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Investigation of Toxicological Profile of Methanol Extract of *Dialium guineense* in Albino Wistar Rats

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Abstract: The era of synthetic drugs has led to plethora of side effects, some of which are life-threatening As a result, ethnopharmacology has become prominent across different climes in recent years. Even in advanced cultures, people are increasingly drawing closer to natural medicine. Humans have suffered untold challenges as a result of paucity of knowledge of toxicological profiles of different medicinal plants, especially in the tropics. The study investigated the acute and chronic toxicological effects of *Dialium guineense* in albino Wistar rats with the view to ascertaining its relative bio-safety. Twenty albino Wistar rats were randomly divided into four groups of four rats each thus: Group1 Normal control, Groups 2-4 (400, 800 and 1200 mg/kg bw) of 80% methanol extract of *Dialium guineense*. The study lasted for 29 days. The acute toxicity was carried out according to Lorke's method (1983). The qualitative and quantitative phytochemical analyses results showed the presence of glycosides, reducing sugar, alkaloids, flavonoids, tannins, total phenolics, steroids and terpenoids in different amounts. The result of acute toxicity indicated no death of any mice at 5000 mg/kg bw. The results of liver markers indicated that alanine aminotansferase and alkaline phosphatase activities and total protein, albumin, total bilirubin and direct bilirubin levels of the treatment were non-significantly (P>0.05) different compared to the normal rats, though AST showed a significant (P<0.05) increase in activity in *Dialium guineense* administered groups compared to the normal control. The results of the serum electrolytes levels sodium and potassium ions indicated a non-significant (P>0.05) increase in concentrations of groups 2-4 compared to group 1. However, Cl^- and HCO_3 levels were significantly (P<0.05) higher in 400 mg/kg bw compared to group1. The results of urea and creatinine showed a significant (P<0.05) decrease in their levels at 1200 mg/kg bw compared to group 1. The results of the lipid profile demonstrated that the lipid panel (total cholesterol, triacylglycerides, low density lipoprotein and high density lipoprotein) of treatment groups was non-significantly (P>0.05) different in concentrations compared to the normal control. The packed cell volume, platelet levels, white blood cells count and differential count showed non-significant (P>0.05) differences in levels and counts of treatment groups compared to normal group. The haemoglobin concentration, red blood cells count showed a significant (P<0.05) differences in treatment groups compared to group 1. Normal renal histo-architecture was observed in normal control group. On the other hand, 400 mg/kg bw showed some epithelial degeneration while **800 mg/kg bw demonstrated mild epithelial changes with mild inflammatory cell infiltration in the surrounding interstitial spaces. Group four (1200mg/kg bw) showed moderate renal tubular atrophy. With respect to liver cells investigations, group one showed normal hepatic histo-architecture whereas group two showed moderate hepatic degeneration. Group three showed marked hepatic vacuolation and disorganization consistent with cellular degeneration. Group four showed a nearly normal hepatic histo-architecture. The cardiac histolology showed that groups one, two and three exhibited normal cardiac histo-architecture while group four showed mild to moderate inflammatory cell infiltration, though no significant necrosis was observed. There was no major challenge to albino mice at acute exposure of methanol extract of *Dialium guineese*. The findings in the albino Wistar rats suggested that caution is needed in prolonged intake of methanol extract of *Dialium guineese*. This is to avoid some of the critical observations in the histo-chemical investigations, though no drug is without side effects. The plant could be employed as a pharmacological agent at a moderate dosage.**

Keywords: *Acute Toxicity, Chronic Toxicity, Liver, Kidneys, Dialium Guineense, Albino Wistar Rats.*

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I. INTRODUCTION

Ethnopharmacology is becoming prominent across different climes in recent years. The era of synthetic drugs has led to plethora of side effects, some of which are life-threatening. Even in advanced cultures, people are increasingly drawing closer to natural medicine. This is no surprise as toxicity has a lot of bearing with the genetic proximity between active molecules and the recipients of the active molecules. By extension, the closer a molecule is to humans, genetically, the more likely the molecule will be biologically accepted, though this does not vitiate the fact that toxic biomolecules abound.

The toxicological profile of a given plant provides a good pointer to the relative safety of the biomolecules. Humans have suffered untold challenges as a result of paucity of knowledge of toxicological profiles of different plants, especially in the tropics. Ethnopharmacological agents are known to possess anticancer potency Nerkar *et al*., (2023).

Plants are replete with active metabolites (phytochemicals and bioactive molecules) in the bark, roots, leaves, fruits and seeds, and have been utilized for medicinal purposes since time immemorial. Reporters have opined that some phytochemicals that have anti-ulcerative properties are alkaloids, tannins, flavonoids, terpenoids, glycosides, carotenoids and saponins (Ghosh *et al* 2016). Different "leads" owe their parent structures to ethnopharmacological investigations, Heinrich and Gibbons (2001).

The plant *Dialium guineense* Wild (family fabaceae, sub family caesalpinioideae), grows in dense savannah forests, shadowy canyons and gallery forests. It is native to Nigeria, Benin Republic, Burkina Faso, Guinea and other West African countries. It is called "icheku" (Ibo, Eastern Nigeria), "awin" (Yoruba, Western Nigeria), "tsayirarkurm" (Hausa, Northern Nigeria), velvet termarind or black tamarind (English) and tamarinier noir (French) (Van *et al*., 2015). The leaves and stem bark are used as forklore remedies for the treatment of infections and other ailments such as diarrhoae, severe cough, bronchitis, wound, stomach ache, malaria fever, jaundice, peptic ulcer disease (PUD), haemorroids and prevention of cancer (Bero *et al*., 2009).

Hepatotoxicity is a common phenomenon especially in excessive use of drugs, including alcohol consumption, microbial challenge, including viral infections. Environmental toxicants are on the increase especially in heavily industrialized cities. Different agents can poison the liver cells. They range from accidentally discharged molecules to agents that got their way into human food web via manufacturing and utilization of different synthetic molecules. Liver injury could be classified as hepatocellular, cholesatatic and mixed, precipitated by different factors Thompson *et al*., (2017).

The integrity of the nephrons is very important metabolically. Xenobiotic metabolism constitutes a heavy challenge to the integrity of the nephrons. Molecules that preserve the nephrons are highly desirable. The interdependence of metabolic processes has made it both desirable and undesirable operations. A significant percentage of molecules that come from the heart are bound to pass through the kidneys, sometimes precipitating nephro-toxicity Shaman *et al*., (2020). A prominent factor to be critically addressed in the renal-restorative cases is prevention of tubular ischemia. Agents that are potent in doing this is of great importance Duke (1999).

The study was aimed at investigating the acute and chronic toxicological effects of *Dialium guineense* in albino mice and albino Wistar rats with the view to ascertaining its relative bio-safety. This is important in having an improved pharmacological knowledge in the usage of this medicinal plant.

II. MATERIALS AND METHODS

A. Plant Materials

The leaves of *Dialum guineense* were collected from Ibagwa community in Igbo-Eze South Local Government area of Enugu State Nigeria and identified by Isaac Ossai, a taxonomist from Biodiversity and Conservation Programme (BDCP) Nsukka. They were shade dried and pulverized. Thereafter they were macerated in 80 % methanol for 72 hours. Filtration followed using muslin cloth (to remove the chaff) and Whatmann No 1 filter paper. Concentration was achieved using magnetic stirrer.

B. Qualitative and Quantitative Phytochemical Analyses

The methods of Harbone (1978) and Trease and Evans (1989) were used to qualitatively and quantitatively determine alkaloids, flavonoids, tannins, total phenolics, glycosides, reducing sugars, steroids and terpenoids.

C. Median Lethal dose (LD_{50})

The median lethal dose of crude methanol extract of *Dialum guineense* was done according to Lorke, ((1983). Two phases of the testing were carried out using albino mice as follows:

- *Phase 1*

Nine mice were grouped three per group as follows:

Groups	Dosages	Observations for death(s) if any
1	10 mg/kg bw	
2	100 mg/kg bw	
3	1000 mg/kg bw	

After twenty-four hours observations were made for signs of toxicity or death(s) if any before phase 2 was undertaken.

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➢ *Phase 2*

Groups	Dosages	Observations for death(s) if any
1	1600 mg/kg bw	
2	2900 mg/kg bw	
3	5000 mg/kg bw	

Observations were made for any possible sign of toxicity or death(s) for twenty four hours.

D. Animals

➢ *Experimental Design*

Sixteen albino Wistar rats were used in this study. They were acclimatized in aluminum cages, exposed to 12hr light/dark cycles and fed poultry mash ad libidum. After acclimatization, they were randomly divided into four groups of four rats as follows:

- Group 1 Normal control
- Group 2 400 mg/kg bw Methanol extract of *Dialium guineense*
- Group 3 800 mg/kg bw Methanol extract of *Dialium guineense*
- Group 4 1200 mg/kg bw Methanol extract of *Dialium guineense*

The study lasted for 29 days. The blood of the rats were collected using ocular puncture and the rats were sacrificed using cervical dislocation before the organs were collected for histological examinations.

E. Determination of Biochemical and Haematological Parameters

The liver function test, renal function test, lipid profile and haematological profile were done in accordance with the methods of Trinder (1951), Terri and Sesin (1958), Skeggs and Hochstrasser (1964), Reitman and Frankel (1957), Jendrassik and Grof (1938), Bartels and Bohmer (1972) and Ochei and Kolhatkar, (2000).

F. Histopathological Examinations

The histopathological examinations of the stomach and duodenum of the albino Wistar rats were done using the method of Drury *et al*., (1967). It involved fixing and washing of the tissues. Dehydration was achieved using alcohol and the embedment was done in paraffin. They were subsequently cleared, infiltrated with paraffin wax, sectioned, mounted on the slides and stained with eosin and haematoxylin. The slides prepared were mounted on photomicroscope one after the other and viewed at x 400 magnification power of the microscope. Qualitative histological interpretations of the different microhistophotograph were made and reported.

G. Statistical Analysis

Data were reported as means ±SD where applicable. One way analysis of variance (ANOVA) was used to analyze the experimental data. Duncan multiple test range was used to separate the means and differences were considered significant at $p<0.05$ using statistical package for service solution (SPSS) version 23.

III. RESULTS

A. The Qualitative and Quantitative Phytochemical Analyses Results

The qualitative and quantitative phytochemical analyses results showed the presence of glycosides, reducing sugar, alkaloids, flavonoids, tannins, total phenolics, steroids and terpenoids in different amounts. However saponins were not detected.

Table 1: Showing the Phytochemical Analyses Results for qualitative and quantitative analyses

Phytochemicals	Bioavailability Amount	mean±SD (mg/100g)
Glycosides	+	16.80 ± 0.042^{a}
Reducing sugar	++	1137.68 ± 42.35^{b}
Alkaloids	++	509.03 ± 51.16^{a}
Flavonoids	+++	1697.09 ± 245.51^{b}
Tannins	++	57.89 ± 0.71^{a}
Total phenolics	+++	11021.00 ± 134.41^{c}
Steroids	+	9.03 ± 0.03^{a}
Terpenoids	++	266.63 ± 4.90^{a}

Superscripts that are the same were considered non-significant ($P>0.05$) while those that were different were considered significant ($P<0.05$)

B. The Result of Median Lethal Dose (LD_{50}) of Dialium Guineense on Albino Mice

There was no death recorded in phases one and two after twenty-four hour durations in albino mice.

Table 2: The Median Lethal Dose (LD_{50}) Results for Phases One and Two Albino Mice Treated with Different Concentrations of *Dialium guineenses*

Groups	Dosages	Observations for death(s)
1	10 mg/kg bw	0/3
2	100 mg/kg bw	0/3
3	1000 mg/kg bw	0/3
4	1600 mg/kg bw	0/3
5	2900 mg/kg bw	0/3
6	5000 mg/kg bw	0/3

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C. The Results of the Liver Function Tests of Albino Wistar Rats Treated with Methanol Fraction of Dialium guineense Leaves

The results of liver markers indicated that alanine aminotansferase and alkaline phosphatase activities and total protein, albumin, total bilirubin and direct bilirubin levels of the treatment were non-significantly (P>0.05) different compared to the normal rats, though AST showed a significant (P<0.05) increase in activity in *Dialium guineense* administered groups compared to the normal control.

Table 3(a): Showing the Results of the Liver Function Tests of Albino Wistar Rats Treated with Methanol Fraction of *Dialium guineense* Leaves

Groups	ALT IU/L	AST IU/L	ALP IU/L	Total bil mg/dl	Direct bil mg/dl
1	11.00±1.87a	11.60±3.13a	31.00±1.41a	0.48a	0.35±0.05a
2	9.60±0.55a	15.20±2.59b	31.40±0.89a	0.58a	0.33±0.08a
3	10.00±1.16a	12.50±1.92a,b	29.75±1.50a	0.63a	0.34±0.11a
4	11.20±1.30a	15.20±1.10b	29.60±1.14a	0.48a	0.29±0.06a

Superscripts that are the were considered non-significant (P>0.05) while those that were different were considered significant (P<0.05)

Table 3(b): Showing the Results of the Liver Function Tests of Albino Wistar Rats Treated with Methanol Fraction of *Dialium guineense* Leaves

Groups	Albumin mg/dl	Total Protein mg/dl
1	4.46±0.56a	4.98±0.79a
2	4.26±0.65a	5.06±0.48a
3	4.30±0.49a	4.43±0.25a
4	4.26±0.47a	4.34±0.61a

Superscripts that are the were considered non-significant (P>0.05) while those that were different were considered significant (P<0.05)

D. The Results of the Kidneys Function Tests of Albino Wistar Rats Treated with Methanol Fraction of Dialium guineense Leaves

The results of the serum electrolytes levels sodium and potassium ions indicated a non-significant (P>0.05) increase in concentrations of groups 2-4 compared to group 1. However, Cl^- and HCO_3 levels were significantly (P<0.05) higher in 400 mg/kg bw compared to group1. The results of urea and creatinine showed a significant (P<0.05) decrease in their levels at 1200 mg/kg bw compared to group 1.

Table 4(a): Showing the Results of the Kidneys Function Tests of Albino Wistar Rats Treated with Methanol Fraction of *Dialium guineense* Leaves

Groups	Na^+ mmol/L	K^+mmol/L	Cl^- mmol/L	HCO_3^- mmol/L
1	$125.60±9.71^a$	$4.02±0.35^a$	$96.40±1.14^a$	$26.20±0.84^a$
2	$139.00±19.13^a$	$4.24±0.53^a$	$100.40±3.65^b$	$28.00±0.71^b$
3	$130.75±7.37^a$	$4.05±0.79^a$	$96.00±1.41^a$	$27.00±0.82^a$
4	$131.00±8.75^a$	$4.58±0.45^a$	$96.80±0.84^a$	$26.00±0.71^a$

Superscripts that are the were considered non-significant (P>0.05) while those that were different were considered significant (P<0.05).

Table 4(b): Showing the Results of the Kidneys Function Tests of Albino Wistar Rats Treated with Methanol Fraction of *Dialium guineense* Leaves

Groups	Urea mg/dl	Creatinine mg/dl
1	$43.80±4.03^b$	$1.56±0.17^b$
2	$38.60±3.65^{a,b}$	$1.36±0.11^{a,b}$
3	$39.00±5.42^{a,b}$	$1.38±0.22^{a,b}$
4	$37.40±2.30^a$	$1.30±0.10^a$

Superscripts that are the were considered non-significant (P>0.05) while those that were different were considered significant (P<0.05)

E. The Results of the Lipid Profile of Albino Wistar Rats Treated with Methanol Fraction of Dialium guineense Leaves

The results of the lipid profile demonstrated that the lipid panel (total cholesterol, triacylglycerides, low density lipoprotein and high density lipoprotein) of treatment groups

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was non-significantly (P>0.05) different in concentrations compared to the normal control.

Table 5: Showing the Results of the Lipid Profile of Albino Wistar Rats Treated with Methanol Fraction of *Dialium guineense* Leaves

Groups	T.Chol	TAG	HDL	LDL
1	5.16±0.47[a]	1.72±0.13[a]	1.62±0.13[a]	3.20±0.59[a]
2	4.92±0.38[a]	1.58±0.23[a]	1.64±0.17[a]	3.06±0.35[a]
3	5.10±0.48[a]	1.65±0.24[a]	1.80±0.14[a]	2.97±0.40[a]
4	4.74±0.09[a]	1.62±0.19[a]	1.76±0.11[a]	2.66±0.11[a]

Superscripts that are the were considered non-significant (P>0.05) while those that were different were considered significant (P<0.05).

F. The Results of the Haematological Profile of Albino Wistar Rats Treated with Methanol Fraction of Dialium guineense Leaves

The packed cell volume showed a non-significant (P>0.05) increase in percentage PCV levels of groups 2-4 compared to group 1. The haemoglobin concentration showed a significant (P<0.05) decrease in Hb level of group 2 compared to group 1 while groups 3 & 4 were non-significantly (P>0.05) different compared to group 1. The result of red blood cells' count showed a significant (P<0.05) increase in RBC level of 800 mg/kg bw compared to group 1 while 400 and 1200 mg/kg bw were non-significantly (P>0.05) higher when compared to group 1. The result of white blood cells' counts indicated a non-significant (P>0.05) decrease in WBC levels of 800 and 1200mg/kg bw compared to group 1. However, 400 mg/kg bw demonstrated a non-significant (P>0.05) increase in WWBC level compared to group 1 and a significant (P<0.05) increase in WBC level compared to 1200 mg/kg bw.

The result of platelet count showed that groups 2 and 3 showed a non-significant (P>0.05) decrease in platelet levels compared to group 1. On the other hand group 4 showed a non-significant (P>0.05) increase in platelet count compared to group 1.

The result of the differential count indicated a non-significant (P>0.05) differences in percentage neutrophils, lymphocytes, monocytes, eosinophils and basophils of treatment groups 2-4 compared to group 1.

Table 6(a): Showing the Results of the Haematological Profile of Albino Wistar Rats Treated with Methanol Fraction of *Dialium guineense* Leaves

Groups	PCV (%)	Hb (mg/dl)	RBC ($x10^9$/L)	WBC ($x10^{12}$/L)	Platelet
1	43.20±2.28[a]	16.92±1.68[b]	256.00±20.74[a]	348.00±21.68[a,b]	274.00±15.17[a]
2	46.80±3.03[a]	13.52±1.50[a]	256.00±15.17[a]	370.00±15.81[b]	270.00±15.81[a]
3	45.75±3.50[a]	15.45±1.79[a,b]	280.00±8.17[b]	345.00±28.87[a,b]	258.00±26.46[a]
4	45.60±3.78[a]	17.14±2.43[b]	268.00±14.83[a,b]	326.00±8.94[a]	278.00±13.04[a]

Superscripts that are the were considered non-significant (P>0.05) while those that were different were considered significant (P<0.05).

Table 6(b): Showing the Results of the Haematological Profile of Albino Wistar Rats Treated with Methanol Fraction of *Dialium guineense* Leaves

Groups	N(%)	L(%)	E(%)	M(%)	B(%)
1	61.60±5.73[a]	37.20±5.59[a]	1.20±1.10[a]	0.00±0.00	0.00±0.00
2	63.60±4.34[a]	35.60±4.98[a]	0.80±1.10[a]	0.00±0.00	0.00±0.00
3	59.50±1.92[a]	39.50±3.00[a]	1.00±1.16[a]	0.00±0.00	0.00±0.00
4	61.60±4.56[a]	37.20±5.40[a]	1.20±1.10[a]	0.00±0.00	0.00±0.00

Superscripts that are the were considered non-significant (P>0.05) while those that were different were considered significant (P<0.05)

G. Histopathological Plates Showing Different Photomicrographs of Different Organs in Albino Wistar Rats Treated with Different Concentrations of Dialium guineense

➢ *Group 1: Showing Photomicrograph of Kidney Section* ***of an Albino Wistar Rat (Normal Control)***

A photomicrograph of kidney section **from an albino rat in this group shows** normal renal histo-architecture. Normal glomeruli (G) in their respective Bowman's capsules surrounded by numerous normal renal tubules (RT) embedded in a highly-vascularized interstitial spaces. Basement membrane (BM). (H&E Stain, x200) as shown in Plate 1A.

➢ *Group 2: Showing Photomicrograph of Kidney Section* ***of an Albino Wistar Rat Treated with*** *400 mg/kg bw Methanol extract of Dialium guineense*

A photomicrograph of a kidney section from an albino rat in this group shows mild renal tubular atrophy, evidenced by variation in tubular lumen size and mild epithelial degeneration (white arrows). A few glomeruli exhibit

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glomerulosclerosis, characterized by increased extracellular matrix deposition and capillary tuft distortion (black arrow). Renal tubule (RT), Glomerulus (G).**(H&E Stain, x200) as shown in Plate 2A.**

- *Group 3: Showing Photomicrograph of Kidney Section* ***of an Albino Wistar Rat Treated with*** *800 mg/kg bw Methanol extract of Dialium guineense*

A photomicrograph of a kidney section from an albino rat in this group shows mild glomerular hyper-cellularity (G) with preserved structural integrity. Renal tubules (RT) appear largely intact, though some show mild epithelial changes (black star). There is mild inflammatory cell infiltration in the surrounding interstitial spaces (white arrows) (H&E stain, x200) as shown in Plate 3A.

- *Group 4: Showing Photomicrograph of Kidney Section* ***of an Albino Wistar Rat Treated with*** *1200 mg/kg bw Methanol extract of Dialium guineense*

A photomicrograph of a kidney section from an albino rat in this group shows moderate renal tubular atrophy (black arrows). Additionally, a few glomeruli exhibit marked congestion (white arrow), and there is mild inflammatory cell infiltration in the interstitial spaces (arrow heads). Normal glomeruli (G) and basement membrane (BM) observed. (H&E stain, x200) as shown in Plate 4A.

- *Group 1: Showing Photomicrograph of Liver Section* ***of an Albino Wistar Rat (Normal Control)***

A photomicrograph of a liver section from an albino rat in this group shows normal hepatic histo-architecture. The hepatocytes are arranged in plates radiating from the central vein (V), with prominent nuclei and intact cytoplasm. The hepatic sinusoids (S) appear normal and well-distributed. Kupffer cells are visible within the sinusoids. The hepatic cords (CH) are well-preserved, with no evidence of inflammation, fibrosis, or necrosis. The endothelial lining (E) of the blood vessels remained intact. (H&E stain, x200) as shown in Plate 1B.

- *Group 2: Showing Photomicrograph of Liver Section* ***of an Albino Wistar Rat Treated with*** *400 mg/kg bw Methanol extract of Dialium guineense*

A photomicrograph of a liver section from an albino rat in this group shows mild to moderate hepatic degeneration. There is marked inflammatory cell infiltration (black arrows) within the portal area (P), suggestive of hepatocellular injury. The hepatocytes exhibit mild disorganization, but no significant necrosis is observed. Additionally, the sinusoids appear mildly congested. (H&E stain, x200) as shown in Plate 2B.

- *Group 3: Showing Photomicrograph of Liver Section* ***of an Albino Wistar Rat Treated with*** *800 mg/kg bw Methanol extract of Dialium guineense*

A photomicrograph of a liver section from an albino rat in this group shows marked hepatic degeneration. There is moderate to severe periportal inflammatory cell infiltration (black arrows) extending into the surrounding hepatic parenchyma, suggestive of hepatocellular injury. Additionally, scattered inflammatory foci (arrowheads) are observed within the hepatic lobules, indicating an ongoing inflammatory response. The hepatocytes appear vacuolated (V) and disorganized, consistent with cellular degeneration. (H&E stain, x200) as shown in Plate 3B.

- *Group 4: Showing Photomicrograph of Liver Section* ***of an Albino Wistar Rat Treated with*** *1200 mg/kg bw Methanol extract of Dialium guineense*

A photomicrograph of a liver section from an albino rat in this group shows a nearly normal hepatic histo-architecture. However, area of necrosis with focal inflammatory cell infiltration (black arrows) are observed within the hepatic parenchyma, suggestive of localized hepatocellular injury. Sinusoids (S), Cord of hepatocytes (CH) and Endothelial cells (E).(H&E stain, x200) as shown in Plate 4 B.

- *Group 1: Showing Photomicrograph of Heart Section* ***of an Albino Wistar Rat (Normal Control)***

A photomicrograph of a heart section from an albino rat in this group shows normal cardiac histo-architecture. The endocardium (EC) appears intact, and the myocardium (MC) consists of well-organized cardiac muscle fibers. Cardiomyocytes exhibit centrally located dark nuclei (DN) without evidence of degeneration or necrosis. (H&E stain, x200) as shown in Plate 1C.

- *Group 2: Showing Photomicrograph of Heart Section* ***of an Albino Wistar Rat Treated with*** *400 mg/kg bw Methanol extract of Dialium guineense*

A photomicrograph of a heart section from an albino rat in this group shows normal cardiac histo-architecture. The endocardium (EC) appears intact, and the myocardium (MC) consists of well-organized cardiac muscle fibers. Cardiomyocytes exhibit centrally located dark nuclei (DN) without evidence of degeneration or necrosis. (H&E stain, x200) as shown in Plate 2C.

- *Group 3: Showing Photomicrograph of Heart Section* ***of an Albino Wistar Rat Treated with*** *800 mg/kg bw Methanol extract of Dialium guineense*

A photomicrograph of a heart section from an albino rat in this group shows largely intact cardiac histo-architecture. However, focal inflammatory cell infiltration (black arrows) is observed within the endocardium (EC). The cardiomyocytes appear largely intact, but there is mild disorganization of muscle fibers. No significant necrosis or fibrosis is observed. (H&E stain, x200) as shown in Plate 3C.

- *Group 4: Showing Photomicrograph of Heart Section* ***of an Albino Wistar Rat Treated with*** *1200 mg/kg bw Methanol extract of Dialium guineense*

A photomicrograph of a heart section from an albino rat in this group shows mild to moderate inflammatory cell infiltration (black arrows) within the endocardium (EC) and adjacent myocardial tissue. The cardiomyocytes appear somewhat disorganized, with areas of darkly stained nuclei (DN). No significant necrosis or fibrosis is observed. (H&E stain, x200) as shown in Plate 4C.

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- *Histopathological Photomicrographs of Different Organs of Albino Wistar Rats*

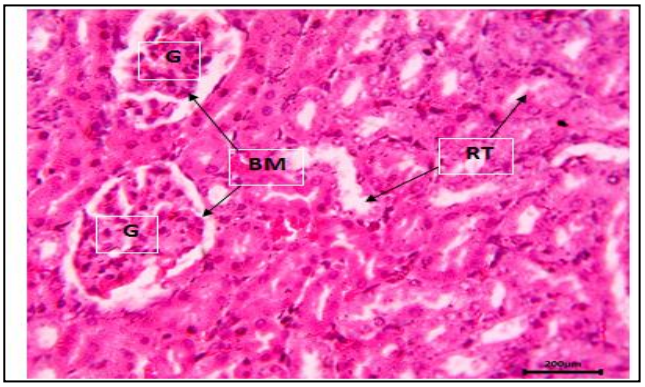

Plate 1(A): Group1 Kidney

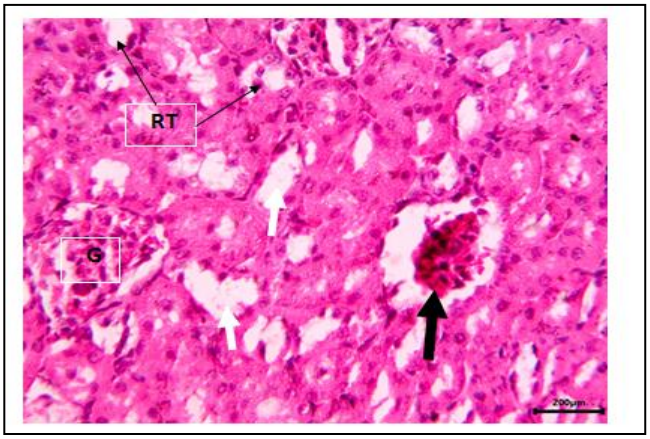

Plate 1(B): Group 2 Kidney

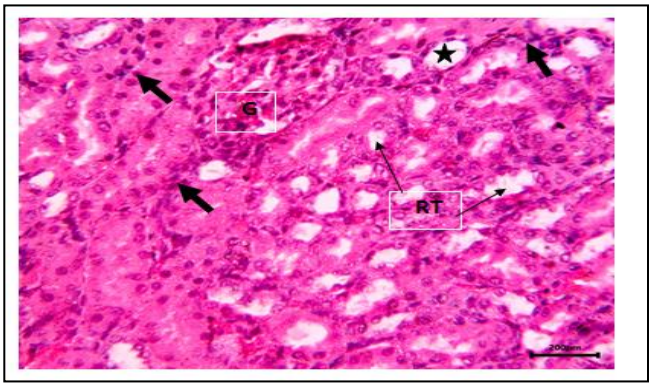

Plate 1(C): Group 3 Kidney

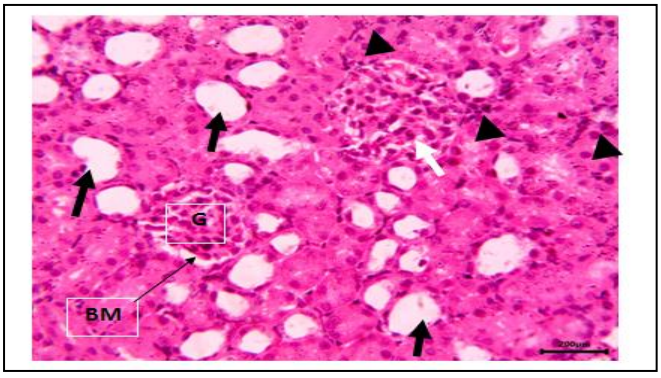

Plate 1(D): Group 4 Kidney

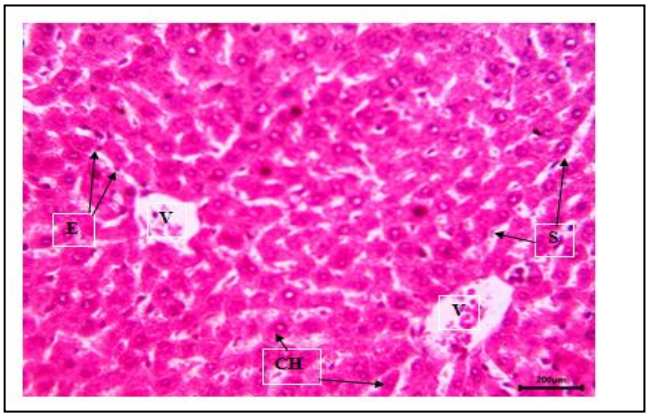

Plate 2(A): Group 1 Liver

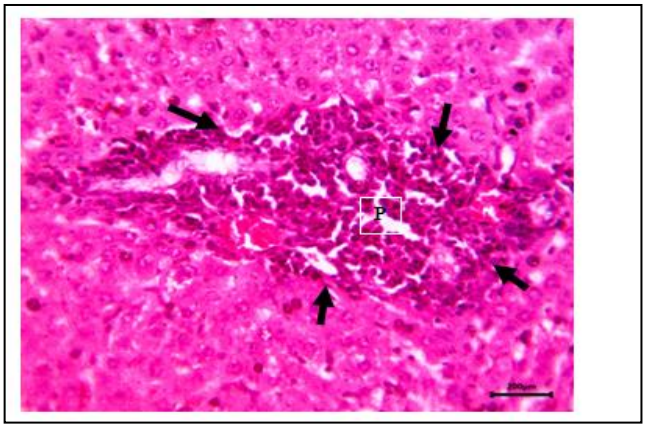

Plate 2(B): Group 2 Liver

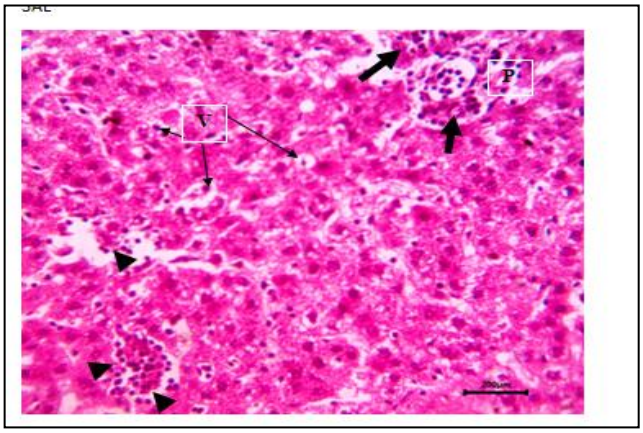

Plate 2(C): Group 3 Liver

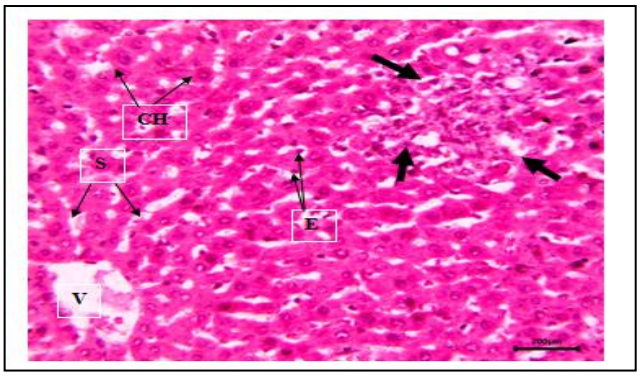

Plate 2(D): Group 4 Liver

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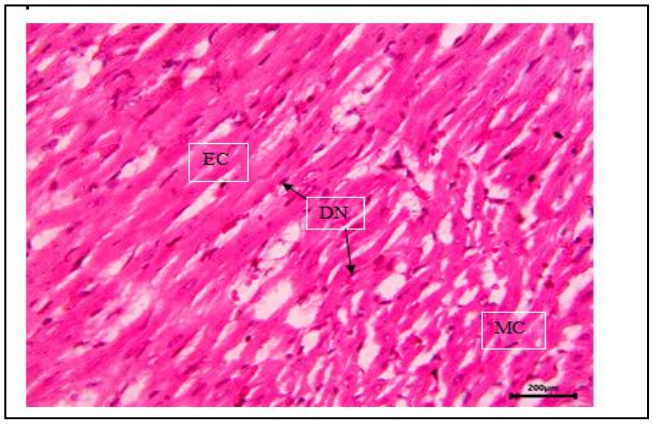

Plate 3(A): Group 1 Heart

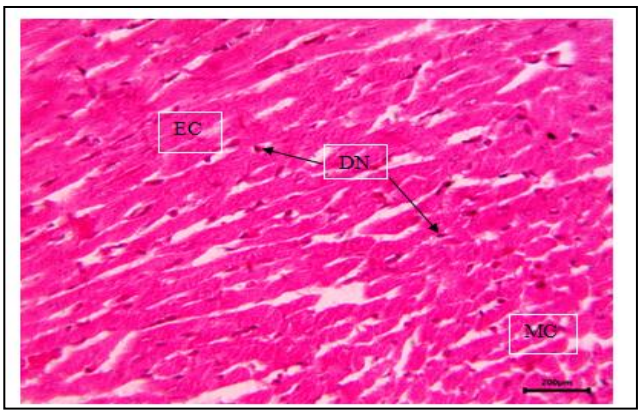

Plate 3(B): Group 2 Heart

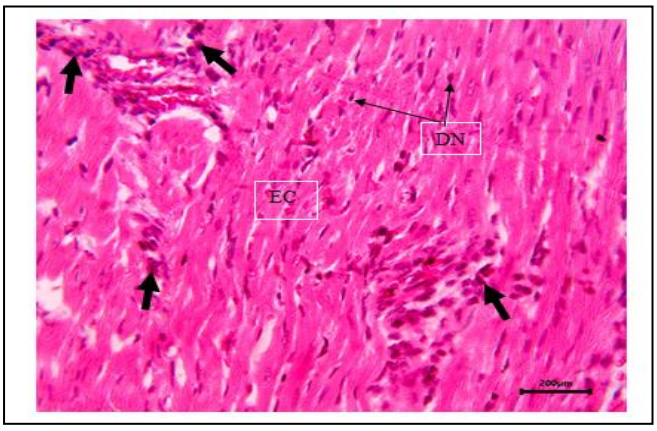

Plate 3(C): Group 3 Heart

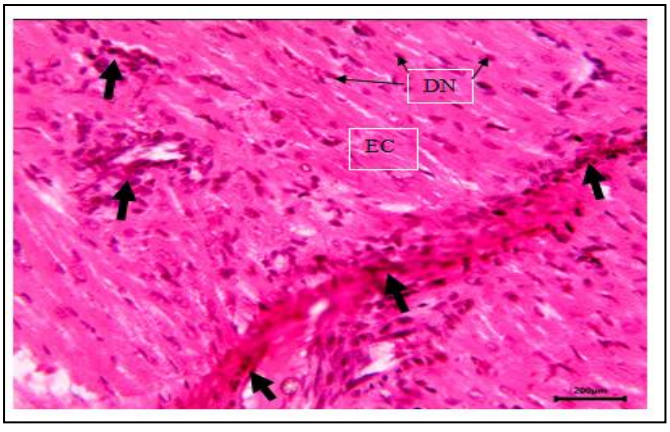

Plate 3(D): Group 4 Heart

IV. DISCUSSION

Ethnopharmacology is becoming prominent across different climes in recent years. The era of synthetic drugs has led to plethora of side effects, some of which are life-threatening. Even in advanced cultures, people are increasingly drawing closer to nature. This is no surprise as toxicity has a lot of bearing with the genetic proximity between active molecules and the recipients of the active molecules. By extension, the closer a molecule is to humans, genetically, the more likely the molecule will be biologically accepted, though this does not vitiate the fact that toxic biomolecules abound.

The use of methanol to extract the active pharmacological agents in *Dialium guineense* was aimed at having a wider range of both polar and non-polar agents for possible better pharmacological activities. Methanol has been viewed as the best solvent for solvent extraction (Patel *et al*., 2012). This informed the choice of methanol in the extraction of active bio-compounds of *Dialium guineense*

Different phytochemicals present in *Dialium guineense* suggested that the plant is rich in phyto-constituents hence some possible pharmacological effects were capable of being elicited. The results of the phytochemical analyses demonstrated that the plant possessed different phytochemicals. These natural bio-compounds are responsible for different potencies claimed in different investigations made on this plant previously. For instance, the presence of tannins, flavonoids and total phenolics could account for antioxidant potency of a plant. Mohamed *et al*., (2014) had suggested that medicinal plants are replete with potent phytochemicals. Alkaloids, cyanogenic glycosides and other nitrogenous compounds could account for toxicity in different plants. Humans have relied heavily on different plants to tackle different diseases. Ahamefula *et al*., ((2018) reported the presence of flavonoids, terpenoids, steroids, phenolics that could elicit biological activity in plants. The result of the acute toxicity pointed to relative safety of Dialium guineense at acute phase, peripherally suggesting the feasibility of continued usage of the plant as already advocated in ethnopharmacology.

The results of liver markers at chronic stage, that indicated that alanine aminotansferase and alkaline phosphatase activities and total protein, albumin, total bilirubin and direct bilirubin levels of the treatment were non-significantly ($P>0.05$) different compared to the normal rats, though AST showed a significant ($P<0.05$) increase in activity in *Dialium guineense* administered groups compared to the normal control suggested that though there might not be a major hepatic disorganization, necrosis or hypertrophy, the liver cells might have been stressed. This observation could possibly be as a result of constant xenobiotic dynamics since it (liver) had the substances in *Dialium guineense* to contented with for twenty-eight days. Liver injury varies in different forms and could be classified as hepatocellular, cholesatatic and mixed, precipitated by different factors Thompson *et al*., (2017). Pharmacological agents that are relatively less toxic are helpful as plethora of agents tend to distort hepatic integrity.

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Haemostasis seemed to have been maintained since there was no distortion in sodium ion levels and potassium ion levels. Anions on the other hand were significantly ($P<0.05$) increased suggesting either a higher level of these anions in the anion gap. The overall predisposing factors of electrolytes disorder could range from age, body mass index, nutritional status, current prescribed medicines among others Alem *et al.*, (2021). An electrolyte imbalance may be a sign of a heart, lung or kidney problem. A significant percentage of molecules that participate in metabolism pass through the kidneys, sometimes giving rise to nephro-toxicity Shaman *et al*., (2020). A major concern to be closely addressed in the renal-restorative cases is prevention of tubular ischemia. Agents that are effective in doing this is of great importance Duke (1999).

Twenty-eight days administration of *Dialium guineense* did not cause dyslipidaemia in albino Wistar rats. The HDL, though non-significant ($P>0.05$) increase at 800 and 1200 mg/kg bw indicated a positive effect with respect to artherosclerosis. HDL is known to ameliorate the pathogenesis of artherosclerosis.

Though a non-significant ($P>0.05$) increase was observed in the packed cell volume of albino Wistar rats treated with *Dialium guineense,* there seemed to be a positive modulation of haematopoietic process suggesting the possibility of using the plant as a quick remedy in anaemic condition. Drug-receptor interaction may account for the significant reduction of Hb of 400mg/kg bw compared to group 1. The increase in RBC level of treatment compared to normal control, reiterate the fact that the leaves of *Dialium guineense* is not just relatively safe, but boosts blood cells' concentrations. Red blood cells are the main determinant of blood viscosity defining the frictional forces exerted by the blood on the arterial wall. In pathology red blood cells collide with the arterial wall inducing both local retention of their membranous lipids and local hemolysis, releasing heme-Fe^{++} with a high toxicity for arterial cells: endothelial and smooth muscle cells, cardiomyocytes, neurons among others Michel and Martin-Ventura (2020). Polycythermia vera is a rare blood disorder in which there is an increase in all blood cells, particularly red blood cells. The increase in blood cells makes the blood thicker. Thick blood can lead blood clots forming in blood vessels. This can cause strokes or tissue and organ damage. The trend observed in WBC suggested that care must be taken to avoid a negative impact with respect to immune-modulatory action of *Dialium guineense* at a very high dose for a long duration. The trend in platelet indicated that the sample did not attenuate inflammation. *Dialium guineense* did not distort the immune system at the end of the twenty-nine days study. This could be as a result of the fact that *Dialium guineense* did not alter the differential counts within this duration.

Normal renal histo-architecture- normal glomeruli in their respective Bowman's capsules were surrounded by numerous normal renal tubules embedded in highly-vascularized interstitial spaces was seen in the kidney cells of normal rat. On the other hand, **group two (400mg/kg bw) showed mild renal tubular atrophy, variation in tubular lumen size and mild epithelial degeneration.** Group three (800mg/kg bw) showed mild glomerular hyper-cellularity with preserved structural integrity. Renal tubules appeared largely intact, though some showed mild epithelial changes. There was mild inflammatory cell infiltration in the surrounding interstitial spaces. Group four (1200mg/kg bw) showed moderate renal tubular atrophy, glomeruli congestion and mild inflammatory cell infiltration.

With respect to liver cells investigations, group one showed normal hepatic histo-architecture whereas group two showed moderate hepatic degeneration marked by inflammatory cell infiltration within the portal area. The hepatocytes exhibited mild disorganization, but no significant necrosis was observed. The sinusoids appeared mildly congested. Group three showed marked hepatic vacuolation and disorganization consistent with cellular degeneration. There was moderate to severe periportal inflammatory cell infiltration extending into the surrounding hepatic parenchyma. Group four showed a nearly normal hepatic histo-architecture. However, area of necrosis with focal inflammatory cell infiltration was observed within the hepatic parenchyma. Different parts of *Dialium guineense* are employed in tackling different ailments. Prolonged exposure of individuals to *Dialium guineense* is inevitable especially in rural settings. The leaves and stem bark are used as forklore remedies for the treatment of infections and other ailments such as diarrhoae, severe cough, bronchitis, wound, stomach ache, malaria fever, jaundice, peptic ulcer disease (PUD), haemorroids and prevention of cancer (Bero *et al*., 2009). Though histological evidence has suggested toxicity at prolonged usage, the pharmacological relevance is a significant reason to systematically exploit the acceptable range of dosage in order to maximize the therapeutic benefits of this ethno-medicine.

The cardiac histolology showed that groups one, two and three exhibited normal cardiac histo-architecture while group four showed mild to moderate inflammatory cell infiltration, though no significant necrosis was observed. In pathology red blood cells collide with the arterial wall inducing both local retention of their membranous lipids and local hemolysis, releasing heme-Fe^{++} with a high toxicity for arterial cells: endothelial and smooth muscle cells, cardiomyocytes, neurons among others Michel and Martin-Ventura (2020).

V. CONCLUSION

There was no major challenge to albino mice at acute exposure of methanol extract of *Dialium guineese*. The findings in the albino Wistar rats suggested that caution is needed in prolonged intake of methanol extract of *Dialium guineese*. This is to avoid some of the critical observations in the histopathological investigations, though no drug is without side effects. The plant could be employed as a pharmacological agent.

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REFERENCES

[1]. Ahamefula, A. A., Onyekwere, B. C., Ogbuchi, E. U. G. I., Ihemetu, G. U. and Echeme, J. B. O. (2018). Chemical Constituents of Methanol Leaf Extract of Aspilia Africana CC. D. Adams by GCMS. *International Journal of Advanced Research in Chemical Science*. 5(10): 21-29 (2018).

[2]. Alem A., Edae C. K., Wabolo E. K. and Wondimu B. Z. (2021). Factors Influencing the Occurrence of Electrolyte Disorders in Cancer Patients. *SAGE Open Medicine*.9:1-7

[3]. Bartels H and Bohmer M. (1972). *In vitro* Determination of Creatinine and Urea. *Clinical Chemistry*, 2: 37-193

[4]. Duke G. J. (1999). Renal Protective Agents: A Review. *Critical Care and Resuscitation* 1(3):265-75.

[5]. Drury, R. A., Wallington, A. and Cameroun, S. R. (1967). In Carlleton's Histhological Techniques. Oxford University Press, New York. pp. 1-420

[6]. Heinrich, M. and Gibbons, S. (2001). Ethnopharmacology in Drug Discovery: An Analysis of its role and Potential Contribution. *Journal of Pharmacy and Pharmacology*. 53(4): 425-32.

[7]. Jendrassik L, and Grof P. (1938). *In vitro* Determination of total and Direct Bilirubin. *Biochemica*, 297: 81.

[8]. Maruna RFL. (1958). Colorimetric determination of sodium. *Clinical Chemistry*. 2:581.

[9]. Michel J. and Martin-Ventura J. L. (2020). Red Blood Cells and Hemoglobin in Human Atherosclerosis and Related Arterial Diseases. *International Journal of Molecular Sciences.* 21(18): DOI:10.3390/ijms21186756

[10]. Mohamed, E. Z., Amani, S. A., Monerah, R. O. and Sahar, K. D. (2014). Phytochemical analysis and antimicrobial activity of medicinal plants against pathogenic microorganisms. *Life Science Journal*. 11(7):350-354.

[11]. Nerkar, A. G., Nagarkar, R. and Badar, S. (2023). Ethnopharmacological Review of Tumeric for Anticancer Activity. *Current Trends in Pharmacy and Pharmaceutical Chemistry*:5(1)10-15.

[12]. Patel, D. k., Prasad, S. K., Kumar, R. and Hemalatha, S. (2012). An overview of antidiabetic medicinal plants having insulin mimetic property. *The Asain Pacific Journal of Tropical Biomedicine*, 2(4): 320-330.

[13]. Reitman S, and Frankel S. (1957). A colorimetric method for the determination of serum glutamic oxaloacetic and glutamic pyruvic transaminases. *American Journal of Clinical Pathology*, 28: 56-63.

[14]. Shaman, S., Jose, J., Kumar, S. and Ahmed, R. (2020). A Brief Study of Nephroprotective Agents. *Indian Journal of Pharmaceutical and Biological Research*. 8(1):9-13.

[15]. Skeggs L. T, and Hochstrasser H. C. (1964). Colorimetric Determination of Chloride. *Clinical Chemistry*, 10:918.

[16]. Terri AE, and Sesin PG. (1958). Colorimetric Determination of Potassium. *American Journal of Clinical Pathology*, 29:86.

[17]. Trinder P. (1951). Colorimetric Determination of Sodium. *Analyst*, 76:596.

[18]. Thompson, M., Jaiswall, Y., Wang, I. and Williams, L. (2017). Hepatotoxicity: Treatment, Causes and Applications of Medicinal Plants as Therapeutic Agents. *The Journal of Phytopharmacology*. 6(3):186-193.