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Histopathological and Organ-Body Weight Ratio Alterations Induced by Oral Administration of AQUEOUS Extract of T*erminaliaschmiperiana* Root in Some Selected Organs in Male WISTAR Rats

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ABSTRACT: The toxic potential of the crude aqueous extract was investigated by evaluating the body-weight ratioand histologic changes of the liver, kidney and testes after oral administration of the extractto rats at various doses (1000, 2000, 3000mg/kg) daily for21 days. On the first, seventh and twenty first day,the rats werehumanely sacrificed under chloroform anesthesiaand the selected organs excised. There was no significant difference ($P<0.05$) in the kidney-body weight ratio when compared with the control group, while the administration at all the doses resulted in significant ($P<0.05$) reduction in liver-body weight ratio.An increase in testes-body weight ratio was also observed at the dose of 2000mg/kg. Histopathologicexamination of the liver, kidney and testes after 21 days of administration of the extract revealed mild changes in the architecture of the tissues.The male wistar rats did not show any treatment-related defects nor overt clinical signs of toxicity neither did the extract produce any sedative effect.

KEYWORDS: Terminalia schimperiana,Organ-body weight ratio, Histopathology, Sedative, Toxicity

I.INTRODUCTION

Histopathology examination is important for detection of any derangement in the tissue architecture of organwhich might occur as a result of the administration of certain toxicities, disease, parasitic infestation or physiological/ metabolic disorders. On the otherhand, organ-body weight ratio mayindicate cell constriction and inflammation. Medicinal plants are widely used for treatment or management of different conditions however; they contain substantial amounts of active ingredients used for their own defense against being foraged on by animals. Such active ingredients could cause adverse effects ranging from temporary physiological disorders to severe pathology states or even death.*Terminalia schimperiana* belong to the order myrtales and family combretaceae, it is known as Idi in Yoruba language. Itis a broadleaved small tree that can reach up to 7–14 m, variably deciduous in the dry season to semi-evergreen, depending on the climate. The leaves are alternate, simple, elliptic to obovate, 9–15 cm long and 3–8 cm broad, green above with pale undersides. The flowers are tiny and form pale spikes at the base of the leaves. The fruit is a samara with a single wing 6–9 cm long that turns brown with age (Arbonnier, 2004) [1]. It can be found in open forest habitats with more than 1300 mm of rainfall per year, when it is found in closed forest, it typically part of the forest canopy and it may be the dominant tree species where it is found (Jones , 1993) [2]. In parts of West Africa, *T. schimperiana* is used as a medicinal plant; the bark being applied to wounds while the twigs may be chewed to promote oral hygiene. In laboratory, experiments on extracts of the plant were found to have *in vitro*antibiotic properties against

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Staphylococcus(Akande and Hayashi, 1998) [3]. The plant extract has been found to also have antifungal properties *in vitro* (Batawila, 2005) [4]. From part of this present study,the plant was found to increase testosterone level, improve sexual motivation and performance in adult male rats. In this present study, the changes in Organ-body weight ratio and histologyof liver, kidney and testes following oral administration of aqueous extract of *Terminalia schimperiana* root wasinvestigated in male wistar rats.

II. MATERIALS AND METHODS

The roots of *Terminalia schimperiana* were collected from Oko, Irepodun local government area of Kwara state in Nigeria. Identification and authentication of plant was carried out at the botany unit of the department of pure and applied biology, Ladoke Akintola College of science and technology, Ogbomoso.The roots of *Terminalia schimperiana* plant wereharvested, cleaned, cut into fine pieces and oven- dried at 40^0C.The dried pieces were then pulverized into powder using an electric grinder. 200g of the powder was percolated in 2000ml of distilled water with constant shaking for 48hrs at room temperature. The extract was filtered with Whatman No.1 filter paper, the filtrate lyophilized and the percentage yield calculated. The resultant yield was reconstituted in distilled water to give the required doses of 1000, 2000 and 3000mg/kg body weight used in this study.Male white albino rats (*Rattus novergicus*) weighing between 250-280 g, were obtained from the Animal Holding Unit of the Department of Biochemistry, Makerere, Kampala, Uganda.

A. Experimental design: For the histopathology studies, male rats were randomly grouped into four groups of 15 animals each. One group served asa control while each of the three remaining groups was administered with one of the doses 1000, 2000, 3000 mg/kg body weight once daily for 21 days. The control received 1.0mlof the vehicle (distilled water) and was treated exactly like the test groups. The experimental rats were allowed free access to rat pellets and water after the daily dose of the extract/distilled water. The rats were sacrificed after 1, 7 and 21 days of oral administration, the selected organs removed and histopathology examinationcarried out on liver, kidney and testes.

B. Determination of Organ-body weight ratio: In order to ascertain the changes in the organ sizes after administration of the extract, the animals was weighed before sacrifice and after sacrifice, the organs of interest were excised and weighed after blotting each in tissue paper to remove blood and water (Yakubu,2006) [5]. The weights were noted and the organ-body weight ratio was calculated using the expression:

$$\text{Organ-body weight ratio} = \frac{\text{Weight of the organ}}{\text{Weight of the animal}}$$

C. Histopathology of the tissues: Following necropsy, the liver, kidneys and gonads were collected into 10% neutral buffered formalin for 72 hours. The tissues were then grossed and processed on a 13 hour 25 minutes schedule in anauto-processing machine (SLEE MTP, German). The processed tissues were embedded and thin sections (4um) made using a rotary microtome (SLEE CUT 4092, German). Sections were stained in Mayers Hematoxylin andEosinaccording to the methods described by Suvarna et al. (2013) [6] and mounted in Distrene plasticizer xylene (DPX).

D. Evaluation of sedative activity: The evaluation of the sedative activity on the Wistar rat was done using the rat hole-board technique. Randomly selected male rats were treated orally with 1ml of the extract at various dosages, three hours later the rats were placed individually on a center of rat hole-board and given a 7.5 min trial period. The number of head dips, rears, locomotive activity and the number of fecal boluses produced was recorded. The time per head drop was then calculated

(Ratnasooriya and Dharmasiri, 2000) [7].

E. Evaluation of Adverse effect: To discover the adverse effect of the extract after administration, all the treated rats were observed at least once daily for food and water intake changes and any overt signs of toxicity such as salivation, ptosis, rhinorrhea, squinted eyes, writhing, and convulsions, tremors, yellowing of fur, lachrymation and loss of hair. Stress (such as erection of fur) and changes in behavior (such as spontaneous movements in the cage, climbing, cleaning of face, non-genital self-grooming).

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F. Statistical analysis: Data was expressed as the mean of five replicates ±SD. Means was analyzed using a one-way analysis of variance (ANOVA) and complimented with student's *t-test*. Post-test analysis was carried out using Turkey's multiple comparison test to determine significant differences in all parameters. All the statistical analysis was done using IBM SPSS statistics 20 while the graphs was plotted with Excel program. Differences with values of P< 0.05 was considered statically significant.

III. RESULTS

G. Toxicity of the crude extract on Organ - body weight ratio

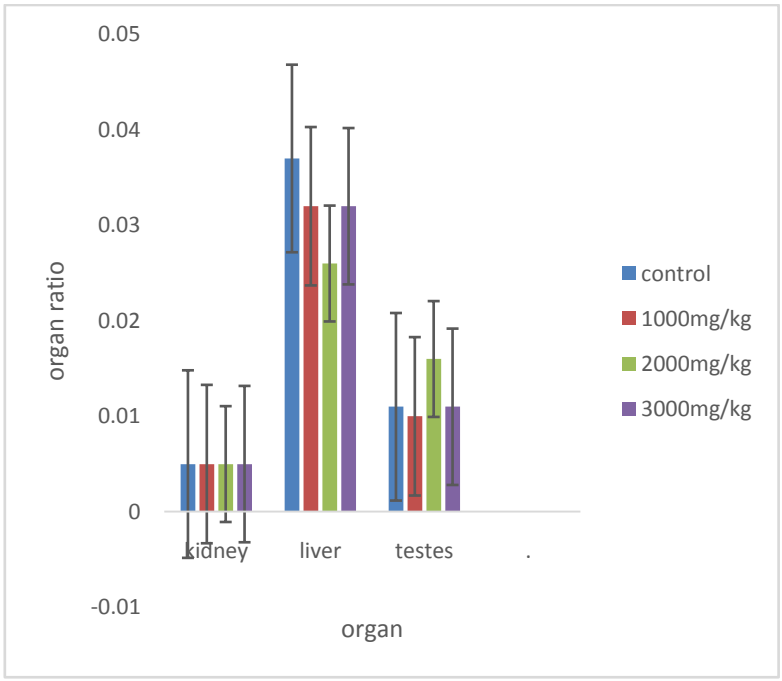

Fig 1; Effect of administration of aqueous extract of *Terminalia schimperiana* root (1000,2000, 3000mg/Kg body weight) on male rat organ -body weight ratio.

Group A (control) was orally administered with 1ml of the vehicle,groups B, C and D were orally administered with 1ml of the extract (1000,2000,3000mg/Kg body weight) respectively once daily at 24hr interval for 21 days. The organ -body weight ratio was determined by the ratio of weight of the rat before sacrifice and the weight of the organ after the sacrifice. Data is expressed as the mean of five replicates ±SD (n=5). Means were analyzed to determine significant differences in all parameters (one-way analysis of variance (ANOVA) and Turkey's multiple comparison tests). Vertical bars show standard errors of the mean. All differences are significant at P<0.05.

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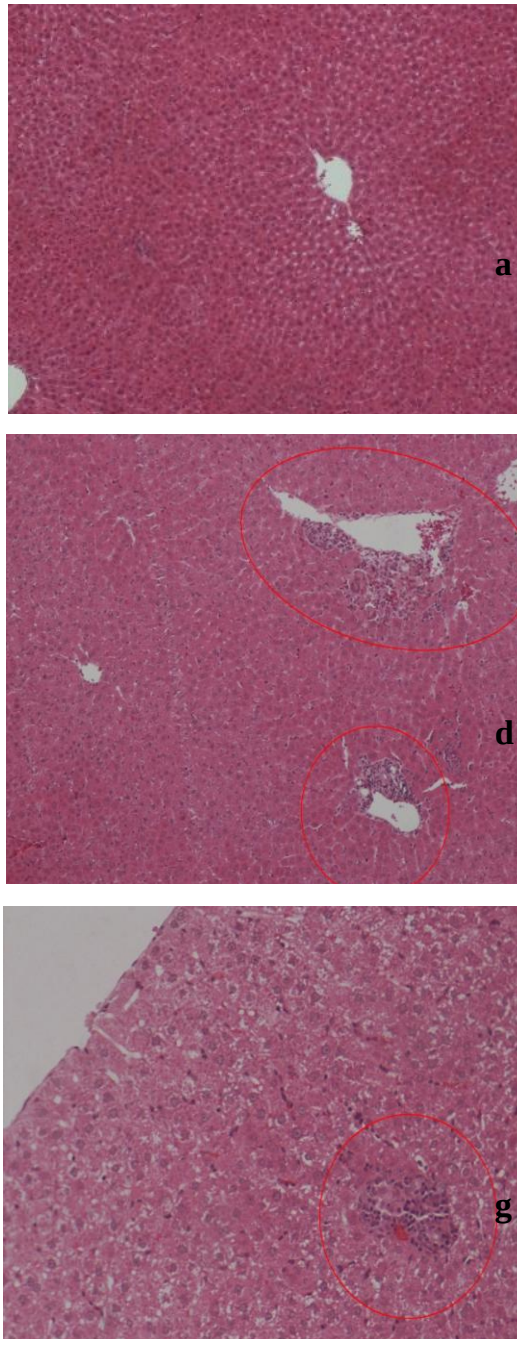
H. HISTOPATHOLOGY



Plates a- i (x10): Mild to moderate histological changes were noticed in the kidney, liver and testes as compared to the respective controls in plates **a**, **b** and **c**. In the liver, mild to moderate, perivascular mononuclear cell infiltration (plate **d**) and focal areas of necrosis (plate **g**) were seen at all doses. The kidneys were congested at low dose (1000mg/kg). At doses of 2000 and 3000mg/kg, mild hemorrhages (plate **e**) and focal renal tubular degeneration (plate **h**) was observed in the kidney sections. A dose dependent testicular atrophy was observed at all doses (plate **f**). Additionally, there was marked increase in interstitial spaces and eosinophilic cellular infiltration at doses of 2000 and 3000mg/kg (plate **i**).

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IV. DISCUSSION

Administration of *Terminaliaschimperiana* root extract at all the doses resulted in no significant difference in the kidney-body weight ratio when compared with the control group In contrast, the administration at all the doses resulted in significant reduction in liver-body weight ratio after 21daysof administration with average values of 0.032, 0.026 and 0.032 when compared with the control ratio value of 0.037 (Figure 1) this suggests that the plant extract might have caused constriction in the cells (hepatocyte).For the testes, apart from the 1000 and 3000 mg/kg body weight dosages which compared favorably with the control, 2000mg/kg dose significantly increased the ratio with ratio value of 0.016 when compared with the control ratio value of 0.011 after 21 days of administration. The increase in the testicular-body weight ratio at 2000 mg/kg dose may be attributed to increased secretory activity of the organ.From the histopathology study no significant lesions were seen in all the control and test subjects after administration of aqueous extract of *Terminalia schimperiana* root for seven days.However, mild dose-independent hepatocellular degeneration was noticed insections of the liver of all test groups after 21 days. This suggests that the prolonged administration of the extract may have adverse effects on the liver. This result further confirms the observations of the liver organ-body weight that has been discussed. Kidney is a very important organ that maintains acid-alkaline concentration of sodium and potassium in the blood and other fluids of the body. For the kidneys,No significant lesions were detected until after 21 days of administration wheremild hyaline degeneration and renal congestion was noticed in kidney when compared with the control group. Hence this supports the fact that no variation was seen in the kidney-body weight ratio between the test and control groups. Dose-dependent testicular degeneration,atrophy,interstitial thickening and eosinophillic infiltration were noticed in all test groups after 21days. (Plate 9-12). . The extract did not have sedative effect on the rats after 7.5 minutes trial with the hole-board technique. The number of head dips and rears were not significant while the locomotive activity was increased at all the doses tested.There was no significant variation in food and water intake between the treatment and control groups. No significant weight variation or treatment-related defects nor overt signs of toxicity, stress or behavioral changeswas also noticed.

V. CONCLUSION

Therefore from this study, the extract did not change the kidney body weight ratio but had effect on the liver and testes organ-body weight ratios, also the extract caused mild histological changes in the testes, liver and kidney tissues in male wistar rat probably as a result of the prolonged continuous use for 21 days.

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