

Hepatoprotective Potential of *Cnidoscolusaconitifolius* Leaf Extract on CCl₄ Treated Wistar Rats

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Abstract: Hepatoprotective potential of *Cnidoscolusaconitifolius* leaf extract on carbon tetrachloride treated wistar rats was investigated. Forty two adult male wistar rats (91-185g) were housed in plastic cages of seven groups with six animals in each. Group one fed with normal rat chow (control). Group two to six were administered CCl₄ subcutaneously on day twenty-one. Group three was administered vitamin C orally on daily basis for twenty-one days, group four, five, six took 50mg/kgbw, 75mg/kgbw and 100mg/kgbw extract dose orally for twenty-one day before administration of CCl₄. Group seven was given subcutaneous dose of olive oil. All the analysis were done using standard methods. In comparison to the disease control and normal control animals, administration of the extract dose (50mg/kgbw, 75mg/kgbw and 100mg/kgbw) dependently lowered creatinine, albumin, calcium, potassium, ALT, AST and total bilirubin. There was no significant difference (p<0.05) in the magnesium concentrations of rats treated with the leaf extract of *C.aconitifolius*. The 50mg/kgbw and 75mg/kgbw extract doses provided better protection. Histopathological examination of the liver showed histological normal liver in normal control rats and distortion of the liver sections in disease control rats which was corrected by the extracts at 50mg/kgbw and 75mg/kgbw doses. The result of this study clearly demonstrated the hepatoprotective effect of *Cnidoscolusaconitifolius* leaf extract against carbon tetrachloride induced hepatotoxicity in rats.

Keywords: *Cnidoscolusaconitifolius*, Hepatoprotective, wistar rats, leaf extracts, Carbon TetraChloride (CCl₄).

I. INTRODUCTION

The use of traditional medicines and medicinal plants has been widely observed in most developing countries, where they are seen as therapeutic agents for the maintenance of good health [1]. For several decades, various fields of research have centered on medicinal plants and their components. There is an increasing interest and demand in research concerning liver diseases in recent years around the World. Liver disease is a global burden and it is among the leading cause of deaths and illness globally [2-3]. The high rate of ingestion of chemicals and substances in form of drugs and herbs poses danger to the body especially the liver. The quest for alternative approach to prevention of hepatotoxic diseases from natural plant sources is on the increase. This has given rise to the need to investigate the claims on use of *Cnidoscolusaconitifolius* leaf extract scientifically for its pharmacological importance.

1.1 Aim

This study investigated the hepatoprotective potential of *Cnidoscolusaconitifolius* leaf extract on carbon tetrachloride treated wistar rats.

1.2 Objectives

The specific objectives are;

- i. Proximate analysis of the leaf extract.
- ii. Evaluation of liver function test in the experimental rats.
- iii. Renal function analysis in the experimental rats.
- iv. Histopathological analysis on the liver of the experimental rats.

1.3 Significance of study

This study will benefit the masses through the result of its investigation and may suggest the possibility of *Cnidoscolusaconitifolius* leaf extract in treating liver diseases. It will help to reduce the over-reliance on drugs and chemicals for treatment and management of liver diseases and in the process protect the liver from unnecessary damage from toxicity.

1.4 *Cnidoscolusaconitifolius*

Cnidoscolusaconitifolius is an interesting tropical plant that can be relevant to different communities because of its numerous potentials when consumed. Yet, it remains under appreciated [4-6]. This review highlights available scientific information on the nutritional relevance and health importance of this plant. *C. aconitifolius* belongs to the family of Euphorbiaceae and is called different names by different tribes according to its diverse traditional uses. It is popularly called 'EfoIyanaIpaja' or 'Efo Jerusalem' in the Southwestern part of Nigeria, 'Hospital Too Far' in the Niger Delta region of Nigeria, 'tree spinach' in English and colloquially as 'Chaya' [7-10].

According to Ihekoronye and Ngoddy[11], vegetables are abundant during rainy season in Nigeria and some African countries but become scarce towards the end of the rainy season and even more during the dry season. *C. aconitifolius* is very easy to grow with high yields that can serve as a good

source of phyto nutrients all year round particularly in the rural areas where people have difficulty to access good supplements [4-6]. It is worthy to note that *C. aconitifolius* can grow well in diverse environmental conditions both in the rainy and dry regions with little relevance for manure and care [5].



Plate 2.1: Cnidoscolumaconitifolius's leaf

II. MATERIALS AND METHODS

2.1 Method of Leaf Extraction

C. aconitifolius leaves were collected fresh, washed, pounded with a mortar and pestle and pressed for aqueous extraction [12]. Suction filtration (vacuum) was employed on the extract and the process repeated severally for decolouration to obtain a soluble compound. The total extract of the leaves was freeze dried at the pharmacognosy laboratory of the Faculty of Pharmaceutical Sciences. The residue obtained was measured and used for analysis.

2.2 Proximate Analysis

Primary components of *C. aconitifolius* leaves were proximately analyzed for moisture content, ash, crude fibre, crude protein, lipids and carbohydrates using the AOAC 1990 methods for the determination of proximate analysis of food samples.

2.3 Experimental Design

Forty two adult male wistar rats weighing 91-185g were purchased from the Animal House of the Department of Physiology University of Port Harcourt. The animals were housed in plastic cages of seven groups with six animals each per group. After one-week acclimatization period on normal rat chow, the treatment commenced. The extract was administered orally on daily basis for twenty one days. The dosage of administration of the extract was adapted, with modification, from Ikewuchi and Ikewuchi[14] and Ikewuchi[15]. The carbon tetrachloride was prepared in a ratio of 1:5 (v: v) in olive oil, and administered subcutaneously at 0.17 mL/kg body weight of rats, on day twenty one after administration of leaf extract. The dosage and method of administration of carbon tetrachloride was adapted from Obi and Uneh[16], with modification. The experimental groups are as follow;

Group I: received daily normal feed and water.

Group II: received daily normal feed and water + subcutaneous dose of CCl₄ (0.17mg/kg) on day twenty one.

Group III: received daily normal feed and water + daily oral dose of Vitamin C + subcutaneous dose of CCl₄ (0.17mg/kg) on day twenty one.

Group IV: received daily normal feed and water + daily oral dose of *C. aconitifolius* extract (50mg/kg) + subcutaneous dose of CCl₄ (0.17mg/kg) on day twenty one.

Group V: received daily normal feed and water + daily oral dose of *C. aconitifolius* extract (75mg/kg) + subcutaneous dose of CCl₄ (0.17mg/kg) on day twenty one.

Group VI: received daily normal feed and water + daily oral dose of *C. aconitifolius* extract (100mg/kg) + subcutaneous dose of CCl₄ (0.17mg/kg) on day twenty one.

Group VII: received daily normal feed and water + subcutaneous dose of olive oil.

Twenty four hours after administration of carbon tetrachloride, the rats were weighed and anaesthetized by exposure to chloroform. While under anesthesia, they were painlessly sacrificed after 8hrs fasting and blood was collected from each rat into heparin sample bottles via cardiac puncture for biochemical and hematological analyses. The liver organs were harvested afterwards and preserved in 10% formalin, for histochemical analysis. The heparin anti-coagulated blood samples were centrifuged at 1000rpm for 10 minutes, after which the plasma was collected and stored for subsequent analysis.

2.4 Biochemical Parameters

The kits used for the determination of potassium, Chloride, Sodium and magnesium concentrations are products of Boditech Med Incorporated, Korea. The kits used for the determination of liver function analysis and renal function analysis are products of Randox Laboratories Ltd., 55 Diamond Road, Crumlin, Co. Antrim, United Kingdom.

2.5 Determination of percentage protection (% protection)

The percentage protection provided by the leaf extract of *Cnidoscolumaconitifolius* against carbon tetrachloride induced liver damage was calculated using the following formula adapted from Al-Qarawiet *al.* (2004).

$$\% \text{ protection} = \frac{\text{Parameter}_{\text{test control}} - \text{Parameter}_{\text{treatment}}}{\text{Parameter}_{\text{test control}} - \text{Parameter}_{\text{control}}} \times 100$$

2.6 Histopathological examination

The liver organs were harvested and kept in a 10% neutral formalin solution for fixation. After 1 week, the organs were washed with water from the tap, afterwards, ethanol of ascending grades was used for dehydration, and then xylene embedded and finally put in paraffin. 4-5µm thickness was obtained while eosin and haematoxylin were used for staining reaction. The set up was examined under a microscope.

2.7 Method of data analysis

Statistical Package for Biological and Social Sciences (SPSS) Inc. 21.0 Software program was used. Mean values (M) ± SD were calculated and one-way analysis of variance (ANOVA) test was performed. Values (P) that was less than 0.05 (P<0.05) was considered statistically significant.

III. RESULTS

3.1 Proximate analysis

The proximate content of *Cnidoscopusaconitifolius* leaves recorded in Table 3.1 shows that the leaves had 82.11 ± 0.85% moisture, 1.38 ± 0.05% ash, 2.80 ± 0.02% lipid, 0.51 ± 0.01% crude protein, 12.34 ± 0.77% crude fibre and 0.86 ± 0.03% Carbohydrate. The result of the proximate analysis of leaf extract of *Cnidoscopusaconitifolius* revealed that the leaves had moisture 82.11 ± 0.85%, which was the highest while crude protein which was 0.51 ± 0.01% was the lowest.

Table 3.1: Proximate analysis of *Cnidoscopusaconitifolius* leaves

| Proximate content | (%) |
|-------------------|--------------|
| Moisture | 82.11 ± 0.85 |
| Ash | 1.38 ± 0.05 |
| Lipid | 2.80 ± 0.02 |
| Crude protein | 0.51 ± 0.01 |
| Crude fibre | 12.34 ± 0.77 |
| Carbohydrate | 0.86 ± 0.03 |

Values are Mean ± SD of triplicate determinations. n=3.

3.2 Biochemical parameters

3.2.1 Kidney function parameters

Table 3.2 shows the concentrations of creatinine and albumin while Table 3.3 shows the concentrations of chloride, potassium, magnesium, calcium and sodium in normal rats and rats administered with CCl₄ and treated with leaf extract of *Cnidoscopusaconitifolius*.

Table 3.2 shows that creatinine concentration was highest and lowest at groups VI (598.50 ± 2.81) µmol/l and IV (178.13 ± 0.61) µmol/l, while albumin concentration was highest at group II (34.47 ± 6.01) g/l and lowest at group VII (5.90 ± 1.77) g/l. There was significant increase (P<0.05) in the creatinine level of rats administered with CCl₄ when compared to the normal. Creatinine levels of treated rats at 50mg/kgbw and 75mg/kgbw dose were decreased significantly when compared to normal and diseased control respectively. There was significant increase in creatinine level at 100mg/kgbw when compared to both the normal and disease control. There was significant increase in the albumin level of rats administered with CCl₄ when compared to the normal and significant decrease in the albumin levels of the treated rats at all concentrations when compared to both normal and disease control.

Table 3.3 shows that the plasma electrolytes was highest and lowest as follows; chloride groups VII (102.17 ± 19.85)

mmol/l and VI (84.63 ± 28.82) mmol/l, potassium groups II (4.37 ± 0.82) mmol/l and IV (2.48 ± 0.71) mmol/l, magnesium groups II (2.20 ± 0.11) mg/dl and III (1.85 ± 0.49) mg/dl, calcium groups I (1.94 ± 0.09) mmol/l and IV (1.63 ± 0.28) mmol/l and sodium groups V (133.76 ± 7.22) mEq/l and I (106.50 ± 13.67) mEq/l respectively. There was significant decrease (P<0.05) in the calcium concentration of rats treated with 50mg/kgbw extract when compared to the normal. There was significant increase in potassium concentration of rats administered with CCl₄ when compared to normal. Also, there were significant increases (P<0.05) in the sodium concentration of rats administered with CCl₄ and the treated rats at all concentrations when compared to the normal.

Table 3.2: Effects of the leaf extract of *Cnidoscopusaconitifolius* on some kidney parameters of normal and CCl₄ induced hepatotoxicity in rats.

| Groups | Creatinine concentration (µmol/l) | Albumin concentration (g/l) |
|----------------------------------|-----------------------------------|-----------------------------|
| Group I (Normal control) | 324.90 ± 0.99 ^a | 6.74 ± 0.77 ^b |
| Group II (Disease control) | 491.63 ± 3.67 ^b | 34.47 ± 6.01 ^a |
| Group III (Vitamin c) | 313.50 ± 1.70 ^a | 33.83 ± 4.29 ^a |
| Group IV (50mg/kg b. w extract) | 178.13 ± 0.61 ^c | 24.75 ± 2.08 ^c |
| Group V (75mg/kg b. w extract) | 334.88 ± 1.19 ^a | 25.54 ± 5.87 ^c |
| Group VI (100mg/kg b. w extract) | 598.50 ± 2.81 ^d | 25.45 ± 4.03 ^c |
| Group VII (Olive oil) | 342.00 ± 1.46 ^a | 5.90 ± 1.77 ^b |

Values are Mean ± SD, n=6. Values in the same column bearing same superscript letters show no significant differences between the groups while those with different superscript letters (a,b,c...) show significant differences between the groups (P<0.05). mg/kg b. w = mg/kg body weight.

Table 3.3: Effects of the leaf extract of *C. aconitifolius* on the plasma electrolyte profiles of normal and CCl₄-induced hepatotoxicity in rats

| Groups | Electrolyte profiles | | | | |
|----------------------------|---------------------------------|----------------------------------|---------------------------------|--------------------------------|------------------------------|
| | Chloride concentration (mmol/l) | Potassium concentration (mmol/l) | Magnesium concentration (mg/dl) | Calcium concentration (mmol/l) | Sodium concentration (mEq/l) |
| Group I (Normal control) | 92.56 ± 16.58 ^a | 2.55 ± 0.43 ^a | 1.91 ± 0.49 ^a | 1.94 ± 0.09 ^a | 106.50 ± 13.67 ^a |
| Group II (Disease control) | 100.59 ± 13.96 ^a | 4.37 ± 0.82 ^b | 2.20 ± 0.11 ^a | 1.77 ± 0.22 ^a | 125.24 ± 4.16 ^b |
| Group III (vitamin c) | 93.24 ± 27.49 ^a | 2.99 ± 0.55 ^a | 1.85 ± 0.49 ^a | 1.84 ± 0.14 ^a | 131.07 ± 12.08 ^b |

| | | | | | |
|-------------------------------------|-----------------------------|--------------------------|--------------------------|--------------------------|-----------------------------|
| Group IV (50mg/kg b. w extract) | 91.00 ± 22.81 ^a | 2.48 ± 0.71 ^a | 2.07 ± 0.22 ^a | 1.63 ± 0.28 ^b | 127.54 ± 14.78 ^b |
| Group V (75mg/kg b. w extract) | 90.55 ± 20.66 ^a | 2.67 ± 0.52 ^a | 1.99 ± 0.26 ^a | 1.73 ± 0.13 ^a | 133.76 ± 7.22 ^b |
| Group VI (100mg/kg b. w extract) | 84.63 ± 28.82 ^a | 3.64 ± 2.04 ^a | 2.08 ± 0.24 ^a | 1.66 ± 0.19 ^a | 124.72 ± 23.48 ^b |
| Group VII (Olive oil) | 102.17 ± 19.85 ^a | 3.05 ± 1.51 ^a | 2.18 ± 0.11 ^a | 1.87 ± 0.49 ^a | 129.00 ± 7.94 ^b |

Values are Mean ± SD, n=6. Values in the same column bearing same superscript letters show no significant differences between the groups while those with different superscript letters (a,b,...) show significant differences between the groups (P<0.05). mg/kg b. w = mg/kg body weight.

3.2.2 Liver function tests

Table 3.4 shows the concentrations of alanine transaminase, aspartate transaminase, alkaline phosphatase, total bilirubin and total protein in normal rats and rats administered with CCl₄ and treated with the leaf extract of *Cnidoscolumaconitifolius*. Table 3.3 shows the effect of the leaf extract on the plasma hepatospecific markers of normal and CCl₄ –induced hepatotoxicity in rats. The parameters ranged as; ALT group V (956.80 ± 158.57) – group VI (1238.53 ± 184.95) μ/l, AST group III (686.67 ± 168.80) – group VII (2441.39 ± 168.26) μ/l, ALP group IV (19.47 ± 7.43) – group II (33.73 ± 6.92) μ/l, Total bilirubin group I (1.26 ± 0.57) – group II (6.85 ± 1.74) μmol/l and Total protein group VII (25.57 ± 5.08) – group VI (57.14 ± 6.57) g/l. There was significant increase (P<0.05) in AST activity of rats administered with CCl₄ when compared to the normal. Also, there were significant decreases (P<0.05) in the AST activity of rats treated with 50mg/kgbw and 75mg/kgbw extract when compared to both normal and disease control while group III showed significant decrease (P<0.05) in the AST levels when compared to disease control. There was significant decrease (P<0.05) in the ALP activity of rats treated with 50mg/kgbw extract when compared to both normal and disease control. There was significant increase (P<0.05) in total bilirubin concentration of rats administered with CCl₄ when compared to the normal. Also, there was significant decreases (P<0.05) in the total bilirubin concentration of rats treated with 50mg/kgbw and 75mg/kgbw extract as well as those

administered with antioxidant when compared to disease control while rats treated with 100mg/kgbw extracts showed significant decrease (P<0.05) in the total bilirubin levels when compared to both normal and disease control. There was significant increase (P<0.05) in total protein concentration of rats administered with CCl₄ and rats treated with the extract at all concentrations when compared to the normal.

Table 3.4: Effect of the leaf extract of *Cnidoscolumaconitifolius* on the plasma hepatospecific markers of normal and CCl₄-induced hepatotoxicity in rats

| Groups | LIVER FUNCTION INDICATORS | | | | |
|----------------------------------|-------------------------------|-------------------------------|----------------------------|--|-----------------------------------|
| | ALT activity (μ/l) | AST activity (μ/l) | ALP activity (μ/l) | Total bilirubin concentration (μmol/l) | Total protein concentration (g/l) |
| Group I (Normal control) | 990.24 ± 104.42 ^a | 691.60 ± 142.80 ^a | 32.57 ± 9.95 ^a | 1.26 ± 0.57 ^a | 37.79 ± 3.43 ^a |
| Group II (Disease control) | 1118.93 ± 44.89 ^a | 886.67 ± 208.14 ^b | 33.73 ± 6.92 ^a | 6.85 ± 1.74 ^b | 53.31 ± 2.60 ^b |
| Group III (Vitamin c) | 1052.00 ± 47.30 ^a | 686.67 ± 168.80 ^a | 29.75 ± 7.45 ^a | 2.19 ± 1.59 ^a | 52.34 ± 4.22 ^b |
| Group IV (50mg/kg b. w extract) | 1013.87 ± 46.53 ^a | 319.10 ± 107.72 ^c | 19.47 ± 7.43 ^b | 2.65 ± 0.86 ^a | 53.72 ± 8.58 ^b |
| Group V (75mg/kg b. w extract) | 956.80 ± 158.57 ^a | 329.33 ± 116.78 ^c | 29.13 ± 8.51 ^a | 3.21 ± 0.73 ^a | 54.54 ± 1.60 ^b |
| Group VI (100mg/kg b. w extract) | 1238.53 ± 184.95 ^a | 707.00 ± 125.74 ^{ab} | 29.29 ± 11.29 ^a | 3.95 ± 1.33 ^c | 57.14 ± 6.57 ^b |
| Group VII (Olive oil) | 1139.20 ± 59.78 ^a | 2441.39 ± 168.26 ^d | 20.55 ± 7.76 ^a | 2.37 ± 0.57 ^a | 25.57 ± 5.08 ^c |

Values are Mean ± SD, n=6. Values in the same column bearing same superscript letters show no significant differences between the groups while those with different superscript letters (a,b,c,d...) show significant differences between the groups (P<0.05).mg/kg b. w = mg/kg body weight.

Figure 3.1 shows the percentage protection of *Cnidoscolumaconitifolius* leaf extract on CCl₄-induced hepatotoxicity in rats. The graph revealed that the leaves had protective potential on alanine transaminase, aspartate transaminase, alkaline phosphatase and total bilirubin concentrations.

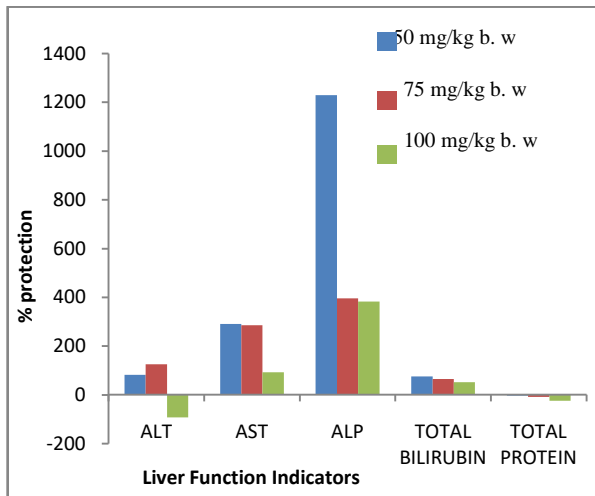


Figure 3.1: Hepatoprotective activity of the leaf extract of *Cnidioscolusaconitifolius* on CCl₄-induced hepatotoxicity in rats

3.3 Histopathological examination

Histopathological examinations of the effects of the leaf extract of *C. aconitifolius* on the normal and CCl₄-induced hepatotoxicity in rats are shown in Plate 3.3.2-3.3.8.

Plate 3.3.2: Photomicrograph of liver section of a normal control rat showed normal central vein, normal hepatic sinusoid and normal hepatocytes cells which implies histological normal liver section.

Plate 3.3.3: Photomicrograph of liver section of rats administered CCl₄ showed ballooning degeneration of hepatocytes cells, vacuolations in isolated areas (arrowed) and infiltration of inflammatory cells in sinusoids. This implies histologically distorted liver section.

Plate 3.3.4: Photomicrograph of liver section of the rats administered vitamin C showed congested central vein, ballooning degeneration of hepatocytes cells in isolated areas, sinusoid infiltrated with inflammatory cells and capillaries with red blood cells. This implies mildly distorted liver section.

Plate 3.3.5: Photomicrograph of liver section of a 50mg/kgbw of *C. aconitifolius* leaf extract treated rat showed patent central vein, sinusoid containing capillaries and buffer cells and normal hepatocytes cells which implies histological normal liver section.

Plate 3.3.6: Photomicrograph of liver section of 75mg/kgbw of *C. aconitifolius* leaf extract treated rat showed congested central vein and normal hepatocytes cells. This implies histologically normal liver section.

Plate 3.3.7: Photomicrograph of liver section of a 100mg/kgbw of *C. aconitifolius* leaf extract treated rat showed congested hepatic artery, sinusoids infiltrated with inflammatory cells and histologically normal liver cells. This implies mildly distorted liver section.

Plate 3.3.8: Photomicrograph of liver section of an olive oil induced rat showed congested hepatic artery and sinusoids infiltrated with inflammatory cells. This implies mildly distorted liver section.

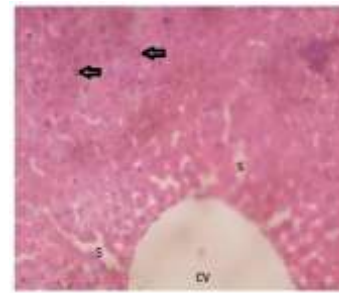


Plate 3.4.2

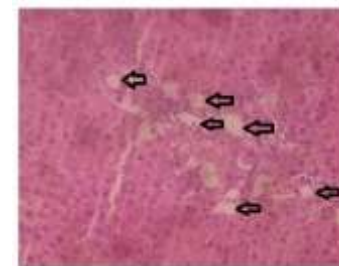


Plate 3.4.3

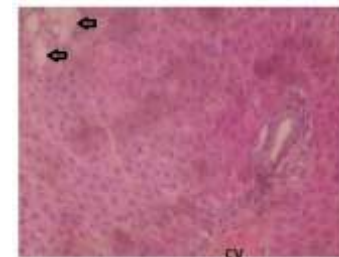


Plate 3.4.4

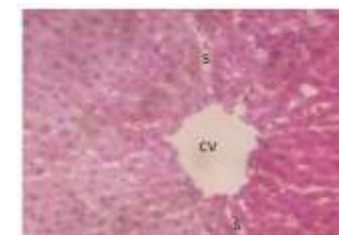


Plate 3.4.5

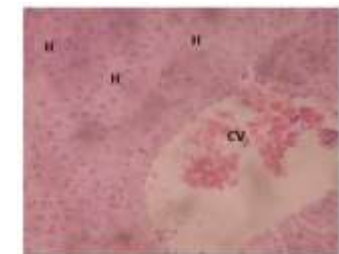


Plate 3.4.6

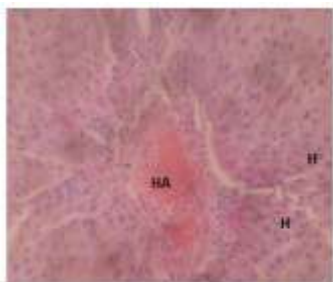


Plate 3.4.7



Plate 3.4.8

IV. DISCUSSION

The result of the proximate composition of *C. aconitifolius* is shown in Table 3.1. From this study, the moisture content was very high (82.11%) which implies that this leaf cannot be stored for long and is prone to microbial attack. The percentage of ash recorded in this study was 1.38%. Ash content represents the concentration of minerals in food sample. The lipid content reported was 2.80%. Lipid in food absorbs and retains the flavour thereby enhancing the palatability of the food [17]. The protein content (0.51%) obtained in this leaf was low. Proteins are necessary for the build-up of tissues and substances like hormones and enzymes. This means that the leaves of this plant may not contribute appreciable amount of the daily protein need. The crude fibre content recorded in *C. aconitifolius* leaf was 12.34%. Dietary fibre is essential in decreasing the risk of many disorders such as diabetes, obesity, constipation and cardiovascular diseases [18-19]. Intake of fibre can stimulate peristaltic movement, weakening of hunger, increased stool bulk and reduced serum levels of cholesterol [20-21]. Also, high fibre content lowers nutrient bioavailability and causes irritation in the intestine [22]. The leaves of this plant can provide the fibre that will provide the above listed biological functions. The carbohydrate content recorded was 0.86%. Carbohydrates are pivotal nutrients needed for a balanced diet [23]. Generally, the low content of protein, lipid and carbohydrate in this leaf implies that it is not a good source of energy.

The effects of extract of *Cnidioscolusaconitifolius* leaves on some renal function indices of normal and CCl_4 induced hepatotoxicity in rats are represented in Table 3.2. There was significant increase ($P < 0.05$) in the creatinine level of rats administered with CCl_4 when compared to the normal. Creatinine levels of treated rats at 50mg/kgbw and

75mg/kgbw dose were decreased significantly when compared to normal and diseased control respectively. There was significant increase in creatinine level at 100mg/kgbw when compared to both the normal and disease control which suggest that the kidney was not compromised. There was a significant increase ($P < 0.05$) in albumin levels of disease control group and the vitamin C group than normal control but were decreased significantly in the treatment group when compared with disease control. Albumin levels reveal the capacity of the glomeruli tubule to function properly [24]. This study showed that there was a significant decrease ($P < 0.05$) in the albumin levels of rats treated with the extracts than the disease control rats which connotes that the leaves extract might have the ability to aid the glomeruli tubule to work well. This finding was in agreement with that of Ikewuchi and Ikewuchi, [14] that found slight increase in plasma albumin of sub-chronic salt-loaded rats treated with aqueous extract of the leaves of *Tridaxprocumbens*. Creatinine levels are associated with the functional state of the nephrons [25] with raised levels signifying damage to the kidney. Results obtained here indicated that serum creatinine levels were increased significantly in the disease control rats and were reduced significantly by the leaves extract treatment at 50mg/kgbw dose. This highlights the ability of *C. aconitifolius* to protect the kidney. Similar findings were obtained by [14].

The effects of the leaf extract of *C. aconitifolius* on the plasma electrolyte profiles of normal and CCl_4 -induced hepatotoxicity in rats are shown in Table 3.3. Plasma calcium levels of animals treated with the leaf extract was lower though not significantly at 75mg/kgbw and 100mg/kgbw doses but significantly lower at 50mg/kgbw dose than the normal control. There was no significant difference in the calcium level of disease control rats when compared to normal control. There was a significant increase ($P < 0.05$) in the sodium levels of disease control rats than the normal control. The extract slightly increased the plasma sodium though not significantly in the vitamin C group and in all extract doses administered than the disease control rats. This increase was very significant when compared to normal control rats. There were no significant difference ($P < 0.05$) on magnesium levels of rats administered CCl_4 and treated rats at all the extract doses when compared to normal and disease control. There was a significant increase ($P < 0.05$) on potassium levels of rats administered with CCl_4 than the normal control but was decreased significantly in the treatment group when compared to the disease control. Results of the plasma electrolyte profile imply that the extract significantly decreased the plasma potassium levels and slightly lowered the plasma calcium levels. Reduced plasma potassium concentrations have been associated with glucose intolerance. Potassium depletion causes glucose intolerance, which is associated with impaired insulin secretion [26]. Calcium fluxes are also important mediators of hormonal effects on target organs through several intracellular signaling pathways, such as phosphoinositide and cyclic adenosine monophosphate

systems [27-28]. In nutshell, these results showed the capacity of the extract to enhance kidney functions.

The effect of the extract of *Cnidoscolumaconitifolius* on the plasma hepatospecific markers of normal and test groups' of administered rats with CCl_4 is expressed in Table 3.4. Results showed that there was a significant increase ($P < 0.05$) in the aspartate amino transferase activity, total protein and total bilirubin of rats administered with CCl_4 while the leaf extracts of *Cnidoscolumaconitifolius* significantly decreased the levels of bilirubin at all doses administered. However, the extracts at all doses increased though not significantly the levels of total protein when compared to disease control rats. There was a significant increase ($P < 0.05$) in the AST activity of rats administered with olive oil when compared to the normal and disease control but significant decrease ($P < 0.05$) in the AST activity of rats treated with 50mg/kgbw and 75mg/kgbw leaf extract of *Cnidoscolumaconitifolius*. There was no significant difference in the activity of ALT in rats administered with vitamin C, with CCl_4 and treated with the leaf extract of *Cnidoscolumaconitifolius*. However, ALT was slightly increased in disease control rats with the treatment of 50mg/kgbw and 75mg/kgbw decreasing the activity of ALT while 100mg/kgbw leaf extract increased ALT activity in rats. Also, there was a slight increase in the ALP activity of disease control rats administered with CCl_4 but was significantly decreased ($P < 0.05$) in rats treated with 50 mg/kgbw leaf extract. Results obtained in this study agrees with the analysis on hepatoprotective effect of aqueous extract of the leaves of *Acalyphawilkesiana* against carbon tetrachloride induced liver injury in rats carried out by Ikewuchiet al.[1], their study showed a significant decrease ($P < 0.05$) in the plasma ALP and AST activities in the treatment rats when compared to the disease control rats.

The hepatoprotective activity of aqueous leaf extract of *Cnidoscolumaconitifolius* on rats induced with CCl_4 hepatotoxicity is represented in Fig 3.1. From the charts, the degree of protection against CCl_4 is dependent on the doses of the leaf extract with 50mg/kgbw dose and 75mg/kgbw dose giving better protection efficiency. The extracts provided protection of about -92.94-125.98 % in ALT, 92.10-290.96 % in AST, 382.76-1229.31 % in ALP, 51.88-75.13 % in total bilirubin and -24.68 - (-2.64) % in total protein.

Furthermore, the prevention of elevation of ALT in rats treated with 50mg/kgbw and 75mg/kgbw leaf extract of *Cnidoscolumaconitifolius*, prevention of elevation of bilirubin in rats treated with the leaf extract at all dosages, significant decrease ($P < 0.05$) in the ALP activity in rats treated with 50mg/kgbw extract and significant decrease ($P < 0.05$) in AST activity in rats treated with 50mg/kgbw and 75mg/kgbw extract doses respectively indicates the potential of the leaf extract of *Cnidoscolumaconitifolius* to restore the normal functional status of the damaged liver, as well as protecting against hepatotoxicity of CCl_4 in rats. Though the mechanism by which the leaf extract exerted hepatoprotective activity is not yet clarified. it is assumed that flavonoid, a constituent of

the leaf is suspected to be responsible for the protective activity against CCl_4 induced hepatotoxicity [15]. Histopathological examination of the liver showed histological distortion of the liver sections in disease control rats which were corrected by the extracts at different doses (Figures 3.3.2-3.3.8). This suggests the hepatoprotective potential of *Cnidoscolumaconitifolius* leaf extract on carbon tetrachloride induced toxicity in rats.

V. CONCLUSION

From this study, it can be concluded that *Cnidoscolumaconitifolius* leaf extract clearly demonstrated hepatoprotective effect against carbon tetrachloride induced hepatotoxicity in rats.

5.1 Recommendation

I recommend further studies on the hepatoprotective potential using the stem and root parts of the *Cnidoscolumaconitifolius* plant as they have been shown to have high phytochemicals [9].

Also, the protective effect of the leaf extract of *Cnidoscolumaconitifolius* plant can equally be carried out to determine its effect on the hormonal balance in rats.

5.2 Contribution to Knowledge

This study has shown that *Cnidoscolumaconitifolius* leaf extract has the potential to reduce the activities of liver function enzyme markers, hence protecting the liver.

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