Research Article

# *In vivo* Evaluation of Genetic and Systemic Toxicity of Aqueous Extracts of *Phyllanthus amarus* in Mice and Rats

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## ABSTRACT

*Phyllanthus amarus* is a broad spectrum medicinal plant which has received world-wide recognition. However, there are concerns on the efficacy and safety of this plants' extract when used as medicinal herb. This study was therefore designed to investigate the genotoxicity of aqueous extract of *P. amarus* using the mouse micronucleus and sperm morphology assays. The potential effects of the extract on histology of the liver, kidney and testis, and blood parameters of rats were also investigated. Five concentrations: 100, 200, 400, 800 and 1600 mg/kg body weight of the extract were utilized and the test animals were orally exposed for ten consecutive days. Distilled water and cyclophosphamide were utilized as negative and positive controls respectively. Compared with the negative control, the extract induced increasing frequency of micronucleated polychromatic erythrocytes and sperm abnormalities at tested concentrations; and this was significant ($p<0.05$) at some of the tested doses. There was significant ($p<0.05$) increase in total white blood cell and lymphocyte counts; and significant pathological changes in the liver, kidney and testis of exposed rats. Tannins, resins, cardiac glycolyside and phenols were analysed in the extract. These findings suggest that aqueous extract of *P. amarus* contained constituents capable of causing systemic and DNA damage in the mouse and rat.

**Keywords**: DNA damage, Histopathology, Haematology, Micronucleus, *Phyllanthus amarus*, Sperm morphology

## INTRODUCTION

The use of plants, plant extracts or plant-derived chemicals to treat diseases is a therapeutic modality that has been explored for centuries. Over 40,000 species of tropical flowering plants are said to possess medicinal properties[1] and are currently in use for various medical conditions. It is estimated that more than 80% of the world's population utilize plants as their primary source of therapeutics[2]. Herbal medicine is generally favoured by the populace because it is believed to have low side effects and can improve the consequences of conventional agents or be a substitute treatment[3]. Therefore, herbs have significant value in current day biomedical antidotes. In a quarter of biomedical medications regularly prescribed nowadays, at least one active ingredient comes from plants and the rest of the substances are chemically produced in laboratories[4]. However, in spite of the extensive use of herbs, there is insufficient scientific evidence validating their efficacy and safety. There are possibilities of toxic effects present due to long term use and unpredictable amounts of the substance that produces the therapeutic effect especially hepatotoxic and nephrotoxic effects[5] as the liver and the kidney are the two most important organs for detoxification process in the body.

*Phyllanthus amarus* is a broad spectrum medicinal plant that has received world-wide recognition[6]. It is a small annual plant that grows 30 – 40cm in height and is found throughout the tropics and sub-tropics. In Nigeria, it is called "Iyin Olobe" in Yoruba language. The traditional preparation is usually just standard infusion or weak decoction of the whole plant or its aerial parts[7]. *Phyllanthus amarus* has been reported to possess many biological activities such as antiviral[8,9], hepatoprotective [10,11], antioxidant[12], antiarthritic[13], antimicrobial[12,14], antiallodynic[15], antitumor[16], antifertility[17], antiinflamatory[18,19], anitidiabetic [20,21], antidiarrhoeal[22], antimalarial[23], antinociceptive[24], diuretic[6], immunomodulatory[25] and antiamnesic[26].

In spite of the potential benefits, *P. amarus* has also been reported to induce cytotoxicity[27] and toxic changes in the microanatomy of liver, kidney and testes of rat[28-31]. There is however scarce report in the literature on its possible genotoxic effect. In this study, we therefore aimed at investigating the potential genotoxicity of the aqueous extract of *P. amarus* on both somatic and germ cells in mice. Additionally, we also investigated the potential toxic effects of the extract on blood parameters and selected organs in rats.

## MATERIALS AND METHODS

*Biological materials*: Young male Swiss albino mice (*Mus musculus*, 6- and 10–11-weeks old respectively) were obtained from the animal breeding unit of the Department of Zoology, University of Ibadan, Nigeria. Male Wistar rats of 6 weeks old were obtained from the

Table 1: Frequencies (mean±SE) of micronucleated polychromatic erythrocytes induced in the bone marrow of mice exposed to aqueous extract of *Phyllanthus amarus*

| Concentrations (mg/kg) | Mean ± SE |
|---|---|
| Distilled water | 0.25 ±0.25 |
| 100 | 0.50 ±0.29 |
| 200 | 1.50 ±0.87 |
| 400 | 1.50 ±0.29 |
| 800 | 2.75 ±0.85* |
| 1600 | 2.50±0.50* |
| Cyclophosphamide (20 mg/kg) | 3.75± 0.85* |

* Significant difference at $p < 0.05$

animal breeding unit of the Department of Physiology, University of Ibadan, Nigeria. The animals were kept in the experimental animal house (with 12 hours light and 12 hours darkness) in the Department of Zoology, University of Ibadan. Food (Ladokun pelleted feed®) and drinking water were supplied *ad libitum*. Animals were cared for according to standard guidelines[32].

*Plant collection, extraction and phytochemical analysis of the extract:* Fresh stem and leaves of *Phyllanthus amarus* were collected within the premises of the University of Ibadan, Nigeria. It was identified and authenticated at the University of Ibadan Herbarium (Voucher no: UIH- 22356). Stem and leaves of the plant were air dried and ground into coarse powder. The ground plant material (400g) was boiled in 4 litres of distilled water for 10 minutes and filtered with Whatman® No 1 (Maidstone, UK) (11 μm) filter paper. The filtrate was concentrated under reduced pressure at 40°C using a rotary evaporator to obtain a brown gelatinous semi-solid extract which was kept at 4°C until use. The extract was screened for the presence of tannins, anthraquinones, flavonoids, alkaloids, saponins, resin, steroids, resin, phenols and cardiac glycosides according to standard procedures[33-35].

*Micronucleus assay*: Mice of 6-8 week old (average weight = 20 g) were utilized for this assay. They were grouped into five test groups (n=4) corresponding to 100, 200, 400, 800 and 1600 mg/kg of the extract as against the negative (distilled water) and positive

(cyclophosphamide, 20 mg/kg) controls. Each mouse was orally administered 0.2 mL of the extract per day for 10 consecutive days. Bone marrow preparation for MN assessment was according to Schmid[36] and Alabi and Bakare[37]. Briefly, mice were sacrificed by cervical dislocation. The femurs were excised and bone marrow cells were flushed with 1 mL foetal bovine serum (Gibco, Brazil). Cells were centrifuged twice at 2000 rpm for 5 minutes, after which slides were prepared and stained with May-Grunwald's and 5% Giemsa stains respectively. 1000 cells were scored per animal at x1000 for the presence of micronucleated polychromated erythrocytes (MNPCEs).

*Sperm morphology assay:* Male mice of 11-15 weeks were utilized. In this assay, same number of treatment groups, type of sample and controls as the MN assay was utilized. Five mice were treated at each concentration, and 5 and 10 weeks exposure periods were considered. Five weeks exposure period was considered because spermatogenesis takes about 34.5 days to complete in mice[38] and 10 weeks exposure period was considered to investigate the possible residual effect after exposure, covering at least two spermatogenic cycles. At 5 and 10 weeks from the first day of exposure, mice were sacrificed by cervical dislocation and their caudal epididymes were surgically removed. Sperm smears were prepared from the epididymes as previously described[39,40]. For each mouse, 1000 sperm cells were assessed at x1000 for morphological abnormalities according to the criteria of Wyrobek and Bruce[41].

*Histopathology and haematology*: Male Wistar rats of ≥ 8 weeks old (weight of 100 – 160 g) were utilized. In this assay, same number of groups, type of sample, treatment and controls as the MN assay were utilized. Twenty four hours after the last exposure, peripheral blood was collected from the retro-orbital sinus and analysed for various haematological parameters using Swelab (Germany) haematology autoanalyzer. The parameters include: White blood cell (WBC), Lymphocytes (LYM), Granulocytes (GRAN), Haemoglobin (HGB), Mean corpuscular haemoglobin (MCH), Mean corpuscular haemoglobin concentration (MCHC); Red blood cell (RBC), Mean corpuscular volume (MCV), Haematocrit
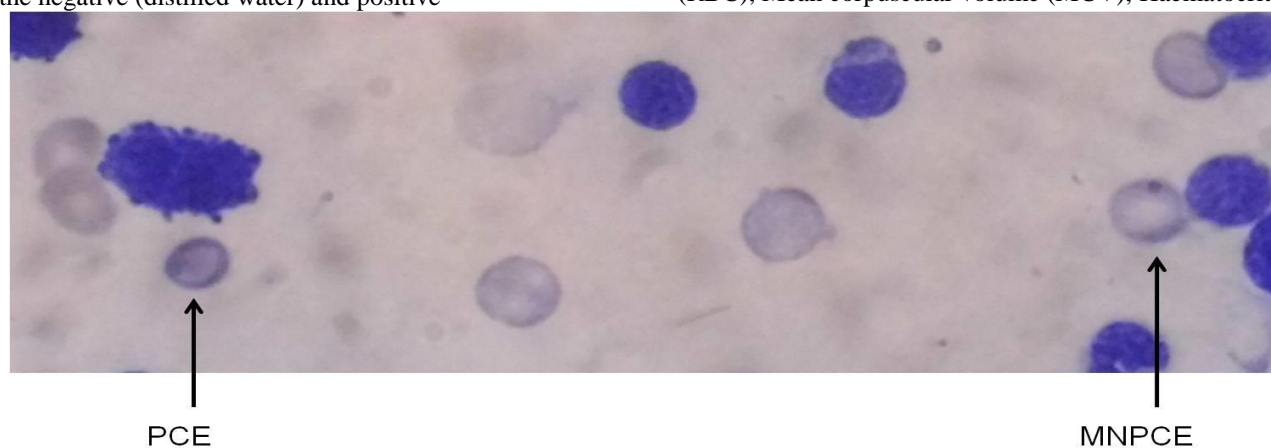


Figure 1: Micronuclei induced in mice exposed to aqueous extract of *Phyllanthus amarus*
(a) Normal polychromatic erythrocyte (PCE) (b) Micronucleated polychromatic erythrocyte (MNPCE) (x1000)

Table 2: Summary (mean ± SE) of aberrant sperm cells induced in mice exposed to different concentration of aqueous extract of *Phyllanthus amarus* after 5 and 10 weeks exposure period

| Treatment (mg/kg) | Mean ± S.E | |
|---|---|---|
| | 5 weeks | 10 weeks |
| Distilled Water | 34.00 ± 11.47 | 23.45 ± 7.88 |
| 100 | 33.60 ±14.00 | 18.00 ± 5.73 |
| 200 | 52.55 ± 26.70 | 51.70 ± 17.08[*] |
| 400 | 66.66 ± 27.46[*] | 27.70 ± 7.34 |
| 800 | 16.00 ± 5.27[**] | 15.09 ± 4.29 |
| 1600 | 21.09 ±7.02 | 31.50 ± 12.38 |
| Cyclophosphamide (20 mg/kg) | 81.27 ± 29.16[*] | 53.27 ± 17.73[*] |

* Significant increase at p<0.05; ** Significant decrease at p<0.05

(HCT), and Red cell Distribution Width (RDW).

The rats were later sacrificed by cervical dislocation and the liver, kidney and testes were surgically removed, rinsed in 0.9 % normal saline, blotted with filter paper, weighed and fixed in 10% neutral buffered formalin (liver and kidney) and Bouin fluid (testes) for histopathological analysis. The relative organ weight was calculated and expressed as g/g percent of body weight using the formula below:

Relative Organ Weight =
$$\frac{\text{Absolute organ weight (g)} \times 100}{\text{Body weight of rat on sacrifice day (g)}}$$

Serial sections of 4μm thick were obtained on glass slides using a rotatory microtome. The deparaffinized sections were stained routinely with Haematoxylin and eosin (H & E) and mounted on the microscope for analysis. Light microscopic examination of tissue sections from each organ in all groups was performed and images representative of the typical histological profile were examined.

*Statistical analysis*: Data were analyzed by SPSS® 17.0 (SPSS Inc., Chicago, IL) and expressed as mean ± SE. Statistical comparisons were performed by one-way analysis of variance (ANOVA) followed by Dunnet's test at the 0.05 probability level. Correlation analysis was performed on data of abnormal sperm cells to establish relationship between the two exposure periods.

## RESULTS

*Phytochemical analysis*

Phytochemical analysis of the aqueous extract of *P. amarus* showed the presence of tannins, resins, cardiac glycolyside and phenols.

*Micronucleus test*

Table 1 shows the results of MNPCE observed in the bone marrow of mice exposed to aqueous extract of *P. amarus*. The induced MNPCE (Figure 1) increased with increasing concentration and was significant (p<0.05) at 800 and 1600 mg/kg concentrations compared to the negative control.

*Sperm morphology test*

The effect of different concentrations of aqueous extract of *P. amarus* on sperm morphology at the end of 5 and 10 weeks exposure periods are presented in Table 2. In the five weeks exposure, the frequency of aberrant sperm cells increased with increasing concentration from 100 -
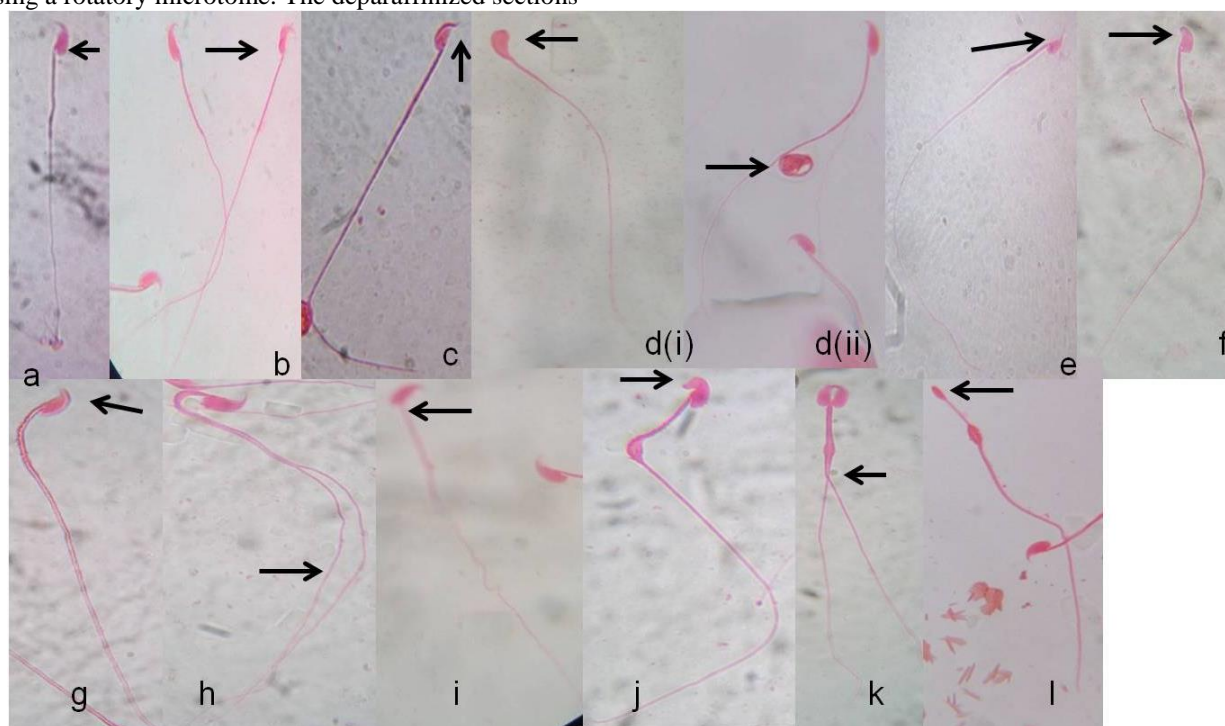


Figure 2: Abnormal sperm morphologies induced in mice exposed to aqueous extract of *Phyllanthus amarus* (a) normal head (b) banana head  (c) knobbed hook (d) i and ii folded sperm head (e) hook at wrong angle (f) No hook (g) short hook (h) double tail (i) wrong tail attachment ( j) amorphous head (k) fused sperm (l) pin head
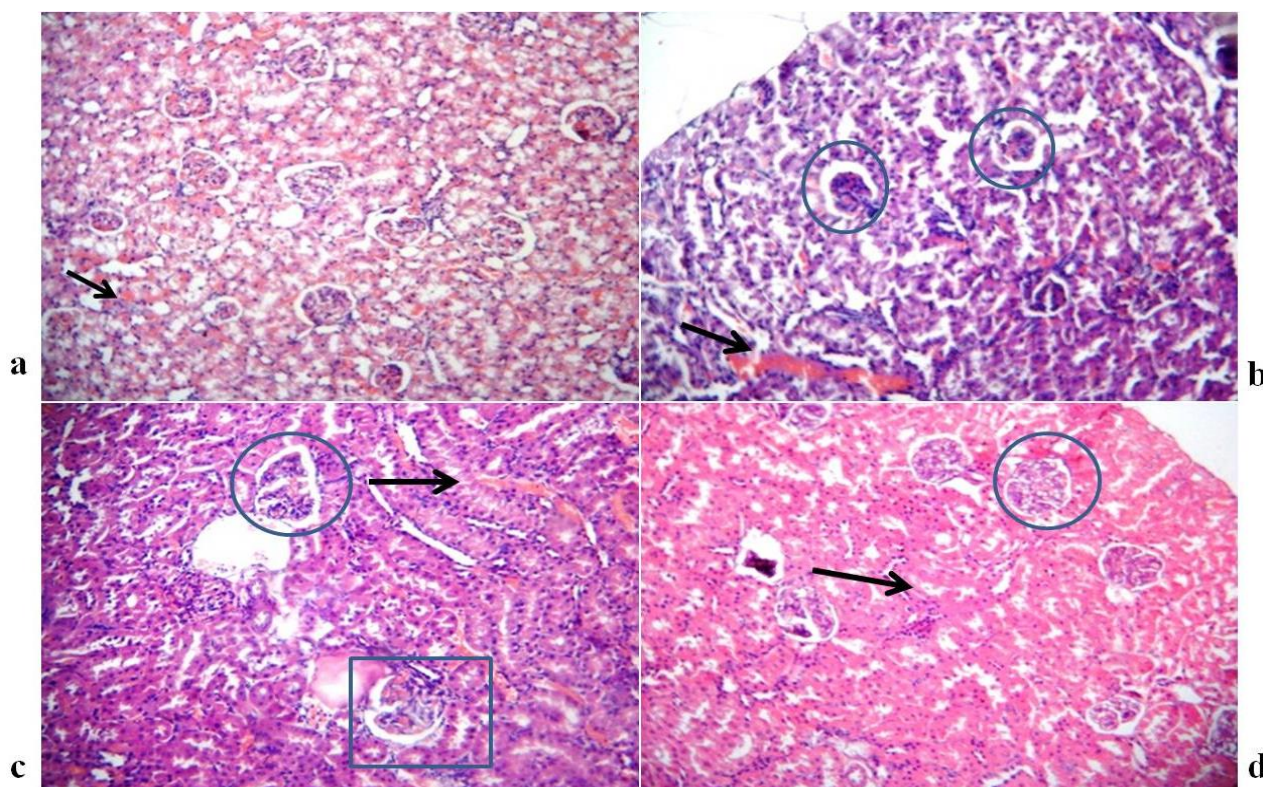
Figure 3: Renal histological analysis stained with hematoxylin and eosin (200x). Kidney with (a) no visible lesion (b) atrophic glomeruli with widened Bowman space (blue ring), congested vessels (arrow) (c) focal glomerular proliferation (ring), dilated and congested blood vessels (arrow), querry degenerate tubules (box) (d) mesangial proliferation (ring), focal tubular degeneration (arrow).

400 mg/kg, and was significant (p<0.05) at the 400 mg/kg compared to the negative control. At 800 and 1600 mg/kg, the frequency of abnormal sperm cell decreased in comparison to the other concentrations and the negative control. At the same doses for the 10 weeks exposure period, the frequency of aberrant sperm cells was not concentration dependent and was only significant at 200 mg/kg compared to the negative control. Generally, the frequency of abnormal sperm cells was lower in the 10 weeks exposure groups than the 5 week exposure groups, and was weakly correlated (r = 0.431). Figure 2 shows the different types of abnormal sperm cells observed in the exposed mice.

*Body weight and relative organ weight*
The body weight of both the controls and treated rats increased throughout the duration of the experiment. However, rats in the control group gained more weight than those in the treated groups (Table 3). The relative organ weight (kidney, liver and testes) of the treated animals were not significantly different from those of the negative control group (Table 3).

*Effect of the extract on haematological parameters*
Table 4a and 4b shows the results of the effect of *P. amarus* on haematological parameters in rats exposed for 10 days. There was a significant increase (p<0.05) in WBC and lymphocytes of exposed rats, which was not concentration dependent compared to the negative control. However, no significant difference was observed in the other parameters compared to the negative control.

*Effect of the extract on histopathology of kidney, liver and testis of rats*
Histopathological examination of sections of the kidney (Fig. 3a), liver (Fig. 4a) and testis (Fig. 5a) of rats in the control group showed normal histological features, while various pathological alterations were observed in these organs in treated rats at concentrations above 100 mg/kg. The kidney section indicated normal glomerulus, capillary vessels and tubule in the control group. The extract however induced injuries such as glomerular mesangial cell proliferation progressing to atrophy, degeneration of the renal tubules with focal necrosis (dead tissue), vascular dilation and congestion with perivascular inflammation in the rats (Figure 3b - c). In the liver of exposed rats, periportal and perivascular lymphocytic infiltration (inflammation), sinusoidal dilation and congestion, focal hepatocytes degeneration and necrosis were observed (figure 4b - c). Similarly, hypospermatogenesis and maturation arrest with focal sloughing of the germ cells in the seminiferous tubules was observed in the testes of rats exposed to *P. amarus* (Figure 5b - c).

**DISCUSSION**
Herbal medicines are widely perceived by the public as being natural, healthful and free from side effects or any potential risks due to their natural origins and are often considered as food supplements and not drugs. Medicinal herbs are usually self-prescribed by the consumers and there is a lack of control and review in terms of dose,
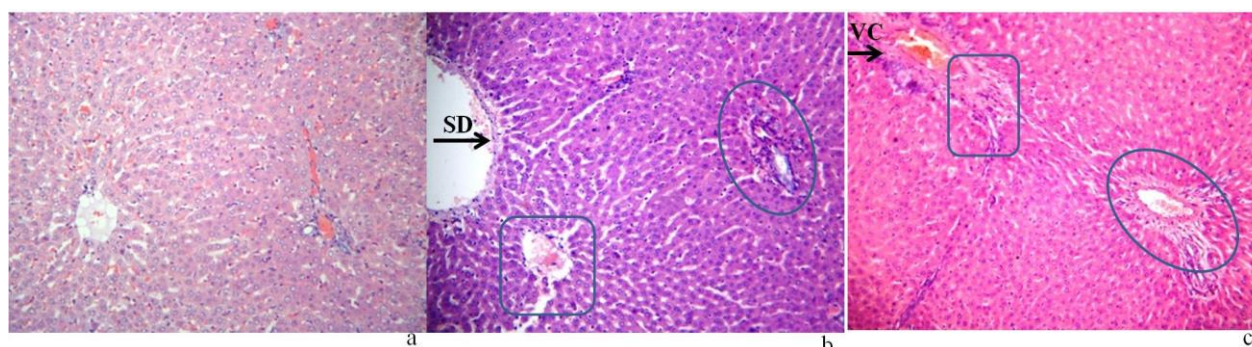
Figure 4: Hepatic histological analysis stained with hematoxylin and eosin (200x). Liver with (a) no visible lesion (b) sinusoidal dilatation (arrow), periportal inflammation, (ring), perivascular inflammation (box) (c) vascular congestion (arrow), perivascular inflammation (box), periportal inflammation (ring).
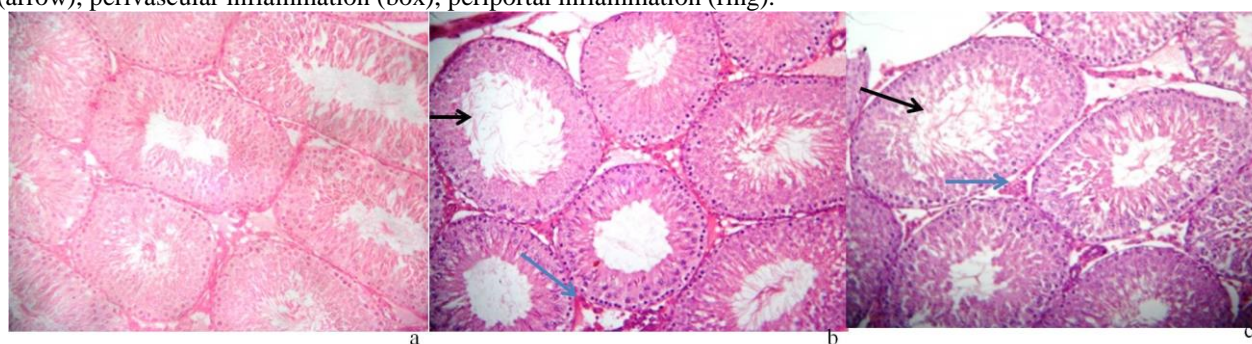


Figure 5: Testicular histological analysis stained with hematoxylin and eosin (200x).
Testes with (a) no visible lesion (b) hypospermatogenesis (black arrow), Leydig cell hyperplasia (blue arrow), (c) maturation arrest (black arrow), Leydig cell hyperplasia (blue arrow)

Table 3: Body and relative organ weights of rats exposed to aqueous extract of *Phyllanthus amarus* for 10 days

| Concentration (mg/kg) | Body weight % increase | Relative organ weight Kidney | Liver | Testes |
|---|---|---|---|---|
| Distilled water | 38.74 | 0.71±0.21 | 4.17±0.49 | 1.13±0.22 |
| 100 | 19.97 | 0.71±0.09 | 3.94±0.47 | 1.16±0.15 |
| 200 | 17.20 | 0.75±0.13 | 4.14±0.31 | 1.14±0.11 |
| 400 | 1.00 | 0.66±0.05 | 3.81±0.23 | 1.18±0.17 |
| 800 | 27.54 | 0.65±0.10 | 3.89±0.25 | 1.13±0.27 |
| 1600 | 8.21 | 0.69±0.10 | 4.09±0.50 | 1.18±0.18 |
| Cyclophosphamide (40 mg/kg) | 13.55 | 0.70±0.14 | 4.00±0.30 | 1.35±0.27 |

manner, and frequency of administration. The chemicals in medicinal herbs may be natural to the plant, but they are not natural to the human body. Plants are known to be composed of several phytochemicals which may work synergistically, additively or antagonistically[42]. The usefulness or toxic effects of the natural medicinal products typically result from combinations of various phytochemicals present in the plant[43-45]. Hence, despite their therapeutic potentials, there is a need to assess the toxicity of all medicinal plants in order to ascertain their safety. In this study, we investigated the toxicity of the aqueous extract of *P. amarus* using genetics, blood and histopathological parameters.

The data of the mouse MN assay showed that aqueous extract of *P. amarus* is genotoxic to somatic cells expecially at high concentration. The evaluation of micronuclei frequency *in vivo* is one of the primary genotoxicity test recommended internationally by regulatory agencies for product safety assessment[46]. The results herein showed that *P. amarus* contained

clastogenic constituents. This is because, in the adult rodent, the bone marrow is the hemopoietic organ. During proliferation, if a given genotoxic or clastogenic agent is administered, it may act during the cell division and cause chromosomal damage, such as breaks. The extract constituents might have also acted on macromolecules, e.g. the tubulin, causing spindle dysfunction[47]. The detection of micronuclei in this study provides a readily measurable index of chromosome breakage and loss. Increase in the frequency of MNPCEs is an indicator of genotoxic insult to the nuclei[48].

The extract induced abnormal sperm cells, and this may imply interference in the genetic process of spermatogenesis. The constituents of the extract might have modified the normal process of gametogenesis and/or the synchronization of the stages in the seminiferous epithelium[49] or cause an abnormal chromosome complement[50]. The observed abnormal sperm cells showed that the extract is capable of causing alteration in testicular DNA. This is because the

Table 4 a: Effect of aqueous extract of *Phyllanthus amarus* on haematological parameters in rats

| Conc (mg/kg) | White blood cell and its indices | | | Platelet |
|---|---|---|---|---|
| | WBC (X10$^9$/L) | LYM (X10$^9$/L) | GRAN (X10$^9$/L) | PLATELET (X10$^9$/L) |
| DDW | 6.03±0.71 | 5.03±0.61 | 0.40±0.10 | 461.00±39.66 |
| 100 | 9.47±0.55* | 7.90±0.53* | 0.73±0.23 | 286.00±171.04 |
| 200 | 10.50±1.76* | 9.27±1.77* | 0.40±0.10 | 360.67±27.50 |
| 400 | 10.53±1.56* | 8.87±1.96* | 0.57±0.12 | 336.33±18.58 |
| 800 | 9.57±1.12* | 8.13±0.86* | 0.67±0.25 | 493.00±28.69 |
| 1600 | 8.47±0.32 | 7.30±0.50 | 0.47±0.15 | 499.67±17.02 |
| CYC (40 mg/kg) | 3.63±1.33 | 2.53±0.91 | 0.67±0.45 | 755.00±31.19* |

DDW: Distilled water; CYC: Cyclophosphamide; WBC: White blood cell;
LYM: Lymphocytes; GRAN: Granulocytes; *Significant at $p<0.05$.

Table 4 b: Effect of aqueous extract of *Phyllanthus amarus* on haematological parameters in rats (Red blood cell and its indices)

| Conc (mg/kg) | HGB (g/L) | MCH (pg) | MCHC (g/dl) | RBC (X10$^{12}$g/l) | MCV(fl) | HCT (%) | RDW (%) |
|---|---|---|---|---|---|---|---|
| DDW | 14.50±0.26 | 19.27±1.22 | 31.43±0.45 | 7.55±0.43 | 61.20±4.20 | 46.13±0.67 | 17.13±0.49 |
| 100 | 14.90±0.70 | 21.37±4.80 | 30.20±4.25 | 7.14±1.16 | 63.40±11.09 | 44.43±0.81 | 20.40±5.04 |
| 200 | 14.20±1.04 | 19.77±0.55 | 31.77±0.40 | 7.18±0.55 | 62.20±1.99 | 42.00±0.69 | 17.67±2.23 |
| 400 | 14.43±0.65 | 18.87±0.25 | 33.53±1.01 | 7.63±0.24 | 47.63±8.89 | 43.07±3.04 | 15.90±0.80 |
| 800 | 13.80±0.60 | 18.07±0.76 | 31.27±0.21 | 7.64±0.56 | 57.77±2.11 | 44.10±2.05 | 17.60±0.92 |
| 1600 | 15.50±1.35 | 19.37±0.35 | 30.47±0.90 | 7.91±0.57 | 63.60±1.56 | 50.37±4.74 | 16.87±0.95 |
| CYC (40 mg/kg) | 14.37±0.49 | 19.13±0.76 | 33.16±0.49 | 7.53±0.48 | 57.67±2.66 | 43.37±0.85 | 16.20±0.62 |

DDW: Distilled water; CYC: Cyclophosphamide; HGB: Haemoglobin;
MCH: Mean corpuscular haemoglobin; MCHC: Mean corpuscular haemoglobin concentration;
RBC: Red blood cell; MCV: Mean corpuscular volume; HCT: Haematocrit;
RDW: Red cell Distribution Width.

characteristics controlling the sperm head shape are carried on the autosomes and sperm abnormality test identifies those agents[51].

Body weight alterations are indices of adverse effects of drugs and chemicals and it is significant if the body weight loss exceeds 10% from the initial body weight[52]. In this study, there was rather an increase in body weight in both the control and the treated groups. This indicates that the extract did not have adverse effects on the body weight of the exposed rats; and might also be an indication that the experimental animals were well fed. The relative organ weight is fundamental to diagnose whether the organ was exposed to injury or not[52]. The liver and kidney are primary organs affected by metabolic reactions caused by toxicants[53] while the testis is a major organ which can reflect the effect of toxicant on male reproductive system. Data herein showed that the aqueous extract of *P. amarus* did not induce any significant toxic effect on the weight of the kidneys, livers and testes of exposed rats compared to the control group.

Assessment of haematological parameters can be used to determine the extent of deleterious effect of foreign compounds such as plant extracts on the blood constituents of an animal[54] and further provides information regarding the status of bone marrow activity and haemolysis[55]. In this study, significant increase was only observed on the white blood cells and lymphocyte count of exposed rats. The increased WBC and differential leukocytes counts indicates that *P. amarus*

has phytochemicals with the ability to boost the immune system through increasing the population of defensive white blood cells. There was no significant difference in the red blood cells and their indices (MCH, MCHC, HGB, PCV and MCV). MCH, MCHC and MCV relates to individual red blood cells while Hb, RBC and PCV are associated with the total population of red blood cells[56]. This result is an indication that there was no destruction of matured RBC's and no change in the rate of production of RBCs. It further shows that the extract does not have the potential to stimulate erythropoietin release in the kidney, which is the humoral regulator of RBC production[57,58]. The non-significant effect on the RBC and HGB also implies that there was no change in the oxygen-carrying capacity of the blood and amount of oxygen delivered to the tissues following the extract administration[59]. This report is however in contrast to those of Adedapo *et al.*[28] who reported significant reduction in the RBC, PCV and HGB in rats exposed to aqueous extract of *P. amarus* for 30 consecutive days. The difference may be due to the difference in exposure duration of the two studies.

The glomerulus is the primary site of action of several chemicals and it may be injured by any toxic, metabolic and immunologic mechanism[60]. The kidney damage reported herein is in concert with earlier reports where *P. amarus* induced deleterious changes on the kidney of exposed rats[28-31]. The observed lymphatic infiltration and periportal area in the liver may be responsible for the

observed increase in WBC and probably a tissue reaction to the presence of the extract[61]. This is similar to those of Adedapo et al.[62], but contrary to the report of Lawson-Evi et al.[27], where both the aqueous and hydro-ethanol extract of *P. amarus* did not induce any pathological changes in rats exposed for twenty eight days.

Testicular degeneration involves a retrogressive change in the germinal epithelium of the seminiferous tubules[61]. In the testes of exposed rats, the observed histopathological effects may be due to impairment of hormones such as Follicle stimulating hormone (FSH) and testosterone[63] which are essential to spermatogenesis and maturation of the spermatozoa[64,65]. These could also have been possible in the testes of exposed mice, and might have been partly responsible for the induction of abnormally shaped sperm cells. Testicular degeneration in the rats and abnormal sperm morphology in the mouse suggests that aqueous extract of *P. amarus* may negatively impact male reproduction in rodents.

Crude extracts are known to be composed of complex mixtures of phytochemicals that are responsible for the benefits and adverse effects of medicinal plants. The observed toxic effects in mice and rats were due to the activities of the phytochemical components of the extract. Some naturally occurring compounds in plants such as polyphenols, alkaloids and tannins, have been implicated in causing chromosomal damage at certain concentrations[66-68]. Hence, the somatic and germ cell damage may be due to the presence of tannins and phenols in *P. amarus*. The histopathological changes in the kidneys, livers and testes of exposed rats may be due to the activities of cardiac glycosides in *P. amarus*[22]; cardiac glycosides have been implicated in the pathological and ultra structural changes in the kidney of rats[69].

In conclusion, this study showed that aqueous extract of *P. amarus* has the potential to induce genotoxicity in somatic and germ cells of mice, haematotoxicity, and histopathological changes in the kidney, liver and testes of rats. Damage to DNA may lead to critical mutations and also to an increased risk of cancer and other genetic abnormalities. It is therefore imperative to isolate the phytochemicals responsible for therapeutic effect in *P. Amarus* and those for toxic effect, as well as establish the concentrations at which these effects occur.

## ACKNOWLEDGMENTS

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