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Biochemical effect of monkey tail on albino rabbits

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Abstract

Monkey tail is an addictive alcoholic drink made from fermented combo of marijuana leaves or seeds and dry/local gin. This study examined the effect of alcoholic concoction of marijuana leaves on blood chemistries of albino rabbits. Sixteen rabbits weighing 1.5 – 2.0 kg were randomly allocated into four groups and orally administered 2.5 ml/kg body weight daily of the marijuana concoction for 0, 10, 20 and 30 days respectively. There was no significant ($p>0.05$) difference in the serum total protein, albumin and globulin concentrations of the treated animals. The serum AST and ALT activities increased significantly ($p<0.05$) with increase in the duration of treatment. There were generally no significant ($p>0.05$) differences in the kidney function parameters of the animals over the treatment duration. Histological analysis of the liver and kidney tissues of the animal administered monkey tail for 30 days showed evidence of fatty liver and acute interstitial nephritis respectively. The results showed that monkey tail on prolonged consumption could have negative hepatic and renal effects in animals with mild alterations in the lipid profile.

Keywords: *Cannabis sativum*; Indian hemp; Alcoholic concoction; Toxicity

1. Introduction

Monkey tail is an alcoholic concoction of *Cannabis sativum* leaves is an addictive hard drink made from a fermented combo of Indian Hemp (Marijuana) leaves or seeds and dry gin or local gin often (Dumbili *et al* 2021). It is mostly touted to flush out typhoid and treat malaria, insomnia, weak erection, premature ejaculation, stomach ulcers, and pains, and serve as blood detox (,(Dumbili, *et al* 2021).

Marijuana-brewed beverage is not novel in Nigeria (Onyeakagbu. 2009). Marijuana is an officially – illegal – but – unofficially – legal drug. Throughout the 1980s and 1990s, monkey tail has been used as a common 'date-rape' drug in Nigeria, often served with other spirits, dessert wines, fruit juices, bits/slices of fruit and sometimes beer or palm-wine, and sweeteners. Other additives may also be added. Sometimes barbiturates were introduced to mask the sickly taste of marijuana (Nane, 2015). Monkey tail is a mainstream commercial drink whose demographics range from the inner-caucus rendezvous of the elite to the volatile "get-togethers" and "shayos" of area boys. Q Monkey tail according to Dumbili (2021) is a locally brewed cannabis and ogogoro liquor (local gin), commonly sold by skilled herbal mixologists in Nigeria.

Monkey tail acts extremely rapidly and its short-term side effects can even be experienced by a person who has drank it just once. Its long-term side effects gradually show up after repeated use, and their intensity depends upon the duration and quantity of use (Nane, 2015). The drink is believed locally to have many advantages including energy giving, improving libido and curing of many diseases such as malaria, typhoid fever, and weakness, and as well makes consumers feel "high", thereby easing their emotional stress. Given the common availability and consumption of this drink, the present study was designed to assess the potential liver and kidney toxicity of the monkey tail drink.

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2. Material and methods

2.1. Plant collection

Fresh leaves of *C. sativa* were collected from a local source at Bayelsa State. They were taxonomically identified and authenticated by Mrs. Stella Etsegbe of the Department of Plant Science and biotechnology, University of Port Harcourt, Rivers State, Nigeria with the Habarium number UPH148.

2.2. Preparation of Monkey tail

The leaves were washed with distilled water, air-dried and ground to powder. Alcoholic concoction of marijuana leaves was prepared by soaking 500g of the ground dried leaves in 1000 ml of local gin, and left to stand for 72 hours to ferment, and then filtered. The concoction was administered based on weight equivalents of the animals given that an average man (75 kg) takes a shot (44ml) of the concoction locally.

2.3. Animal Handling

Sixteen Albino rabbits (1.5-2k g) were bought from the Department of Biochemistry, University of Port Harcourt, Rivers State, Nigeria. They were acclimatized to the laboratory conditions for 7 days before proceeding with administration.

2.4. Animal Grouping

The rabbits were divided into four groups namely; "Control", "X", "Y" and "Z". Then, 2.5 ml per kg body weight of monkey tail were orally administered daily to the animals for a duration of 10, 20 and 30 days to "X", "Y" and "Z" groups respectively, while the "Control" group was fed water *ad libitum.*

2.5. Blood and organ samples collection

The rabbits were anesthesised with diethyl ether before blood samples were collected from the heart using the method postulated by (Hoofs *et al.,* 2000). Immediately after blood sample collection, the animals' liver and kidney were harvested, washed, blotted dry and stored in formalin for histological examination.

2.6. Biochemical analysis

2.6.1. Liver function profile

Serum aspartate and alanine aminotransferases (AST & ALT) activities were assayed using UV kinetic method, while total protein and albumin concentrations were estimated quantitatively using Biuret and bromocresol green (BCG) methods respectively as described with the aid of commercial kits from Rand ox Laboratories (United Kingdom). Serum globulin concentration was derived by subtracting the albumin concentration from the total protein concentration (George 2009).

2.6.2. Kidney function profile

Serum urea and creatinine concentrations were estimated by diacetyl monoxime and Jaffes methods respectively (Bonsnes *et al*., 1945) Ion selective electrode (ISE) (analyzer ISE 4000) was used for the determination of the serum concentrations of sodium, potassium and chloride as described by (Balci *et al.,* 2013). Serum uric acid concentration was estimated quantitatively by the uricase method using Agappe test kit (Agappe Diagnostics, Switzerland).

2.6.3. Lipid profile

Serum total cholesterol concentration and serum Triacylglycerol concentration were estimated quantitatively using Agappe test kit of Agappe Diagnostics (Switzerland)

2.6.4. Histological Studies of the tissues.

Histological studies was carried out on liver, kidney and heart samples, using the method described by Okoro (2002) was used with minor modification.

2.7. Statistical analysis

Data generated was analyzed using one-way analysis of variance (ANOVA) with the aid of GraphPad Prism version 5.3. Values were adjudged significant at $p \leq 0.05$.

3. Results and discussion

Monkey tail drink is a hard drink from fermented marijuana leaves or seeds and dry gin or local gin, mostly touted to flush out several diseases (Dumbili, *et al,* 2021). Toxicity due to the use of medicinal plants has been reported, and has been worsened by the fact that in ethno medicine, most of the herbal preparations are presented in an alcohol base, which may further increase toxicity due to possible synergy between alcohol and the toxic components of the plant material used (Oshilonya *et al*., 2015). Several biochemical parameters such as the levels of liver and renal function parameters, have remained major indicators of toxicity and good health and have over the years been used to measure the health status of both humans and animals (Oshilonya *et al*., 2015). Furthermore, to meet the nutritional and medicinal needs of human and animals, the appropriate nutrients and active ingredients in foods, beverages and even drugs must be provided in the right quantity. This however has been the bane of most locally produced and marketed unregulated foods and beverages (Abdeloubaheb and Amadou, 2012). This is also the case of monkey tail as no specific dosage have been shown to be applicably or standardly used by locals for a specific condition of interest. Due to inconsistency in dosage and abuse seen in the intake or consumption of monkey tail, it is therefore expedient to evaluate its effect on the health status of the liver and kidney of potential consumers.

Measurement of changes in the concentration of liver biomarkers is insightful and may therefore be a means of evaluating the toxic effects of substances (Nane, 2015). The serum chemistry parameters of albino rabbits administered alcohol concoction of marijuana leaves for 0 to 30 days shown in Figure 1 gave ranges for total protein (67.09 ± 5.58 to 74.23 ± 3.29 g/L), albumin (36.05 ± 3.40 to 40.38 ± 2.42 g/L), and globulin (31.03 ± 2.59 to 33.85 ± 2.14 g/L) with minimum values obtained at day 0 while maximum values were seen on day 30. These values were shown to have increased with time as maximum values of these parameters were observed by day 30 of treatment. However, the increase in value seen was not significantly different within the study duration.

The non-significant increase in total protein, albumin and globulin concentrations observed on continuous administration of monkey tail from day 0 to day 30 depicted that the intake of monkey tail did not affect the protein, albumin and globulin levels in serum yet by day 30, and as such does not depict an uncompromised liver. Thus, the non-significant increase observed in protein concentrations is not suggestive of possible liver dysfunction, (Oshilonya *et al*., 2015).

Figure 2 showed the serum aspartate and alanine aminotransferase (AST and ALT) activities (U/L) of the albino rabbits administered alcoholic concoction of marijuana leaves. The result revealed that AST and ALT activities (U/L) by days 0 and 10 of treatment were non-significantly different from each other, but significantly lower than those of days 20 and 30 of treatment. The activities of AST and ALT were found to be significantly ($p<0.05$) higher on day 20 through day 30. This shows that the intake of monkey tail could compromise the integrity of the liver after day 10 of continuous intake. These findings corroborated the reports of Robert *et al*. (2009) showing that toxic compounds alter the integrity of the liver. Previous reports showed that elevated levels of liver function enzymes' markers are often found in blood circulation when the integrity of the liver is compromised (Green and Flamm, 2002; Robert *et al*., 2009). Our observation may be due to possible damage caused by metabolites generated from the repeatedly administered monkey tail (Shahjahan *et al*., 2004). Goeringer *et al*. (1997) had reported that metabolites from ingested toxic substances may cause greater toxic effects to liver cells than their parent substances and may in the process also affect other organs like the kidneys, hence the observed increase in the levels of serum urea and creatinine.

Renal function parameters were variably affected in blood following intake of monkey tail. Figure 3 showed the serum creatinine concentrations on day 0 of treatment to be non-significantly higher than that observed on day 10 but lower than those of days 20 and 30. However, creatinine concentration at day 10 was shown to be significantly lower than those of days 20 and 30. On the other hand, the serum urea concentration (Figure 4) of day 0 treatment was significantly ($p<0.05$) lower than the values observed at days 10, 20 and 30, indicating increase in urea concentration with increased monkey tail dose administration. Figure 5 revealed that uric acid concentrations of albino rabbits administered monkey tail showed higher but non-significantly values when compared with the control (Day 0). However, the uric acid value of the animals administered monkey tail for 10 days was significantly higher than those of days 20 and 30. Serum sodium and chloride ion concentrations (Figures 6 & 8) of all the treatment groups were not significantly different. On the other hand, serum potassium ion concentrations (Figure 7) of the monkey tail treated animals were non-significantly higher than that of the control group expect for the 30 days treated group. For serum bicarbonate ion concentrations (Figure 9), by day 0 of treatment values obtained were not significantly different with values seen on day 10 of treatment but both were significantly ($p<0.05$) lower than values seen on days 20 through 30 of treatment. Though, the bicarbonate ion concentration obtained on day 10 was significantly lower than those of the other treatment, the values obtained for days 20 and 30 were not significantly different with each other.

The results of the present study are similar to the report of Atici *et al.* (2005) on the impact of tramadol on the renal function parameters of rats. The findings of our study also corroborated the report of Aaron *et al*. (2022) on comparative effects of *Cannabis sativa* in ethanol and tramadol, in which they noted induced similar degrees of toxicity in adult female albino rabbits. Electrolyte balance is key to the maintenance of a stable internal environment for metabolic activities. In diabetic patients for example, osmotic diuresis accompanied by water loss and electrolytes-induce glycosuria may result from electrolytes imbalance (Balci, et al, 2014). Thus, electrolytes must be carefully monitored in patients as any medication that negatively affects electrolyte balance should be discontinued as it could pose threat to the functionality of the liver and kidney.

The lipid profile of the monkey tail treated rabbits showed significantly increased total cholesterol concentrations with a non-significant reduction in the value of triacylglycerol at varying days of administration. Figure 10 showed that the serum total cholesterol concentrations (mmol/L) of day 0 treatment group to be non-significantly different with values seen at days 10, 20 and 30. However, the value obtained on day 10 was found to be significantly lower than that of day 30. Serum triacylglycerol concentrations (Figure 11) of all the treatment groups were found to be non-significantly ($p>0.05$) different. Similarly, several plants have been shown to reduce or increase lipid profile of Wistar rats. Some plant leaves have been previously reported to exhibit hypo-lipidemic effect, significantly reducing serum cholesterol (Tzu-Li *et al*., 2007), and low-density lipoprotein, while increasing high density lipoprotein (Gurrola-Diaz *et al*., 2010) reported significant decrease in plasma total cholesterol with no significant changes in triacylglycerol levels in *H. sabdariffa* extract treated animals.

The observed potential hepatorenal toxicity of monkey tail was further elucidated in the histopathology of the liver and kidney tissue sections. The monkey tail toxicity was evidenced with the presence of fatty liver (Figure 12) and acute interstitial nephritis (Figure 13) by days 20 through day 30. These observations further buttressed the earlier discussed increase observed in AST and ALT activities as well as creatinine and urea concentration of days 20 and 30 treatment groups. On the other hand, there were no observed abnormalities in the heart tissue sections (Figure 14) of all the animal groups treated with the monkey tail.

Biochemical parameters of rabbits administered alcoholic concoction of marijuana leaves.

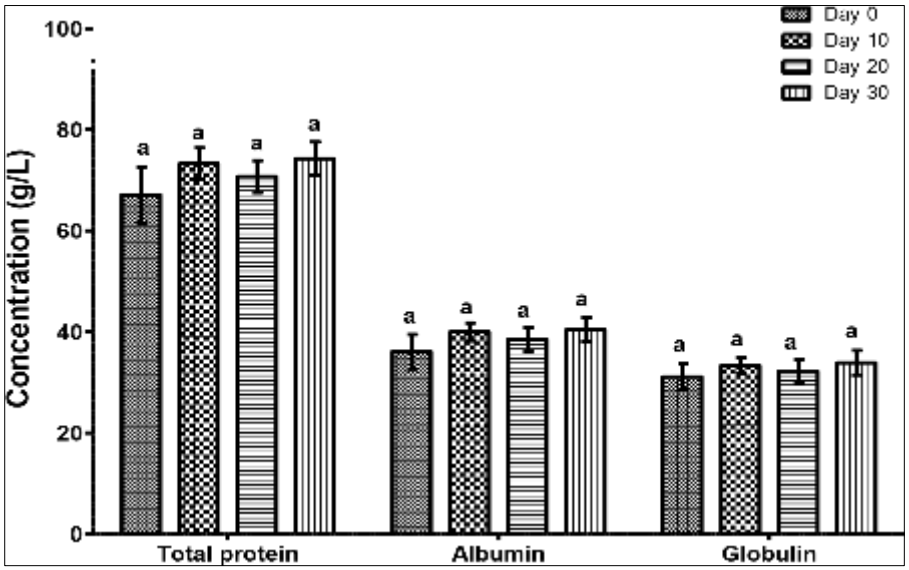

Figure 1 Serum total protein, albumin and globulin concentrations (g/L) of albino rabbits administered alcoholic concoction of marijuana leaves. Bars are mean ± standard deviation of quadruple determinations. Bars bearing similar alphabet letters per parametric group are not significantly difference ($p>0.05$)

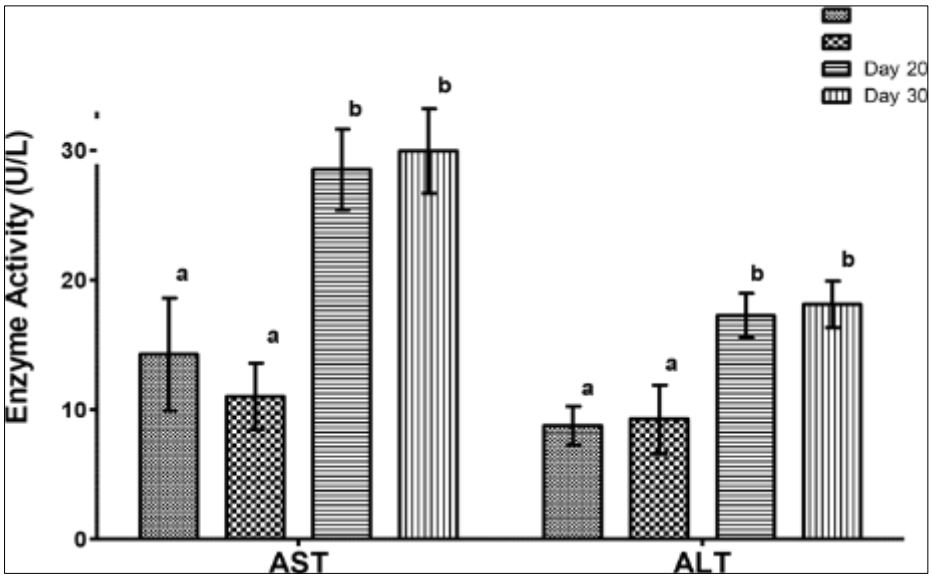

Figure 2 Serum aspartate and alanine aminotransferase (AST and ALT) activities (U/L) of albino rabbits administered alcoholic concoction of marijuana leaves. Bars are mean ± standard deviation of quadruple determinations. Bars bearing similar alphabet letters per sample group are not significantly difference (p>0.05)

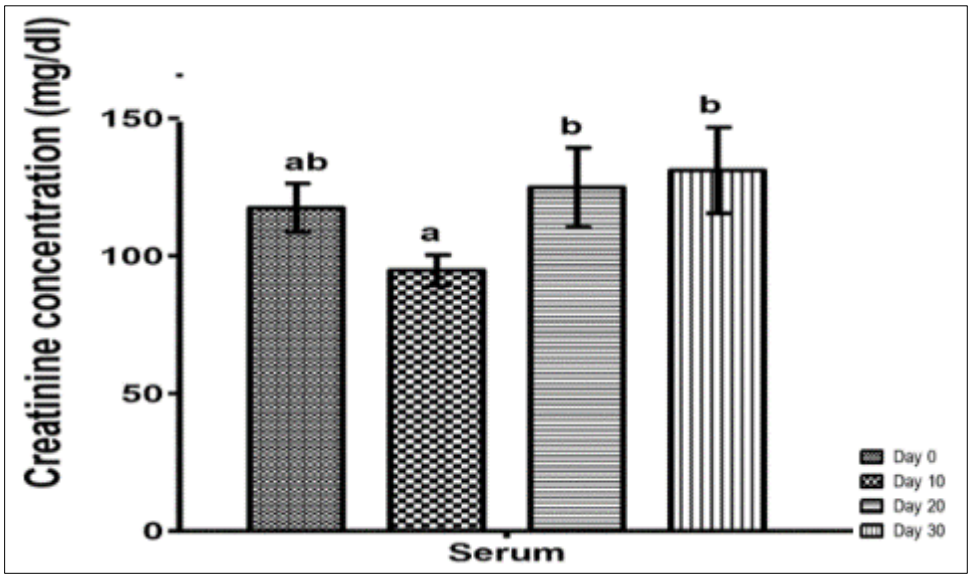

Figure 3 Serum creatinine concentrations (mg/dl) of albino rabbits administered alcoholic concoction of marijuana leaves. Bars are mean ± standard deviation of quadruple determinations. Bars bearing similar alphabet letters per sample group are not significantly difference (p>0.05)

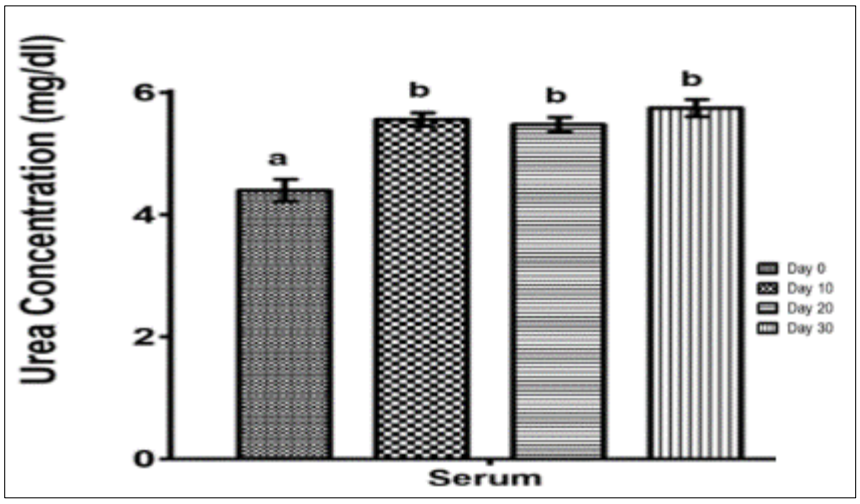

Figure 4 Serum urea concentrations (mg/dl) of albino rabbits administered alcoholic concoction of marijuana leaves. Bars are mean ± standard deviation of quadruple determinations. Bars bearing similar alphabet letters per sample group are not significantly difference (p>0.05)

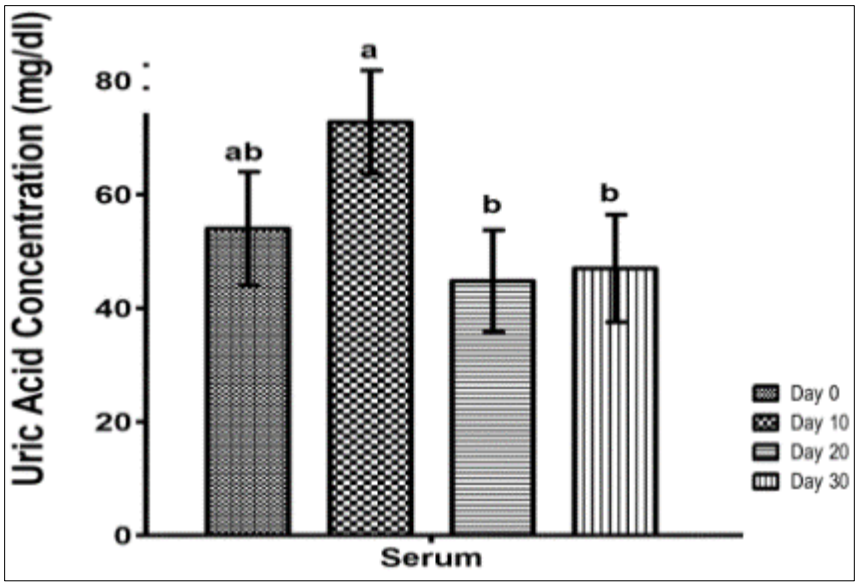

Figure 5 Serum uric acid concentrations (mg/dl) of albino rabbits administered alcoholic concoction of marijuana leaves. Bars are mean ± standard deviation of quadruple determinations. Bars bearing similar alphabet letters per sample group are not significantly difference (p>0.05)

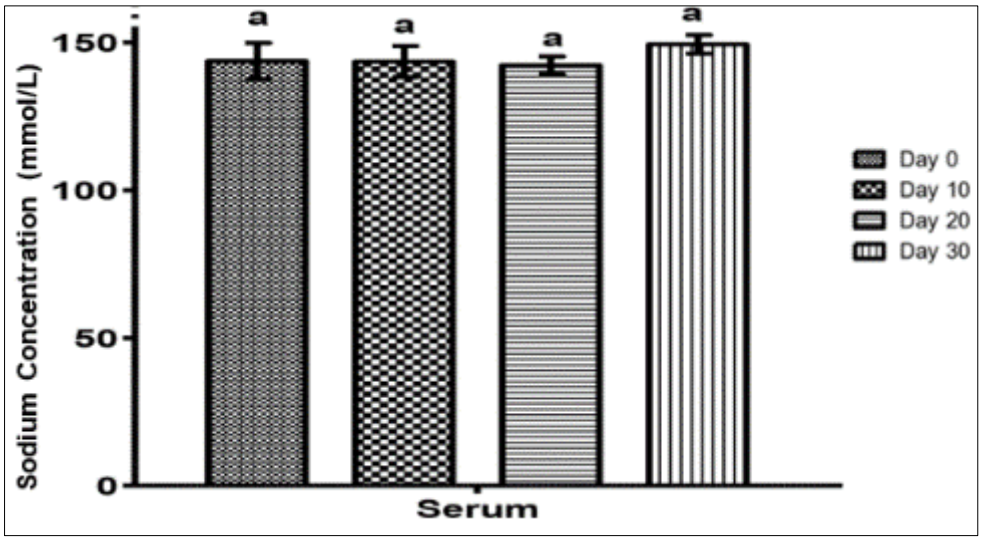

Figure 6 Serum sodium ion concentrations (mmol/L) of albino rabbits administered alcoholic concoction of marijuana leaves. Bars are mean ± standard deviation of quadruple determinations. Bars bearing similar alphabet letters per sample group are not significantly difference (p>0.05)

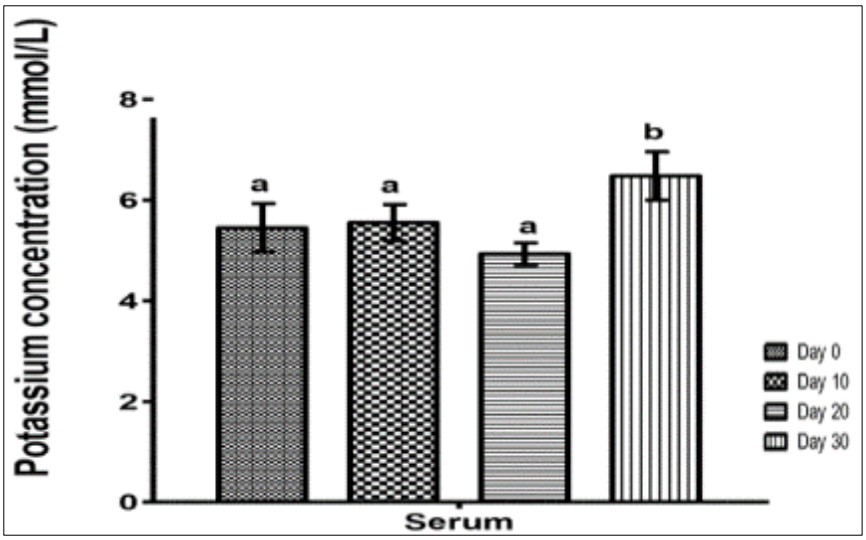

Figure 7 Serum potassium ion concentrations (mmol/L) of albino rabbits administered alcoholic concoction of marijuana leaves. Bars are mean ± standard deviation of quadruple determinations. Bars bearing similar alphabet letters per sample group are not significantly difference (p>0.05)

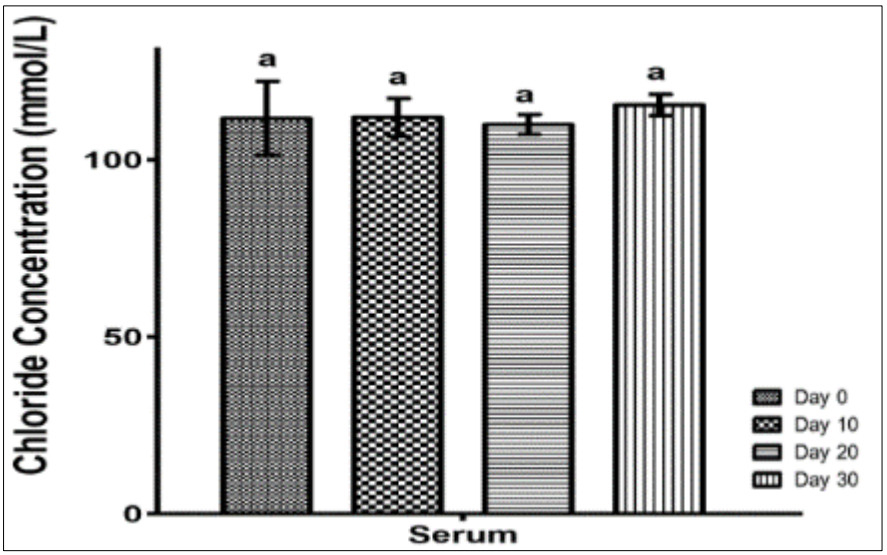

Figure 8 Serum chloride ion concentrations (mmol/L) of albino rabbits administered alcoholic concoction of marijuana leaves. Bars are mean ± standard deviation of quadruple determinations. Bars bearing similar alphabet letters per sample group are not significantly difference (p>0.05)

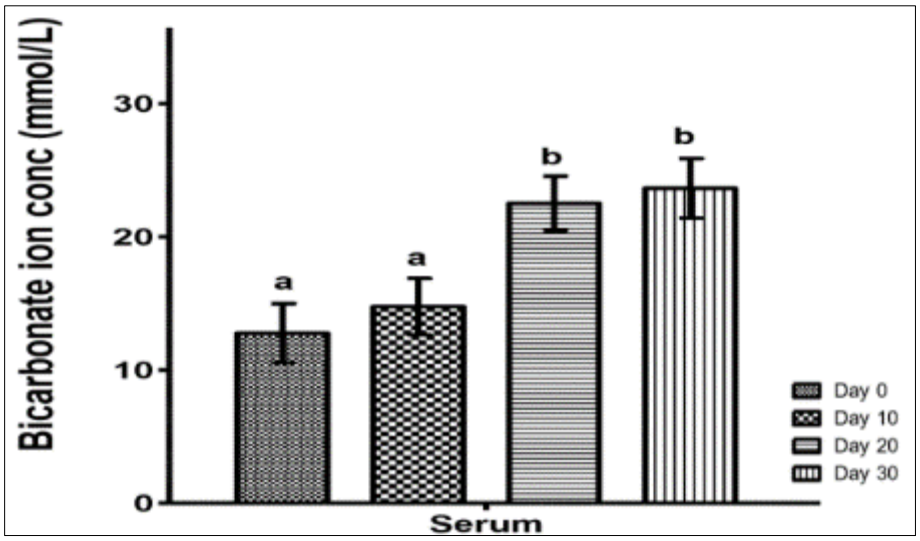

Figure 9 Serum bicarbonate ion concentrations (mmol/L) of albino rabbits administered alcoholic concoction of marijuana leaves. Bars are mean ± standard deviation of quadruple determinations. Bars bearing similar alphabet letters per sample group are not significantly difference (p>0.05)

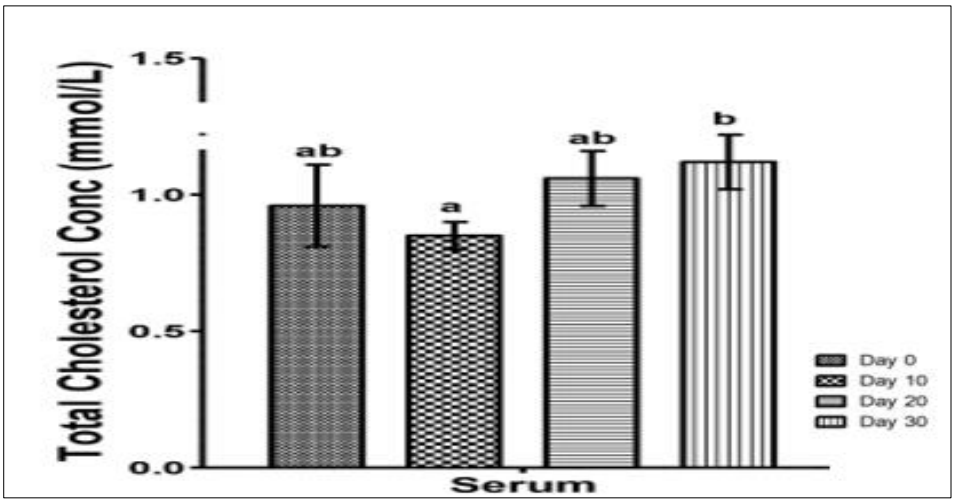

Figure 10 Serum total cholesterol concentrations (mmol/L) of albino rabbits administered alcoholic concoction of marijuana leaves. Bars are mean ± standard deviation of quadruple determinations. Bars bearing similar alphabet letters per sample group are not significantly difference (p>0.05

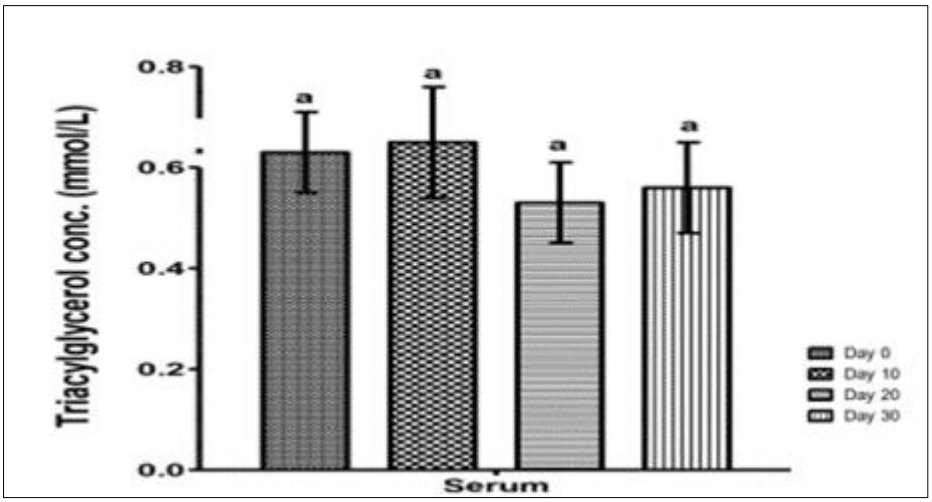

Figure 11 Serum triacylglycerol concentrations (mmol/L) of albino rabbits administered alcoholic concoction of marijuana leaves. Bars are mean ± standard deviation of quadruple determinations. Bars bearing similar alphabet letters per sample group are not significantly difference (p>0.05)

3.1. Histopathology of Wister Rabbits administered monkey tail.

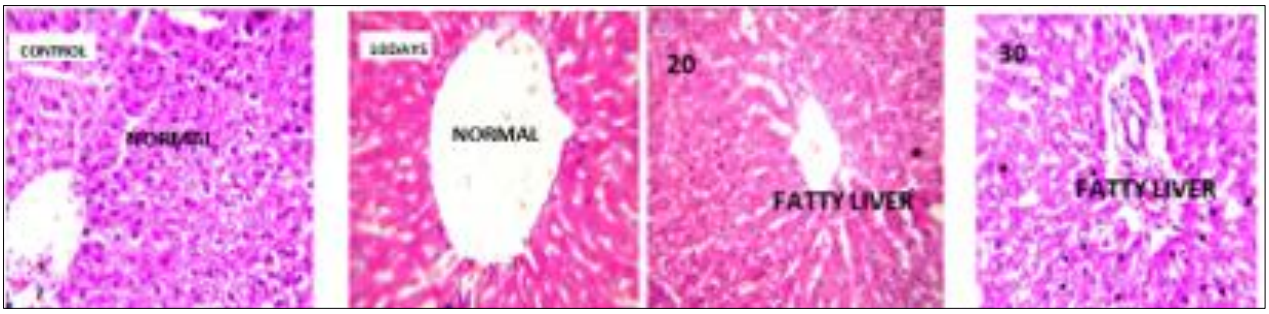

Figure 12 Transverse section of the liver tissue of rabbits administered alcoholic concoction of marijuana leaves

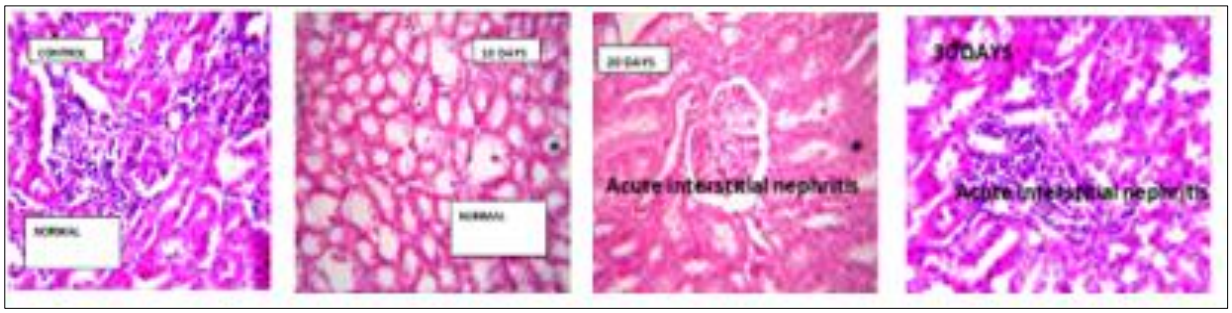

Figure 13 Transverse section of the kidney tissue of rabbits administered alcoholic concoction of marijuana leaves

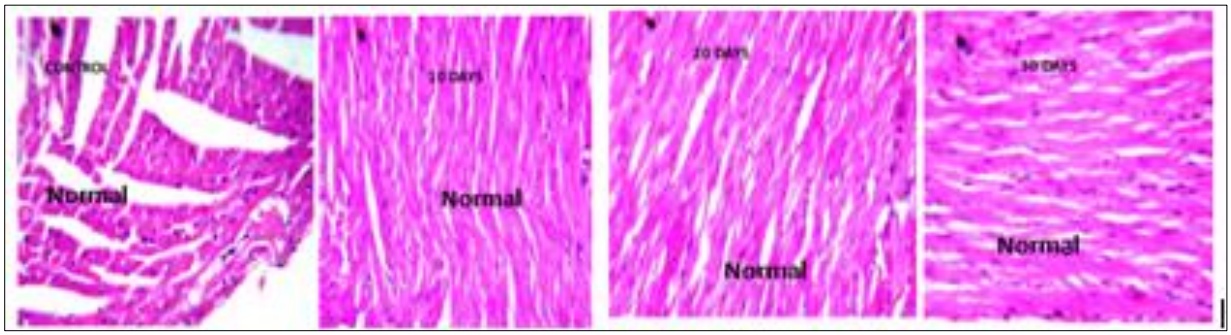

Figure 14 Transverse section of the heart tissue of rabbits administered alcoholic concoction of marijuana leaves

4. Conclusion

Results of our study showed that prolonged administration of monkey tail for up to 20 to 30 days could have significant hepatorenal toxic effect on consumers given the observed effects on ALT, AST, creatinine, urea and other related toxicity marked studied. The observed hepatorenal toxicity was further strengthened with results of the histological examination of the liver and kidney tissue sections, with monkey tail toxicity evidenced in presence of fatty liver and acute interstitial nephritis following prolonged intake. However, the cardiac tissues were seen not to be histologically prone to toxicity effect within this study period. This study in totality revealed that monkey tail drink intake continuous for a duration beyond 10 days could be significantly toxic to the kidney and liver organs. Hence, it is advised that monkey tail's usage for any reason, should be discontinued after 10 days of exposure.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to disclosed.

Statement of ethical approval

This study was ethically approved by the ethical unit of federal University of Technology Owerri.

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