis of both the aqueous and ethanol leaves extracts revealed the presence carbohydrate (reducing sugars, ketones and pentoses), gallic tannins, saponins, alkaloids, sterols and triterpenes, flavone aglycones, emodols and cardiac glycosides (Mahre *et al*., 2009; Aji *et al*., 2010a). Hypoglycemic, anticholesterolemic and body weight lowering effect of the aqueous leaves extract (Aji *et al*., 2010b) and its antibacterial effect against *Pseudomonas aeroginosa*, *Staphylococcus aureus*, *Salmonella typhi* and *Klebsiella pneumonia* was also reported (Aji *et al*., 2010a). The extract was also reported to prolong the anesthetic effect of thiopentone sodium and ketamine hydrochloride (Peter *et al*., 2013).

Most scientific evaluations of medicinal plants are mainly concerned with validating their traditional use and identifying the active components of their extract preparations. Therefore, the continued examination of these plants is required, not only to establish the scientific basis for their activity, but also to enable the quantity, efficacy, and safety of these preparations to be better assessed (Palombo, 2006). In spite of the use of *O. hochstetteri* Bak as medicinal plant, serum biochemistry and tissue changes following prolonged oral administration of its leaves extract has not been evaluated. The objective of this study was to determine the effect of prolonged oral administration of the aqueous leaves extract of *O. hochstetteri* Bak. on serum biochemical parameters (aspartate aminotransferase - AST, alanine aminotransferase - ALT, alkaline phosphatase - ALP, total and conjugated bilirubin, total protein, albumin, urea, and creatinine) and to study histopathological changes associated with its effects on the liver and kidney of Wistar albino rats.

## II. MATERIALS AND METHODS

### A. COLLECTION AND IDENTIFICATION OF PLANT MATERIAL

Fresh leaves of *O. hochstetteri* Bak. were collected from Mafa Local Government Area of Borno State. The leaves were identified and authenticated by a taxonomist in the Department of Biological Sciences, University of Maiduguri, Nigeria and voucher specimen (Spp Vet. 206 A) was deposited at the University Herbarium. The leaves were shade-dried, crushed and pulverized and then kept in cellophane bags till extraction.

### B. PREPARATION OF AQUEOUS LEAVES EXTRACT

The aqueous leaves extract was prepared according to the methods of Mittal *et al*. (1981) and Fernando *et al*. (1989). Two hundred grams of the powdered leaves was mixed with 1 L of water in a 5 L beaker. The mixture was warmed at 65$^{o}$C for one hour, allowed to cool and mixed vigorously. It was filtered using Whatman No.1 filter paper, concentrated by evaporation at 60$^{o}$C in a water bath and the extract stored at 4 $^{o}$C. A stock solution of 0.5 g/mL was prepared for administration.

### C. THE EXPERIMENTAL ANIMALS

Sixty Wistar strain albino rats of both sexes weighing 109 ± 19 g were used in the experiment. They were kept in clean rat cages in the Department of Veterinary Physiology Laboratory of University of Maiduguri and were given water and commercially prepared feeds *ad libitum*. They were allowed to adjust for 3 weeks before the start of the experiment. Since ethical endorsement was not applicable as at the time of the experiment, animals were handled according to the international guiding principles for biomedical research involving animals (CIOMS, 1985).

### D. TREATMENT OF ANIMALS WITH PLANT EXTRACT

The rats were randomly separated into four groups (A, B, C and D) of 15 rats each. Rats in groups A, B and C were treated with daily oral dose of the extract at 250, 500 and 1000 mg/kg body weight respectively for 3 weeks. Rats in group D received only distilled water for the same period.

### E. SAMPLE COLLECTION

At the end of each week, from the commencement of extract administration, 5 rats were randomly picked from each group (A, B, C and D) and humanely sacrificed by decapitation. Blood samples were collected into sterile plain sample bottles and the sera obtained were used for biochemical analyses. Their livers and kidneys were also collected for histopathological study.

### F. SERUM BIOCHEMICAL ANALYSES

Aspartate aminotranferase (AST) and Alanine aminotranferase (ALT) were determined calorimetrically using standard procedures in accordance with the principles of Reitman and Frankel (1957). Serum alkaline phosphatase (ALP) levels were photometrically determined (Klein *et al*., 1960; Babson *et al*., 1966). To determine serum total and conjugated bilirubin, the calorimetric method based on the principles described by Jendrassik and Grof (1938) was adopted. Serum total protein level was determined calorimetrically using the Biuret method (Weichselbaum, 1946). The bromocresol green method was adopted for the determination of serum albumin, (Doumas *et al*., 1971). Blood urea was determined by diacetyl method of Natelson (1951) while serum creatinine was determined based on Jaffe's reaction.

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### G. POSTMORTEM EXAMINATION AND HISTOPATHOLOGICAL STUDIES

Postmortem examination of all sacrificed rats was done according to standard procedure (Igbokwe, 1989). The liver and kidneys were prepared and stained with Haemayoxylin and Eosin (H&E) for histological examination using standard techniques (Junqueira and Carneiro, 2005) and were photographed at × 400 objective.

### H. STATISTICAL ANALYSIS

Data obtained from the studies were analyzed using computer software (GraphPad InStat, Instant Biostatistics, Version 3, by GraphPad Software Inc®, U.S.A.). The results obtained were presented as Mean ± Standard deviation (SD) and differences between means were analyzed using ANOVA with Tukey-Kramer multiple comparisons post test. Level of significance of differences between means was considered at $P \leq 0.05$.

## III. RESULTS AND DISCUSSION

### A. THE EFFECTS OF AQUEOUS LEAVES EXTRACT OF *O. HOCHSTETTERI* BAK. ON SERUM ENZYMES OF RATS

The effect of aqueous leaves extract of *O. hochstetteri* Bak. on the enzymes AST, ALT, and ALP are presented in Table 1. There was very significant ($P<0.01$) increases in the serum levels of the 3 enzymes at all the treatment dosages i.e. 250 mg/kg, 500 mg/kg and 1000 mg/kg at day 7, 14, and 21 of treatment when compared with their respective control groups.

This study revealed that oral administration of aqueous leaves extract of *O. hochstetteri* Bak. to rats at all the treatment regimens significantly increased the serum enzymes, AST, ALT and ALP indicating that the liver was adversely affected. An elevation in the serum activity of AST is known to occur in association with liver diseases in a variety of animal species and it is of diagnostic importance in the assessment of the level of liver cell damage especially if no disease exists in other tissues in which it is found in high concentration (Coles, 1974). Similarly, a concurrent rise in plasma activities of ALT and AST (as observed in this study) is an indication of liver damage (Kaplan *et al*., 1998; Murray *et al*., 2006). The elevation of these enzymes is a sensitive indicator of damage to the cytoplasmic and/or mitochondrial membranes of liver cells resulting to altered membrane permeability and cell necrosis (Coles, 1974; Mayne, 1994). In a similar study (Farag *et al*., 2006), the crude leaf juice of a closely related plant, *Olea europaea* administered to rats at 200 ppm for 6 weeks was reported to induce significant increases in the serum AST, ALT and ALP.

### B. THE EFFECT OF AQUEOUS LEAVES EXTRACT OF *O. HOCHSTETTERI* BAK. ON SERUM TOTAL AND CONJUGATED BILIRUBIN, TOTAL PROTEIN AND ALBUMIN OF RATS

The effects of the extract on serum total and conjugated bilirubin (Table 2) indicated that there was no significant ($P>0.05$) changes in the serum levels of total bilirubin for the 250 mg/kg treatment group at day 7 and 14 of treatment and so also for the 500 mg/kg treatment group at day 7. However, there was significant ($P<0.05$) increase in serum total bilirubin in the 250 mg/kg treatment group by day 21 while very significant ($P<0.01$) increases were observed for the 500 mg/kg treatment group at days 14 and 21 and for 1000 mg/kg group at day 7, 14, and 21 with the highest mean level of 14.00 µmol/L attained at treatment with 1000 mg/kg of extract by day 21. The serum levels of conjugated bilirubin was not significantly ($P>0.05$) affected throughout the course of treatment with extract at 250 mg/kg. Similar result was observed for the 500 mg/kg at day 7 and 14 however, this group showed very significant ($P<0.01$) increase in serum conjugated bilirubin by day 21. At extract dosage of 1000 mg/kg, there was significant ($P<0.05$) rise in serum conjugated bilirubin by day 7 and 14, which became very significant ($P<0.01$) by day 21. The effect of the extract on serum total protein and albumin of rats are presented in Table 3. Daily dosage of 250 mg/kg of extract did not produce any significant ($P>0.05$) change in the levels of serum total protein and albumin by day 7, 14 and 21. Similarly, by day 7 of treatment with 500 mg/kg, there was no significant ($P>0.05$) changes observed in both serum total protein and albumin levels, however at day 14 and 21, they were very significantly ($P<0.01$) decreased. Very significant ($P<0.01$) decreases in both serum total protein and albumin were observed throughout the treatment period with 1000 mg/kg of extract.

The above findings in conjunction with the serum enzymes, AST, ALT and ALP is an indication that liver damage due to treatment of rats with aqueous leaves extract of *O. hochstetteri* Bak. at 250 mg/kg is mild. At treatment dosages of 500 and 1000 mg/kg however, liver (and possibly other organs) damage are severe as indicated by the very significant increases in serum enzymes (ALT, AST and ALP), increase in total and conjugated bilirubin and decrease plasma protein involving primarily the albumin fraction. Cotran *et al*. (1999) noted that decrease in total serum protein occur in severe renal disease, and a decrease in plasma protein involving primarily the albumin fraction results when there is damage to the renal glomeruli leading in defective glomerular filtration. The decrease in plasma albumin levels following administration of 500 mg/kg and 1000 mg/kg of extract at 2 to 3 weeks of treatment could be ascribed to renal insufficiency.

### C. THE EFFECT OF AQUEOUS LEAVES EXTRACT OF *O. HOCHSTETTERI* BAK. ON SERUM UREA AND CREATININE LEVELS OF RATS

The effect of the extract on the serum levels of urea and creatinine of treated rats are shown in Table 4. Rats that received 250 mg/kg of extract did not exhibit any significant ($P>0.05$) changes in their serum urea and creatinine levels throughout the treatment when compared to the control group.

Similarly, treatment with 500 mg/kg did not produce significant (P>0.05) change in serum urea level by day 7 but significantly (P<0.05) increased serum creatinine. The same treatment produced very significant (P<0.01) increases in both urea and creatinine by day 14 and 21. At 1000 mg/kg, very significant (P<0.01) increases in serum urea and creatinine levels were observed throughout the treatment period. In a Similar study (Farag *et al*., 2006), increased serum urea and creatinine values were observed following oral administration of 200 ppm of *O. europaea* to rats for 6 weeks with severe damage to the kidneys observed during histopathological examination.

Urea is derived in the liver from amino acids and therefore from protein. The normal kidney can excrete large amount of urea. If for any reason the rate of production exceeds the rate of clearance, plasma concentration rises (Kaplan *et al*., 1998; Murray *et al*., 2006). Creatinine on the other hand is derived from endogenous sources by tissue creatine breakdown. Its rate of excretion depends on the functional state of the kidneys (Mayne, 1994). Rising serum values of urea and creatinine as those caused by treatment with 500 mg/kg and 1000 mg/kg in this study is an indication of kidney damage (Cole, 1974; Mayne, 1994; Kaplan *et al*., 1998). According to reports by Mendelssohn *et al*. (1999), mildly elevated serum creatinine levels is in most cases an indication of lost of about 50% of renal filtration function and the presence of mild to moderate renal insufficiency.

### D. OUTCOME OF POSTMORTEM EXAMINATIONS AND HISTOPATHOLOGICAL STUDIES OF THE LIVER AND KIDNEY OF RATS TREATED WITH AQUEOUS LEAF EXTRACT OF *O. HOCHSTETTERI* BAK.

There was no gross pathologic change in the group that received 250 mg/kg of the extract throughout the period of the experiment. Carcasses of rats in the 500 mg/kg and 1000 mg/kg groups appeared much leaner than those of control group and the group that received 250 mg/kg by day 21. In addition, 3 rats in the 500 mg/kg treatment group and 11 in the 1000 mg/kg treatment group showed evidence of hepatomegaly and a general pale appearance of their muscles.

Various types of histopathologic changes were observed in the livers and kidneys of all the rats that were treated with the extract. The severity of these changes appeared to be dose and time-dependant. Histopathologic changes observed in the liver of rats treated with 250 mg/kg of extract at day 7 and 14 included mild vacuolar degeneration and diffused mononuclear cellular infiltration of the hepatic parenchyma (Fig: 1a), severe periportal mononuclear cellular infiltration and moderate to severe congestion of the liver (Fig: 1b and Fig: 1c). Similar histopathological changes were observed in the liver of rats treated with 500 mg/kg (Fig. 1d) and 1000 mg/kg (Fig. 1e) of the extract at day 7 and 14 except that the lesions were of greater severity and magnitude. There was necrosis of the hepatocytes observed at day 14 of treatment with 1000 mg/kg of the extract (Fig: 1e). These histopathologic findings further substantiates the evidence of liver damage earlier revealed by the elevated serum enzymes (AST, ALT and ALP) and decreased plasma proteins mainly of the albumin fraction. Similar histopathologic finding (Farag *et al*., 2006) was reported for treatment of rat with 200 ppm of crude leaves juice of *O. europaea* for 6 week.

The kidneys of rats treated with 250 mg/kg of the aqueous extract. showed evidence of diffused interstitial mononuclear cellular infiltration, hydrophic changes with degeneration of renal tubular epithelia and necrosis at day 21 (Fig: 2a). In rats treated with 1000 mg/kg of extract, severe renal damages were observed as early as day 7 of treatment. These included severe degeneration of renal tubular epithelia, interstitial haemorrhages and marked necrosis of the renal glomerular and tubular epithelial cells (Fig: 2b). At day 21, there was evidence of amyloidosis within the renal glomerular corpuscles (Fig: 2c) and the presence of protein casts deposited within the renal tubular lumen (Fig: 2d). These findings strongly support the evidence of renal damage as revealed by the serum biochemical studies and are a confirmation of toxicity of the extract of *O. hochstetteri* Bak. to the kidneys.

## IV. CONCLUSION

This study revealed that prolonged oral administration of 250 to 1000 mg/kg of aqueous leaves extract of *O. hochstetteri* Bak. caused significant changes in serum biochemical parameters and various histipathologic changes in liver and kidney tissues of albino rats, indicating toxicity. The extract can however be given at a dosage not exceeding 250 mg/kg for a period not exceeding 1 week. We recommend further toxicological studies on the leaf extract of *O. hochstetteri* Bak. with the view to understanding its effects on other organs of the body, such as the hematopoietic, cardiovascular, respiratory, nervous, reproductive and digestive organs.

| Enzyme | Groups (n=5) | Enzyme concentration (U/L) in serum (Mean ± SD) | | |
|---|---|---|---|---|
| | | Treatment days | | |
| | | day 7 | day 14 | day 21 |
| Aspartate aminotransferase (AST) | Control | 65.80±2.43 | 63.20±1.93 | 64.60±1.84 |
| | 250 mg/kg | 104.20±4.09** | 135.60±6.96** | 134.20±6.54** |
| | 500 mg/kg | 115.20±6.56** | 136.75±8.09** | 153.40±6.67** |
| | 1000 mg/kg | 164.60±7.49** | 196.20±9.83** | 208.00±10.89** |
| Alanine Aminotransferase (ALT) | Control | 26.60±1.96 | 24.20±0.97 | 25.40±1.76 |
| | 250 mg/kg | 58.80±1.79** | 66.40±2.53** | 76.40±3.02** |
| | 500 mg/kg | 74.20±2.07** | 76.75±3.78** | 82.20±1.36** |
| | 1000 mg/kg | 74.00±3.88** | 90.60±1.32** | 91.80±3.98** |
| Alkaline Phosphatase (ALP) | Control | 88.60±3.99 | 85.80±1.37 | 89.40±2.51 |
| | 250 mg/kg | 133.20±1.23** | 176.40±3.31** | 176.00±3.73** |
| | 500 mg/kg | 147.60±3.62** | 191.20±2.31** | 200.20±4.36** |
| | 1000 mg/kg | 207.00±3.82** | 208.80±5.38** | 226.00±6.52** |

** = *Very significant (P<0.01) compared with control.*

*Table 1: Effect of aqueous leaves extract of O. hochstetteri Bak. on serum levels of enzymes, AST, ALT and ALP*

| | Groups (n=5) | Concentration in serum (Mean ± SD) | | |
|---|---|---|---|---|
| | | Treatment days | | |
| | | day 7 | day 14 | day 21 |
| Total bilirubin (µmol/L) | Control | 6.00±0.58 | 6.00±0.71 | 5.8±0.87 |
| | 250 mg/kg | 6.02±0.14 | 6.50±0.92 | 6.95±0.18* |
| | 500 mg/kg | 6.60±0.14 | 7.60±0.30** | 8.75±0.71** |
| | 1000 mg/kg | 9.00±0.71** | 13.40±0.30** | 14.00±0.80** |

| | | | | |
|---|---|---|---|---|
| Conjugated bilirubin (µmol/L) | Control | 3.08±0.84 | 3.40±0.89 | 3.18±0.48 |
| | 250 mg/kg | 3.40±0.54 | 3.06±0.52 | 3.21±0.20 |
| | 500 mg/kg | 3.40±0.55 | 3.00±0.29 | 2.32±0.32** |
| | 1000 mg/kg | 4.44±0.91* | 2.20±0.35* | 1.98±0.28** |

** = Significant (P<0.05) compared with control.*
***= Very significant (P<0.01) compared with control.*

*Table 2: Effect of aqueous leaves extract of O. hochstetteri Bak. on serum total and conjugated bilirubin*

| | Groups (n=5) | Concentration in serum (Mean ± SD) Treatment days | | |
|---|---|---|---|---|
| | | day 7 | day 14 | day 21 |
| Total Protein (g/L) | Control | 76.40±1.01 | 76.80±0.87 | 74.60±1.05 |
| | 250 mg/kg | 75.50±1.04 | 75.70±0.88 | 74.60±1.08 |
| | 500 mg/kg | 74.70±0.62 | 69.00±1.25** | 51.80±1.30** |
| | 1000 mg/kg | 53.20.±1.06** | 47.80±0.35** | 34.40±0.67** |
| Albumin (g/L) | Control | 38.00±0.52 | 37.80±1.09 | 37.00±1.23 |
| | 250 mg/kg | 37.40±0.16 | 37.10±0.17 | 37.00±0.71 |
| | 500 mg/kg | 37.10±1.08 | 33.80±0.72** | 30.20±1.03** |
| | 1000 mg/kg | 33.40±0.86** | 28.33±1.06** | 20.60±1.08** |

*** = Very significant (P<0.01) compared with control.*

*Table 3: Effect of aqueous leaves extract of O. hochstetteri Bak. on serum total protein and albumin.*

| | Groups (n=5) | Concentration in serum (Mean ± SD) Treatment days | | |
|---|---|---|---|---|
| | | day 7 | day 14 | day 21 |
| Urea (mmol/L) | Control | 4.05±1.05 | 3.85±0.67 | 3.82±0.57 |
| | 250 mg/kg | 4.08±0.60 | 4.07±0.79 | 4.13±0.31 |
| | 500 mg/kg | 4.66±1.27 | 5.53±0.47** | 5.64±0.63** |
| | 1000 mg/kg | 5.82±0.36** | 6.78±0.84** | 8.42±0.81** |
| Creatinine (µmol/L) | Control | 68.96±1.08 | 70.08±0.65 | 70.04±0.38 |
| | 250 mg/kg | 70.07±0.60 | 69.88±0.36 | 70.91±0.93 |
| | 500 mg/kg | 70.50±0.61* | 78.75±0.67** | 94.46±0.90** |
| | 1000 mg/kg | 77.00±1.00** | 98.40±0.93** | 114.40±1.09** |

** = Significant (P<0.05) compared with control.*
*** = Very significant (P<0.01) compared with control*

*Table 4: Effect of aqueous leaves extract of O. hochstetteri Bak. on serum urea and creatinine*

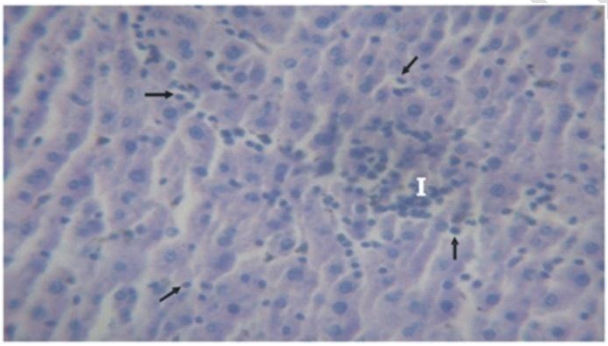


*Fig 1a: Photomicrograph of liver of rat treated with 250 mg/kg of extract. Note the diffused interstitial mononuclear cellular infiltration (I) and mild vacuolar degeneration (arrows). H&E x400*

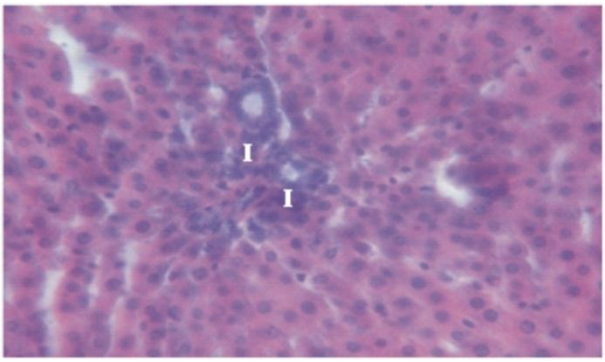


*Fig 1b: Photomicrograph of liver of rat treated with 250 mg/kg of extract at day 7. Note the marked periportal mononuclear cellular infiltration (I). H&E x400*

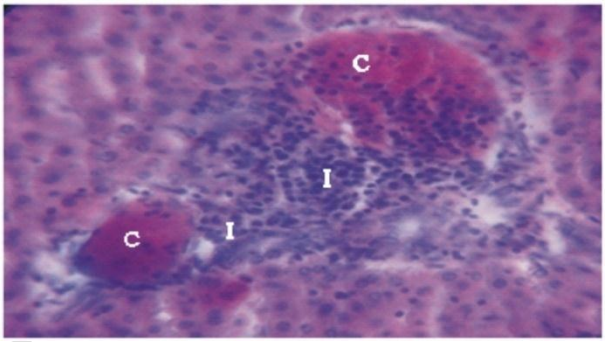


*Fig 1c: Photomicrograph of liver of rat treated with 250 mg/kg of extract at day 14 showing areas of marked congestion (C) and periportal mononuclear cellular infiltration (I). H&E x400*

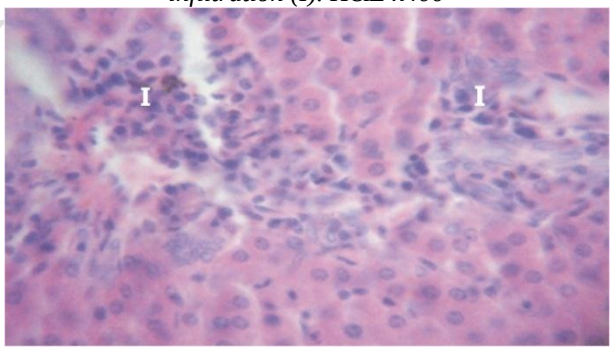


*Fig 1d: Photomicrograph of liver of rat treated with 500 mg/kg of extract at day 14. Note the severe periportal mononuclear cellular infiltration (I). H&E x400*

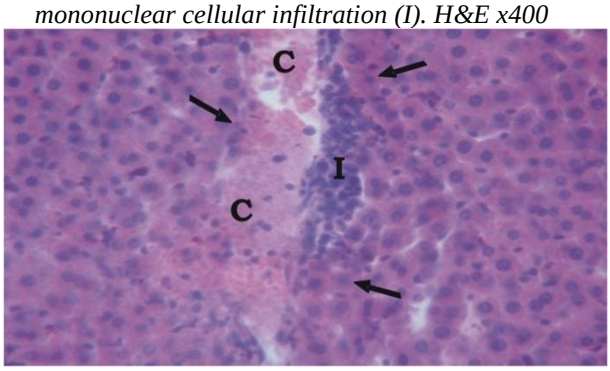


*Fig 1e: Photomicrograph of liver of rat treated with 1000 mg/kg of extract at day 14 showing severe congestion (C), marked mononuclear cellular infiltration (I) and necrosis of hepatocytes (arrows). H&E x 400*

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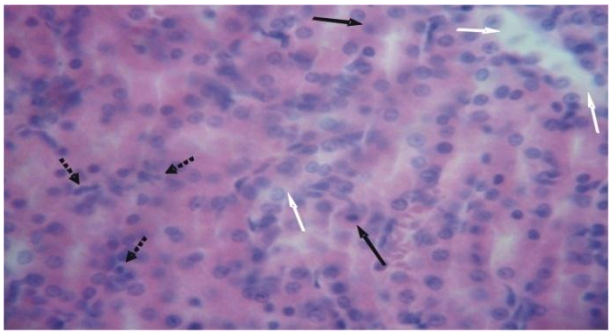

*Fig 2a: Photomicrograph of kidney of rat treated with 250 mg/kg of extract at day 21 showing interstitial mononuclear cellular infiltration (dotted arrows), degeneration of renal tubular epithelium (white arrows) and necrosis (black arrows). H&E x400*

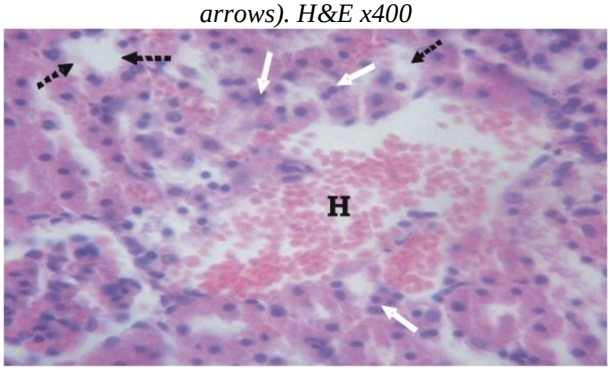


*Fig 2b: Photomicrograph of kidney of rat treated with 1000 mg/kg of extract at day 7. Note the severe haemorrhage (H), degeneration of the tubular epithelium (dotted arrows), and necrosis of the tubular epithelial cells (white arrows). H&E x400*

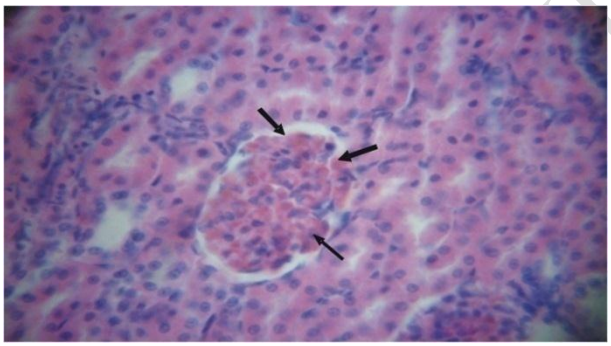

*Fig 2c: Photomicrograph of kidney of rat treated with 1000 mg/kg of extract at day 14 showing homogenous pinkish substance suspected to be evidence of amyloid deposits (arrows) within the renal glomerular corpuscle. H&E x400*

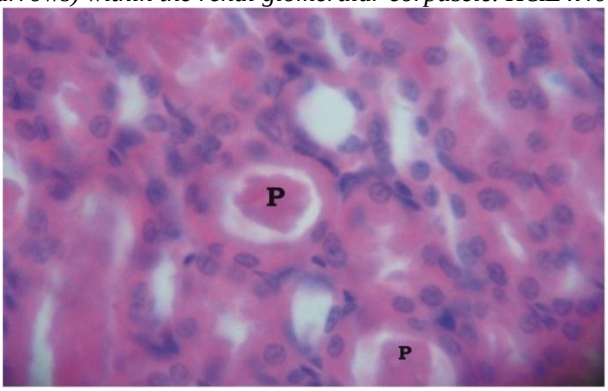


*Fig 2d: Photomicrograph of kidney of rat treated with 1000 mg/kg of extract at day 21. Note the presence of protein cast deposits (P) within the renal tubular lumen. H&E x800.*

## REFERENCES

[1] Aji, S.B., Auwal, M.S., Onyeyili, P.A. and Dawurung, C.J. (2010a). Phytochemical screening and antibacterial evaluation of the leaves extracts of Olea hochstetteri Bak. (Oleaceae). Research Journal of Pharmacology, 4(2): 26-30

[2] Aji, S.B., Sandabe, U.K. and Egbe-Nwiyi, T.N.C. (2010b). Hypoglycemic, anticholesterolemic and body weight-lowering effect of aqueous leaves extract of Olea hochstetteri Bak. in rats. Research Journal of Biological Sciences, 5(11): 745-749

[3] Babson, L.A., Greeley, S.J., Coleman, C.M. and Phillips, G.D. (1966). Serum alkaline phosphatase determination. Clin. Chim., 12: 482-490.

[4] CIOMS (1985). International guiding principles for biomedical research involving animals. Council for International Organization of Medical Sciences (CIOMS) c/0 World Health Organization 1211, Geneva 27, Switzerland.

[5] Coles, E.H. (1974), Veterinary Clinical Pathology. 2nd edition. Philadelphia, London, Toronto: W.B. Saunders Company.

[6] Cotran, R.S., Kurma, V. and Collins, T. (1999). Pathologic basis of disease. 6th edition. Philadelphia/Pennsylvania, USA: W.B. Saunders Company

[7] Doumas, B.T., Watson, W.A and Biggs, H.G. (1971). Albumin standards and the measurements of serum albumin with bromocresol green. Acta., 31: 87-96.

[8] Farag, R.S., Mahmoud, E.A., Basuny, A.M., and Ali, R.F.M. (2006). Influence of crude olive leaf juice on rat liver and kidney functions. Int. J. Food Sci. Tech., 41(7): 790-798.

[9] Fernando, M.R., Wickramasinghe, M.D., Thabrew, M.I. and Karnayaka, E.K. (1989). A preliminary investigation of the possible hypoglycemic activity of Asteracanthus longifolia. J. Ethnopharmacol., 27: 7-14.

[10] Hutchingson, J. and Dalziel, J.M. (1958). Flora of West Tropical Africa. 2nd edition. Volume 2. Revised by R.W.J. Keay. Wilbank, London: Crown Agents for Oversea Government and Administration

[11] Igbokwe, I.O. (1989). Postmortem examination of carcasses. Zaria Veterinarian, 4(2): 145-152.

[12] Jandrassik, L. and Grof, P. (1938). Calorimetric method for the determination of total bilirubin. Biochem 2, 297: 8.

[13] Junqueira, L.C. and Carneiro, J. (2005). Basic histology: Text and atlas. 11th edition. U.S.A.: McGraw-Hill Companies Inc.

[14] Kaplan, A., Szabo, L.L, and Opheim, K.E. (1998). Clinical chemistry: interpretation and techniques. 3rd edition. Philadelphia: Lea and Febiger.

[15] Keay, R.W.J., Onochie, C.F.A. and Stanfield, D.P. (1964). Nigerian trees. Vol 2. Ibadan, Nigeria: Federal Department of Forest Research.

[16] Klein, B., Read, P.A. and Babson, L.A. (1960). Calorimetric method for the determination of alkaline phosphatase. Clin. Chim., 6: 269-275.
[17] Mahre, M.B., Sandabe, U.K. and Abdulrahman, F.I, (2009). Phytochemical analysis of aqueous leaf extract of Olea hochstetteri Bak. (Oleaceae) and its effects on hematological parameters in rats. Nigerian Veterinary Journal, 29(4): 24-33.
[18] Mayne, P.D. (1994). Clinical chemistry in diagnosis and treatment. 6th edition. London, New York, New Delhi: Arnold.
[19] Mendelssohn, D.C., Barret, B.J., Brownscombe, L.M., Ethier, J., Greenberg, D.E., Kanani, S.D. Levin, A. and Toffelmire, E.B. (1999). Elevated levels of serum creatinine: Recommendation for management and referral. Canadian Medical Association Journal, 161(4): 413-423.
[20] Mittal, G.C., Aguwa, C.N., Eziefe, V.U. and Akabue, P.L. (1981). Preliminary pharmacological studies on antivenom action of Diodia scandens. Nig. J. Pharm. 12: 32-36.
[21] Murray, R.K., Granner, D.K., and Rodwell, V.W. (2006). Harper's illustrated biochemistry. 27th edition. U.S.A: McGraw-Hill Companies Inc.
[22] Natelson, S. (1951). Routine ultramicro-methods in clinical laboratory. Am. J. Clin. Pathol., 21: 1153-1172.
[23] Palombo, E.A. (2006). Phytochemicals from traditional medicinal plants used in the treatment of diarrhea: mode of action and effects on intestinal function. Phytother. Res., 20: 717-724.
[24] Peter, I.D., Sandabe, U.K. and Adamu, J.Y (2013). Aqueous leaf extract of Olea hochstetteri modulates thiopentone- and ketamine-induced anesthesia in rats. Journal of Experimental and Integrative Medicine, 3(2): 141-144.
[25] Reitman, S. and Frankel, s. (1957). A calorimetric method for the determination of serum glutamic oxaloacetic and glutamic piruvic transaminases. Am. J. Clin. Pathol., 28: 56-63.
[26] Weichselbaum, T.E. (1946). The Biuret method for total protein detection. Amer. J. clin. Path. Tech. Sect., 16: 40.