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Effects of Ethanolic Extracts of Fruits of Acacia nilotica and Flowers of Calotropis procera on Liver Function of Aspirin-Induced Male Albino Rats

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Abstract

This study examined the effects of ethanolic extracts of fruits of *Acacia nilotica* and flowers of *Calotropis procera* on the liver function of male albino rats. *Acacia nilotica* fruits and *Calotropis procera* flowers are commonly consumed by natives in northern Nigeria for therapeutic purposes. Eighty-four (84) healthy male albino rats were used for this study. The animals were randomly distributed into six groups of ten animals each, while 24 rats were used for lethal dose (LD50) analysis. The animals were administered ethanolic extracts of fruits of *Acacia nilotica* and flowers of *Calotropis procera* for 5 and 14 days respectively after induction with aspirin. The study was carried out in two phases: The animals were induced with 500 mg/kg body weight of aspirin 12 hours before the commencement of the experiment, the test animals were administered ethanolic extract of fruit of *Acacia nilotica* and flowers of *Calotropis procera* for 5 and 14 days and sacrificed at the end of each test phase respectively, blood was

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collected for biochemical analysis. The liver was harvested at the end of each phase, processed for histological investigation and photomicrographs taken. The result of liver function parameters showed that the mean values of alanine transaminase (ALT) after five-day of treatment decreased non-significantly (p>0.05) in groups 4, 5 and 6 and decreased significantly (p<0.05) in group 4 when compared to normal control. Also, ALT increased significantly (p < 0.05)in group 2. There was a general decreasing trend in the mean values of aspartate transaminase (AST) across all groups except group 2. The mean values of alkaline phosphatase (ALP) decreased significantly (p < 0.05) in all groups, but increased non-significantly in group 2. The same trend was observed in the values of ALT, AST, and ALP after 14 days of treatment. Photomicrograph of the liver section of normal rats showed normal central vein, sinusoids and hepatocytes as well as that of the treated animals. Photomicrograph of liver section of rat administered aspirin only showed slightly dilated portal triad after five days. The study suggests that the ethanolic extracts from fruits of Acacia nilotica and flowers of Calotropis procera may have active ingredients that are capable of improving some liver functions.

Keywords: Acacia nilotica, Calotropis procera, Liver function, Histology, Acute toxicity

INTRODUCTION

Medicinal plants are used as an alternative remedy mostly by people living in developing nations. Hundreds of plants have traditionally been utilized throughout Africa to manage and control opportunistic infections (1). Medicinal plants produce a diverse spectrum of compounds with medicinal properties, making them promising candidates for the development of new antibacterial and antioxidant medications (2)(3). Nigerian ethnic groups have used various plant products for a variety of purposes. Medicinal plants have been used for ages to treat a variety of diseases as well as to protect against disease vectors (4).

Acacia nilotica belongs to the subfamily of Babula tree, also known as Indian gum arabic tree, which is the common name for this tree (5). The species is found in the drier parts of Africa, from Senegal through Egypt and down to South Africa, as well as Asia, from Arabia east to India, Burma, and Sri Lanka. It can resist temperatures as low as -1°C and as high as 50°C, however it is frost susceptible when young. The annual rainfall ranges from 250 to 1500 millimetres. During the dry season, trees are mostly deciduous, while riverine species



can be virtually evergreen (6). Acacia nilotica is a tree that can be used for a variety of purposes. In agroforestry systems all throughout the world, it is employed as a green fertiliser, wood tree, and fodder tree (7). In Niger, it is used as an urban forestry tree species for shading (8). Acacia nilotica is a safe, biodegradable, and renewable source of pharmaceuticals with a high therapeutic index (9). The lifespan of Acacia nilotica is approximately 40 years (10).



Acacia nilotica fruit (11)



Calotrpis procera plant grows in dry habitats with an annual precipitation from 150 to 1000 mm, as well as in areas with excessively drained soil with up to 2000 mm of yearly precipitation. The plant can also be found in common habitats such as roadside and seaside dunes, as well as being commonly disturbed in urban areas. *C. procera* can also be found at elevations of up to 1,000 metres (12). The leaves of *C. procera* were used in sun worship since the Vedic era (13). Hindu healers employed secretions from the root bark to cure skin illnesses, coughs, intestinal worms, ascites, and anasarca, as well as enlargements of the abdominal viscera, among other things (14).



Calotropis procera plant (11)



MATERIALS AND METHODS

Plant material

The dried fruits of *Acacia nilotica* was purchased from old market in Wukari, Nigeria. It was ground into powder. The fresh flowers of *Calotropis procera* was collected from Aguwan Roger Road, Wukari, Nigeria. It was dried at room temperature and then ground into powder. The chemical constituents of the powdered samples were extracted using 70% ethanol.

Extract preparation

Seventy percent (70%) ethanol was prepared and it was used to soak the two grounded samples (fruits of *Acacia nilotica* and flowers of *Calotropis procera*) separately. The mixtures were filtered after 48 hours. The filtrates were concentrated using a water bath set at 68°C in order to eliminate the ethanol. The concentrated extract was diluted with normal saline before administration to the test animals.

Animals care and management

Eighty-four (84) healthy male albino Wister rats (about 8 weeks old) were used for this experiment. The rats were purchased from Hema Farms Federal Housing Estate Bajaburie Yola, Adamawa State, Nigeria. The rats were maintained under standard laboratory conditions and were allowed free access to standard diet and water *ad libitum*. They were allowed to acclimatize for 14 days before the experiment.

Acute toxicity study

The plant extracts were screened for toxicity using the method of (15) with slight modification for oral routes in rats. The method consists of two phases using 24 rats. In the first phase, 3 groups of 3 rats each were administered the extracts in doses of 100, 400, and 800 mg/kg body weight orally and were observed for signs of toxicity and death for 24hours. In the second phase, 3 groups each containing 1 rat were administered new doses of the extract: 1,000, 2,000 and 3,000 mg/kg body weight orally, they were also observed for any sign of toxicity and death for 24 hours. This was done for the two plant parts extracts.



Experimental design

Sixty (60) healthy male albino rats were used for this experiment. The animals were randomly distributed into six (6) groups and received the plant extracts and aspirin orally as follows:

Group	1	2	3	4	5	6
Treatment	Normal	Negative	Aspirin	Aspirin	Aspirin	Aspirin
	control	control:	(500	(500	(500	(500
		Aspirin	mg/kg)	mg/kg)	mg/kg) +	mg/kg) +
		(500	+ Acacia	+ Acacia	Calotropis	Calotropis
		mg/kg)	nilotica	nilotica	procera	procera
			fruit	fruit	flower	flower
			extract	extract	extract	extract
			(200	(400	(200	(400
			mg/kg)	mg/kg)	mg/kg)	mg/kg)

Table 1: Experimental design

Aspirin was given as a single dose of 500 mg/kg body weight to 24hours fasted rats, the animals were allowed to stay for 12 hours before the start of treatment. The ethanolic extracts of the two plant parts were given to the animals for 5 days and 14 days before sacrifice respectively.

Animal sacrifice and blood collection

Five albino rats from each group were sacrificed after 5-day treatment with the plants extract, while the remaining five rats were sacrificed after 14 days of treatment with the plants extract, blood was collected via cardiac puncture into a plain sample bottle container for biochemical analysis and the liver was harvested for histological analysis.

Determination of levels of selected liver marker indices

Levels of selected indices of liver function such as Alanine aminotransferae (ALT), Aspartate aminotranferase (AST), Alkaline phosphatase (ALP), Total protein (TP), Albumin and Bilirubin were determined using an auto-Chemistry analyser (Landwind LW E60B, China).

Histopathological study of the liver

The liver of the animals was harvested and fixed in 10% formalin, dehydrated in gradual ethanol (50-100%), cleared in xylene, and embedded in paraffin wax. The sections, which were 5-6 mm thick was then prepared using rotary microtome (Leica RM 2125 RTS,



Singapore) and stained with hematoxylin and eosin dye for microscopic observation of histopathological changes in the liver.

Statistical analysis

Statistical analysis was carried out using ANOVA and further with Duncan's multiple comparison test and results were expressed as mean \pm standard error. The statistical analysis was performed using Statistical Package for Social Sciences (SPSS) version 23 and significance was at P < 0.05.

RESULTS

Table 2: Acute toxicity test in rats administered ethanolic extracts of fruits of Acacia nilotica (phase 1)

Dose (mg/kg. bw)	Number of rats	Number of dead rats after 24 hours
100	3	0
400	3	0
800	3	0

The animals did not show any sign of toxicity after administration of ethanolic extract of fruits of *Acacia nilotica* at doses 100, 400, and 800 mg/kg body weight, and there was no death recorded after 24hours.

Table 3: Acute toxicity test in rats administered ethanolic extracts of fruits of Acacia
<i>nilotica</i> (phase 2)

Dose (mg/kg. bw)	Number of rats	Number of dead rats after 24 hours
1000	1	0
2000	1	0
3000	1	0

The animals did not show any sign of toxicity after the second phase of LD50, the animals were administered ethanolic extract of fruits of *Acacia nilotica* at doses 1000, 2000, and 3000 mg/kg body weight, and there was no death recorded after 24hour.



Dose (mg/kg. bw)	Number of rats	Number of dead rats after 24 hours
100	3	0
400	3	0
800	3	0

 Table 4: Acute toxicity test in rats administered ethanolic extracts of flowers of

 Calotropis procera (phase 1)

The animals did not show any sign of toxicity after administration of ethanolic extract of flowers of *Calotropis procera* at doses 100, 400, and 800 mg/kg body weight, and there was no death recorded after 24hours.

Table 5: Acute toxicity test in rats administered ethanolic extracts of flowers of Calotropis procera (phase 2)

Dose (mg/kg. bw)	Number of rats	Number of dead rats after 24 hours
1000	1	0
2000	1	0
3000	1	0

The animals did not show any sign of toxicity after the second phase of LD50, the animals were administered ethanolic extract of flowers of *Calotropis procera* at doses 1000, 2000, and 3000 mg/kg body weight, and there was no death recorded after 24hours.



Table 6: Concentration of selected liver function parameters in rats administered ethanolic extracts of fruits of Acacia nilotica and flowers of Calotropis procera (after

Parameters	Group 1 (normal control)	Group 2 (aspirin 500 mg/kg. Bw	Group 3 (aspirin 500 mg/kg. bw + A. nilotica 200 mg/kg. bw)	Group 4 (aspirin 500 mg/kg. bw + A. nilotica 400 mg/kg. bw)	Group 5 (aspirin 500 mg/kg. bw + C. procera 200 mg/kg. bw)	Group 6 (aspirin 500 mg/kg. bw + C. procera 400 mg/kg. bw)
ALT	152.39 ±	190.97 \pm	102.63±	153.33±	$148.05 \pm$	123.56±
(IU/L)	0.99c	3.09 ^d	1.05 ^a	4.55°	2.95°	0.45 ^b
AST	$470.84 \pm$	513.13 ±	73.87 ±	$160.51\pm$	$127.51\pm$	123.09±
(IU/L)	8.07 ^d	31.01 ^d	1.69ª	6.72 ^b	2.34 ^b	4.32 ^b
ALP	12.61 ±	$18.48 \pm$	8.45 ± 0.38^{b}	$7.73 \pm 0.19^{\text{b}}$	2.05 ± 0.26^{a}	2.89 ± 0.31^{a}
(IU/L)	0.54 ^c	0.81 ^d				
ТР	6.22 ±	$6.56 \pm$	6.78 ± 0.02^{a}	6.86 ± 0.15^{a}	7.12 ± 0.10^{a}	8.62 ± 1.03^{b}
(gm/dL)	0.13ª	0.17ª				
Albumin	$2.04 \pm$	2.19 ±	$2.74 \pm 0.04^{\circ}$	3.07 ± 0.08^{d}	2.99 ± 0.02^{d}	0.37 ± 0.05^{a}
(gm/dL)	0.45 ^b	0.06 ^b				
Globulin	4.18 ±	4.37 ±	4.04 ± 0.04^{a}	3.78 ± 0.13^{a}	4.12 ± 0.07 a	8.25 ± 1.07^{b}
(gm/dL)	0.22ª	0.18ª				
TB	$0.55 \pm$	$0.90 \pm$	0.78 ± 0.03^{a}	0.69 ± 0.13^{a}	0.65 ± 0.12^{a}	0.66 ± 0.08^{a}
(mg/dL)	0.03ª	0.22ª				
DB	$0.02 \pm$	$0.31 \pm$	$0.03 \pm 0.00^{\mathrm{a}}$	0.31 ±	0.34 ±	$0.16 \pm$
(mg/dL)	0.00ª	0.20 ^{b,c}		0.03 ^{b,c}	0.09 ^{b,c}	0.04 ^{a,b}
INDB	$0.53 \pm$	$0.38 \pm$	$0.75 \pm 0.03^{\rm b}$	$0.38 \pm$	0.31 ± 0.10^{a}	$0.49 \pm$
(mg/dL)	0.04 ^{a,b}	0.20 ^{a,b}		0.14 ^{a,b}		0.12 ^{a,b}

5 days treatment)

Results represent mean \pm standard error of group results obtained (n=5).

Means in the same row having different superscripts are statistically significant (p < 0.05).

Legend: ALT= Alanine transaminase, AST= Aspartate transaminase, ALP= Alkaline phosphatase, TP= Total protein, TB= Total bilirubin, DB= Direct bilirubin, INDB= Indirect bilirubin

The result of selected liver parameters showed that ALT level increased non-significantly (p>0.05) in group 4 when compared to the control group, it increased significantly in group 2 and decreased significantly in all other test groups except group 5 where it decreased non-significantly. AST increased non-significantly in group 2 when compared to group 1 but decreased significantly (p < 0.05) in all test groups. The level of ALP decreased significantly in all test groups when compared to the control group except group 2. The TP increased non-significantly in all test groups from the control group except group 6 where



it increased significantly. The level of albumin increased non-significantly in group 2 when compared to the control group and increased significantly in groups 3, 4 and 5 while it decreased significantly in group 6. Globulin increased non-significantly in group 2 and significantly in group 6 when compared to group 1 and it decreased non-significantly in all other test groups. Total bilirubin increased non-significantly across all test groups when compared to the control group while direct bilirubin increased significantly across all test groups. The level of indirect bilirubin decreased in all test groups when compared with group 1 except in group 4 where it increased non-significantly.

Table 7: Concentration of selected liver function parameters in rats administered ethanolic extracts of fruits of *Acacia nilotica* and flowers of *Calotropis procera* (after 14 days treatment)

Parameters	Group 1 (normal control)	Group 2 (aspirin 500 mg/kg. Bw	Group 3 (aspirin 500 mg/kg. bw +A. Nilotica 200 mg/kg. bw)	Group 4 (aspirin 500 mg/kg. bw + <i>A.</i> <i>nilotica</i> 400 mg/kg. bw)	Group 5 (aspirin 500 mg/kg. bw + <i>C.</i> <i>procera</i> 200 mg/kg. bw)	Group 6 (aspirin 500 mg/kg. bw + C. procera 400 mg/kg. bw)
ALT	149.46 ±	187.27 ±	117.14 ±	218.46 ± 17.79^{d}	133.70±4.35ª,b	141.48 ±
(IU/L)	5.05 ^b	1.28°	3.92ª			0.88 ^b
AST	396.42 ±	478.43 ±	$160.09\pm$	$143.19 \pm 3.52^{a,b}$	$122.78 \pm 1.95^{a,b}$	118.71 ±
(IU/L)	24.44 ^d	9.98°	1.05 ^b	6 4 6 L 0 05		2.11ª
ALP	11.72 ±	13.42 ±	7.71 ± 0.21^{d}	$6.46 \pm 0.25^{\circ}$	1.77 ± 0.24^{a}	1.85 ± 0.31^{a}
(IU/L)	0.23e	0.72 ^f	5 5 6		5 10 1 0 11	
TP	6.24 ±	5.73 ±	5.73 ±	$6.68 \pm 0.26^{\circ}$	5.42 ± 0.11^{a}	$6.87 \pm 0.20^{\circ}$
(gm/dL)	0.14 ^{b,c}	0.10 ^{a,b}	0.14 ^{a,b}	0.04	0.40 1 0.04	0.07 1.0.00
Albumin	$2.70 \pm$	2.84 ±	2.73 ± 0.02^{b}	$3.24 \pm 0.14^{\circ}$	0.19 ± 0.04^{a}	0.37 ± 0.09^{a}
(gm/dL)	0.04 ^b	0.02 ^b	2 00 1 0 10-	2 42 1 0 40-	5 00 L 0 001	
Globulin	3.53 ±	2.89 ±	3.00 ± 0.12^{a}	3.43 ± 0.18^{a}	$5.23 \pm 0.03^{\text{b}}$	$6.50 \pm 0.26^{\circ}$
(gm/dL)	0.14ª	0.09^{a}	0.70 ±	0.22 ± 0.07	0.70 ± 0.07 h s	$0.(1 \pm 0.07)$
TB	$0.85 \pm 0.02^{\circ}$	$0.62 \pm 0.02^{\rm b}$	$0.78 \pm 0.03^{b,c}$	0.33 ± 0.07^{a}	$0.70 \pm 0.07^{\rm b,c}$	0.61 ± 0.07^{b}
(mg/dL) DB		0.02° $0.29 \pm$		0.17 ± 0.03^{a}	0.20 ± 0.05 h	0.25 ±
		0.29 <u>1</u> 0.08 ^{a,b}	0.15 ± 0.03^{a}	0.17 ± 0.03^{a}	$0.39 \pm 0.05^{\text{b}}$	$0.25 \pm 0.10^{a,b}$
(mg/dL) INDB	0.04^{a} 0.66 ±	$0.08^{a,b}$ $0.32 \pm$	0.63 ± 0.07^{b}	0.15 ± 0.09^{a}	0.31 ± 0.08^{a}	$0.10^{a,b}$ 0.35 ± 0.05^{a}
			$0.03 \pm 0.07^{\circ}$	0.13 ± 0.09^{a}	0.31 ± 0.08^{a}	0.35 ± 0.05^{a}
(mg/dL)	0.06 ^b	0.09ª				

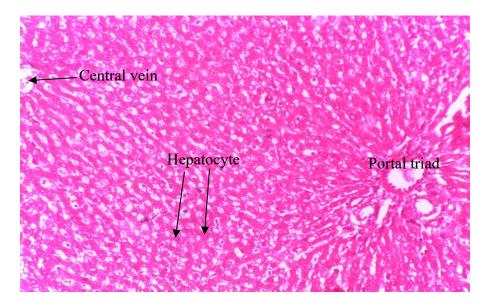
Results represent mean \pm standard error of group results obtained (n=5).

Means in the same row having different superscripts are statistically significant (p<0.05).

Legend: ALT= Alanine transaminase, AST= Aspartate transaminase, ALP= Alkaline phosphatase, TP= Total protein, TB= Total bilirubin, DB= Direct bilirubin, INDB= Indirect bilirubin.



The result showed the liver enzyme ALT when compared to group, increased significantly in groups 2 and group 4 while it decreased in groups 3, 4 and 6. The AST level increased significantly in group 2 and decreased in all other test groups when compared to the control group. The ALP level increased significantly in group 2 but decreased in all test groups when compared to group 1. Total protein level when compared to the control group decreased in all test groups except group 4 and 6 where it increased significantly. Albumin level increased non-significantly across the group except group 5 and 6 where it decreased significantly. Globulin level decreased non-significantly in groups 2, 3 and 4 when compared to the control group and increased significantly in groups 5 and 6. Total bilirubin level was decreased across all treatment groups. Direct bilirubin increased in groups 2, 5 and 6 when compared to the control group and decreased non-significantly in groups 3 and 4. The indirect bilirubin level decreased significantly across the test group except for group 3 where it decreased non-significantly.



Histological analysis results of the liver of the animals after five days treatment

Figure 3: Photomicrograph of liver section of normal control rat (group 1) showing normal central vein, portal triad, sinusoids and lamella of hepatic cells.

Stain: Hematoxylin and Eosin. Mag: x200.



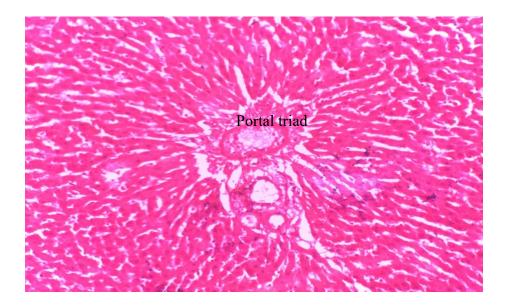


Figure 4: Photomicrograph of liver section of rat administered aspirin (500mg/kg) only (group 2) showing slightly dilated portal triad. However, other stromal elements appear unremarkable.

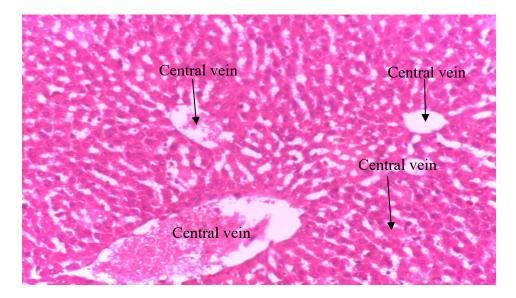


Figure 5: Photomicrograph of liver section of rat administered aspirin (500mg/kg) and *Acacia nilotica* fruit extract (200mg/kg) (group 3) showing two normal and one dilated central vein within a normal tissue stroma. Other tissue elements appear unremarkable.



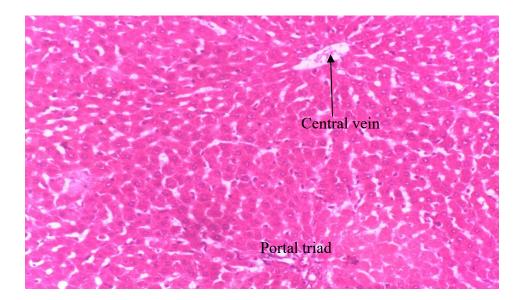


Figure 6: Photomicrograph of liver section of rat administered aspirin (500mg/kg) and *Acacia nilotica* fruit extract (400mg/kg) (group 4) showing normal central vein and portal triad within a normal tissue stroma.

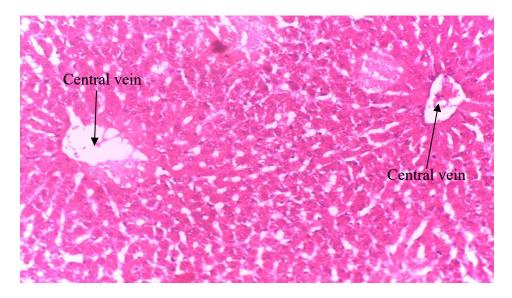


Figure 7: Photomicrograph of liver section of rat administered aspirin (500mg/kg) and *Calotropis procera* flower extract (200mg/kg) (group 5) showing normal central veins within a normal tissue stroma.



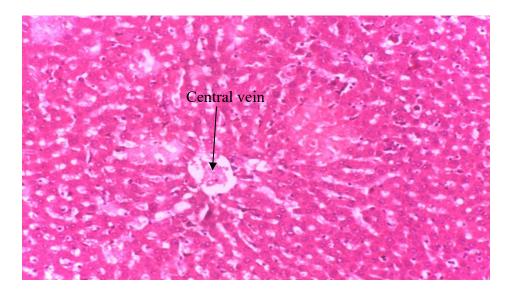
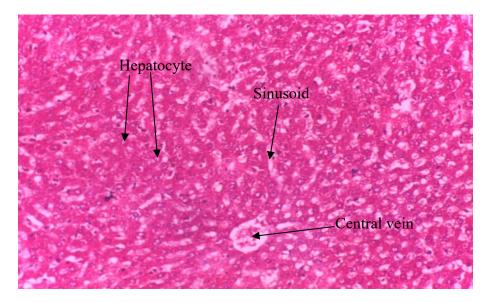


Figure 8: Photomicrograph of liver section of rat administered aspirin (500mg/kg) and *Calotropis procera* flower extract (400mg/kg) (group 6) showing normal central vein within a normal tissue stroma.



Histological analysis results of the liver of the animals after fourteen days treatment

Figure 9: Photomicrograph of liver section of normal rat control (group 1) showing normal central vein, sinusoids and hepatocytes.

Stain: Hematoxylin and Eosin. Mag: x200.



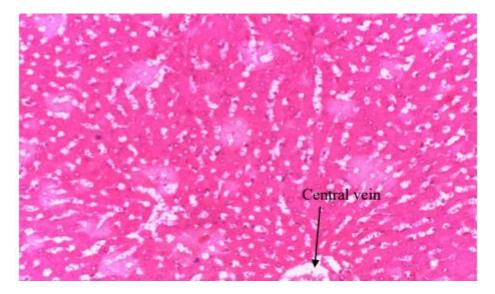


Figure 10: Photomicrograph of liver section of rat administered aspirin (500mg/kg) only (group 2) showing normal central vein, sinusoids and lamella of hepatic cells.

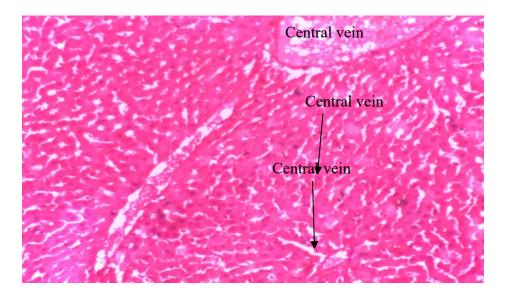


Figure 11: Photomicrograph of liver section of rat administered aspirin (500mg/kg) and *Acacia nilotica* fruit extract (200mg/kg) (group 3) showing a dilated central vein and a normal central vein within a normal tissue stroma.



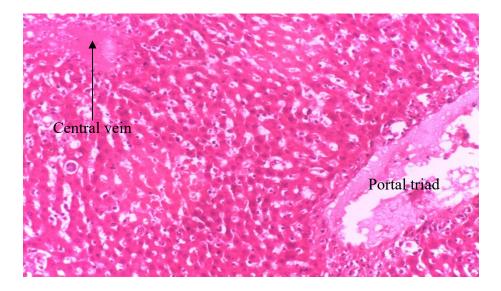


Figure 12: Photomicrograph of liver section of rat administered aspirin (500mg/kg) and *Acacia nilotica* fruit extract (400mg/kg) (group 4) showing one normal central vein and a dilated portal triad within a normal tissue stroma.

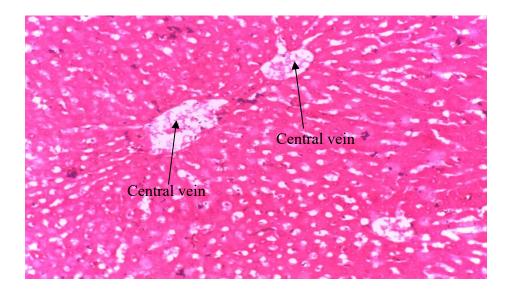


Figure 13: Photomicrograph of liver section of rat administered aspirin (500mg/kg) and *Calotropis procera* flower extract (200mg/kg) (group 5) showing normal central within a normal tissue stroma. Other tissue elements appear unremarkable.



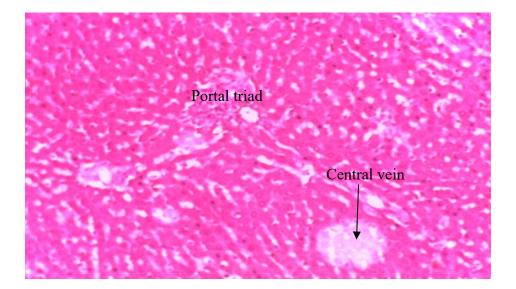


Figure 14: Photomicrograph of liver section of rat administered aspirin (500mg/kg) and *Calotropis procera* flower extract (400mg/kg) (group 6) showing normal central vein and portal triad within a normal tissue stroma.

DISCUSSION

The ethanolic extract of fruits of *Acacia nilotica* at 3000 mg/kg body weight did not produce any mortality after 24 hours of administration. Research carried out by (16) also did not show any toxic effect of the plant extract at that dose. Also (17) reported that the plant part to be safe at 3000 mg/kg body weight, but recorded death at 5000 mg/kg body weight. The ethanolic extracts of flowers of *Calotropis procera* did not show any toxicity at 3000 mg/kg body weight, this is in agreement with the research carried out by (18). A research by (19) observed that the oral administration of a single dose of 3000 mg/kg body weight of flower extract of *Calotropis procera* did not cause any death or toxic symptoms in terms of behavioral changes, skin effects, breathing, food and water intake, therefore, the study concluded the estimated lethal dose of flower extract of *Calotropis procera* to be 5000 mg/kg body weight.

This study showed significant decrease in the liver enzyme (ALT) activity after 5-days and 14-days treatment on the group administered fruit extract of *Acacia nilotica* (200 mg/kg body weight) when compared to the normal control group (Table 6 and 7). The administration of fruits extract of *Acacia nilotica* (400 mg/kg body weight) increased non-significantly (P>0.05) after 5 days of treatment and increased significantly after 14 days of



treatment. The decrease in ALT values implies that the ethanolic extracts of fruit of Acacia nilotica did not cause harm to the liver cells. However, the significant increase recorded after 14-days treatment with 400 mg/kg body weight of the extract may be due to prolong use of the extract. The ALT value of group administered flowers extract of Calotropis procera in low dose decrease non-significantly from the normal group, but the group given 400 mg/kg body weight dose decreased significantly (p < 0.05). The AST and ALP activities decreased significantly in groups administered the fruit extract of Acacia nilotica in doses of 200 mg/kg and 400 mg/kg body weight when compared to the control group (Table 6) and it also decreased significantly after 14 days of treatment (Table 7). This research result is in agreement with the work done (17), but a study by (20) showed a contrasting result. The activities of ALP and AST decreased significantly across all groups administered the flower extract of Calotropis procera when compared with the normal control (Table 6). The administration of the plant part extract after 14 days of treatment decreased the activities of ALT, AST and ALP significantly for groups 5 and 6 except the ALT value of group 6 (Table 7). This research study is in tandem with the work of (19). The liver marker enzymes have significant metabolic activity within cells and are used to detect liver diseases (17). High levels of ALT, AST and ALP are reported in hepatoxicity (19) and a breakdown in the functional integrity of the cell (21). High ALP level is associated with increased synthesis of the enzyme and due to coronary artery disease (17). The decreased levels of the liver marker enzymes in this study is an indication that the two plant parts extract can be used in ameliorating liver complications, and can be used in boosting liver function.

The liver is the major site of plasma protein synthesis and alterations in the level of serum protein is an indication of liver dysfunction. A decrease in serum protein (such as albumin) is an indication of chronic damage to hepatocytes (since almost all proteins are synthesized by the hepatocytes) (17). This study showed an increased level of total protein in all treatment groups when compared to the normal control group, the same trend was seen in the levels of albumin and globulin. This increase implies that the ethanolic extracts of both plant parts have the potency to promote the synthetic function of the liver, since, a decrease in the protein level is an indication of impaired hepatocellular function. The concentration of total bilirubin and direct bilirubin increased slightly after treatment with both plant parts ethanolic extract for 5 days and decreased after 14 days treatment, this is an indication that the liver clears bilirubin properly. The administration of ethanolic extracts of liver



marker enzymes (ALT, AST, ALP) activities in the blood and caused an increase in the concentrations of serum protein. Therefore, the extracts of both plant parts may be utilized clinically in the treatment of hepatic dysfunction.

The result of histopathology of the liver of normal control group after both 5-days and 14days treatment showed a normal central vein, portal triad, sinusoids and lamella of hepatic cells. Group 2 animals administered aspirin only at a dose of 500 mg/kg body weight showed a slightly dilated portal triad but other liver cells appeared unremarkable, however, after 14 days, the result showed a normal central vein, sinusoids and hepatic cells. The photomicrograph of the liver section of rats administered fruit extract of Acacia nilotica in dose of 200 mg/kg body weight after 5 days showed two normal and one dilated central vein with other liver tissues appearing unremarkable, the result also showed same for animals treated for 14 days with the same extract. Administration of the fruit extracts at a dose of 400 mg/kg body weight after 5 days showed a liver section with normal central vein and all other tissues appeared normal, while after 14 days of treatment, the photomicrograph showed a normal central vein and a dilated portal triad within a normal tissue stroma. This research study is in agreement with a work done by (17) where aqueous leaves extracts of Acacia nilotica was used. Groups 5 and 6, administered flower extract of Calotropis procera at 200 mg/kg body weight and 400 mg/kg body weight doses respectively after 5 days treatment showed a normal central vein and other tissues appeared unremarkable. Also, the groups given same doses of the extract after 14 days of treatment showed normal central vein, normal portal triad with other tissues appearing unremarkable. The results of the histological study of the liver showed ability of the two plant parts extracts to treat liver toxicity.

CONCLUSION

The liver marker enzymes have significant metabolic activity within cells and are used to detect liver diseases. This study showed that the ethanolic extracts of fruits of *Acacia nilotica* and flowers of *Calotropis procera* did not show any harm caused on the liver cell and may have ingredients that are capable of improving certain liver functions.

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Competing interest

All the contributing authors declare no competing interest

Authors contribution

Author 1 and 2 designed the experiment, author 2 reviewed the literature, all authors participated in laboratory analysis and author 1 and 2 performed the data analysis.

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