World Journal of Pharmaceutical and Life Sciences
WJPLS
www.wjpls.org
SJIF Impact Factor: 7.409

EVALUATION OF ACUTE AND SUBCHRONIC TOXICITY OF ETHANOL EXTRACT OF SCLEROCARYA BIRREA ROOTS

Antoine Vayaraï Manaoda*[1], Judith Caroline Ngo Nyobe[2], Bigued[4], Jacques Yinyang[1], Charles Christian Ngoule[3], Gisèle Etame Loe[3] and Jean Claude Ndom[2]

[1]Pharmacology and Toxicology Laboratory of The Faculty of Medicine and Pharmaceutical Sciences, University of Douala.
[2]Photochemistry Laboratory of the Faculty of Medicine and Pharmaceutical Sciences, University of Douala.
[3]Department of Pharmaceutical Sciences of the Faculty of Medicine and Pharmaceutical Sciences, University of Douala, Cameroon. B.P. 2701 Douala, Cameroon.
[4]Department of Biological Science of the Faculty of Sciences of the University of Maroua.

***Corresponding Author: Antoine Vayaraï Manaoda**
Pharmacology and Toxicology Laboratory of The Faculty of Medicine and Pharmaceutical Sciences, University of Douala.

Article Received on 08/05/2024 **Article Revised on 29/05/2024** **Article Accepted on 18/06/2024**

SUMMARY
To this day, medicinal plants play a vital role in the art of healing throughout the world. The use of extracts from the bark and leaves of these plants is used to relieve various mild pathologies. However, long-term use of extracts and excessive doses can affect the normal functioning of organs through the presence of phytotoxins. The objective of this study is to evaluate the acute and subchronic toxicity of Sclerocarya birrea extracts, in order to assess whether it confirms its reputation. The leaves and bark of the trunk and roots of Sclerocarya birrea were harvested respectively in December 2022 in Djingliya-Koza (latitude 10.847 longitude 13.85 and lat) in the department of Mayo Tsanaga region of the Far North. Rats of the Wistar strain of the Rattus norvegicus species will be used for toxicity in this work. The phytochemical screening carried out on the parts of the plant revealed the presence of polyphenols, flavonoids and tannins. Acute toxicity was achieved by oral administration at a single dose of 5000 mg/kg. The subchronic toxicity evaluation was carried out orally with the ethanoic extract of the root bark at doses of 500, 1000 and 2000 mg/kg body weight for 28 days. The acute toxicity results were dosed at 5000 mg per kg and showed no mortality or behavioral changes during the 14 days of observation. The lethal dose (LD50) would be greater than 5000 mg/kg body weight. The 1000 and 2000 mg/kg body weight acute toxicity batches showed an increase in body weight throughout the treatment period, but not significantly ($p<0.05$) different from that of the control group. A significant reduction in subchronic toxicity in the 1000 and 2000 mg/kg batches ($p<0.05$) in body weight was observed every week respectively, which proves toxicity at the hepatic and renal level with long-term use.

KEYWORDS: Subchronic toxicity, Sclerocarya birrea extract and roots.

INTRODUCTION

Since ancient times, humanity has used various plants found in its environment to treat and cure all kinds of illnesses. Among which, Sclerocarya birrea known as marula, one of the most used plants of the Anacardiaceae family for the treatment of different pathologies in rural areas.[1] Different parts of this plant are traditionally used: the fruits are eaten or processed to make beer or jam; almonds are eaten or used for oil extraction; the leaves are used as fodder for livestock; stem, root and leaf bark extracts of S. birrea are used against human diseases reported that methanolic extracts of S. birrea roots inhibited the growth of Candida spp. and Cryptococcus neoformans.[2] Methanol and water root extracts have also been shown to act as potent antioxidants and hypoglycemic effects of methanolic and acetone leaf and bark extracts.[3] Additionally, water and acetone extracts of the stem bark of S. birrea showed anticancer and proapoptotic activities.[3] Polar extracts from the leaf bark and stem (inner bark) have antibacterial and antifungal activities.

In Nigeria a further evaluation of subchronic toxicity with a significant reduction ($p < 0.05$) in weight was observed in rats administered extracts at 3,000 mg/kg body weight at the 4th week compared to the control group. For rats receiving 4000 mg/kg body weight, a

significant ($p < 0.05$) reduction in weight was observed from 1st to 4th week.[4,5]

Information on the toxicity of S. birrea bark in Cameroon is not revealed. Therefore, this paper reports the safety evaluation of the plant bark extract by acute and subchronic oral administration in rats.

Long-term use or at inadequate doses can cause organ toxicity due to the presence of phytotoxins in the plant.

MATERIAL AND METHODS

1. Identification of secondary metabolites by reduced pressure chromatography (HPTLC)

The fragments of roots, bark and leaves of Sclerocarya Birrea were collected, treated and dried in a suitable room, protected from the sun and dust, then cut and crushed. Maceration was done with ethanol. The chemical profile of the ethanolic stem extract was developed using ultra-performance liquid chromatography-time-of-flight quadrupole mass spectrometry (UPLC-Q-TOF-MS).[6]

It makes it possible to highlight the subfamily of secondary metabolites. This is a technique that allows the different compounds contained in a mixture to be analyzed quickly, reliably and precisely.

Three steps were then used

Step 1: Submission of migration sites.

The different spots were deposited in horizontal bands. An initial concentration of 10 mg/mL of each extract was prepared in distilled water (10 mg in 1 mL, w/v). After filtration through fine 0.2 µm filters, 20 µL of solution of each extract were automatically deposited in strips by spraying liquid nitrogen onto HPTLC 60F254 type plates (glass holder 20 cm x 10 cm wide). Merck, Darmstadt, Germany). Sample deposition was carried out using a Linomat5 semi-automatic applicator (Camag® Muttenz, Switzerland) controlled by the CATS vision base software (CAMAG® HPTLC). In this study, quercetin, coumarin, and tannic acid were used as positive controls for flavonoids, coumarins, and tannins, respectively.[6]

Step 2: Migration

After drying the spots, the migration was carried out (7 cm path) using a saturated CAMAG type container for 30 minutes with a specific mobile phase depending on the case: hexane - ethyl acetate (20:4 , v/v) for the migration of sterols and triterpenes, ethyl acetate/methanol/water/chloroform (18:2, 4:2.4:6; v/v/v/v) for tannins, acetate ethyl/diethylamide/toluene (70:10:20, v/v/v) for the migration of alkaloids and chloroform/ethyl acetate/methanol/water (6:18:2.4:2.4, v/v) /v/v) for the migration of flavonoids. After development, the individual plates were dried at 110 °C for 2 min on a hot plate (ThermoFischer®) and the chromatographic fingerprints were observed in the visible and UV wavelengths of 254 nm and 366 nm in a camera. C In.[6]

Step 3: Disclosure of secondary metabolites

Neu's reagent and ferric chloride (5%) were used as developers for flavonoids and tannins respectively. The Liberman-Buchard reagent was used for the revelation of sterols and triterpenes. Dragendorff's reagent was used to highlight alkaloids under 366 nm UV light.

2. Quantitative analyzes

- Total phenol content

Quantification of TPC was performed using the Folin-Ciocalteu colorimetric method with slight modifications. First, a stock solution of 1 g/mL was prepared for each extract. 1 mL of stock solution was added to 1 mL of Folin Ciocalteu reagent and incubated for 10 min at 105 °C in an incubator. Subsequently, 2 mL of sodium carbonate solution (7.5%) was added to the previous mixture to incubate for 30 min at room temperature. At the end of this time, the absorbances were read using a UV-visible spectrophotometer at 760 nm (model UV-1800 240V, UV spectrophotometer, Shimadzu Japan). A cascade dilution of the stock solution was used as a blank control. The TPC was then calculated using the gallic acid (GA) equation used as a reference: $Y = 10.46X + 0.03$ ($r2 = 0.99$).[6,7]

3. Acute toxicity

The acute toxicity test was carried out following the "dose adjustment" method of OECD line 425, (2022) at a single dose of 5000 mg/kg. The test was carried out on 6 rats (3 males and three females), clinical examinations were observed for 14 days.

The table below shows the evolution of the clinical parameters of rats for 15 days after a single administration of 5000 mg/kg body weight according to the OECD. No signs of toxicity or death were observed during this test.[4,8]

4. Subchronic toxicity

The acute toxicity test was carried out following the "dose adjustment" method of OECD line 407 (2018). The test substance is administered orally daily at different dose levels to several groups of mice, at one dose level per group, for a period of 28 days.

The study was conducted following OECD guideline 407 (2018). It was carried out on 24 albino Wistar rats divided into eight equal groups of 3 males and 3 females.

Rats were fed and hydrated ad libitum, then weighed every 3 days to calculate weight mass.

A behavioral observation was carried out 3 h after administration of the substances. Then hydration and nutrition were carried out daily for 28 days. During this period, signs of toxicity include coat changes, motility, tremors, mass, grooming, breathing, sensitivity to noise after metal impact, appearance of stools, mobility as well as death were noted.[4,8]

5. Histological study

The histological sections of the liver tissue were made according to the technique proposed by Martoja and Martoja[9], using hematoxylin/eosin. For observation using an optical microscope and taking micrographic photos, the sections examined are the result of technical procedures which require several successive steps: fixation, inclusion, cutting, coloring, assembly.[10]

RESULTS

1. Identification of secondary metabolites by reduced pressure chromatography (HPTLC)

The result of the qualitative phytochemical analysis of the roots, bark and leaves of Scle-rocarya birrea shows the following secondary metabolites: flavonoids, Polyphenols, Phenols, quinones, saponins, tannins, triterpenoids and sterols.

2. Quantitative analyzes

➢ **Content of sterols and triterpenes**

Figure N0: x shows the chromatographic profile of sterols and triterpenes of extracts from the roots, bark and leaves of Sclerocarya Berri. Analysis of this figure shows that the Liberman-Buchard reagent highlighted the sterols and triterpenes which appear in the form of purple, blue, pink, gray, green and red spots on the different chromatographic plates. The 'RSHB and ESB showed stains of greater intensity when observed under UV light.

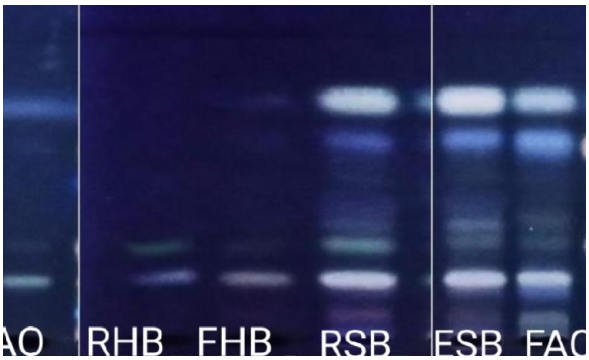


Figure 1: Chromatogram for detection of sterols and triterpenes.

Elution Solvent: Hexane/Ethyl Acetate (20:4, v/v)
Developer: Liebermann Buchard

➢ **Flavonoid content**

Figure 2 shows the chromatographic fingerprint of the flavonoids in the extracts. The chroma-togram revealed blue, green, yellow, orange, greenish and fluorescent dots in the different samples tested. Flavonoids are known to interact with the Nue reagent to generate complexes with brilliant colors that glow under UV light. Yellow spots and greenish spots reveal the presence of flavonols. Furthermore, the presence of flavanones and aurones was indicated by the presence of green spots. We note that all the extracts contain flavonoids. Green spots were observed on the ESB and RSB extracts, which indicates the presence of aurones and flavones in these extracts.

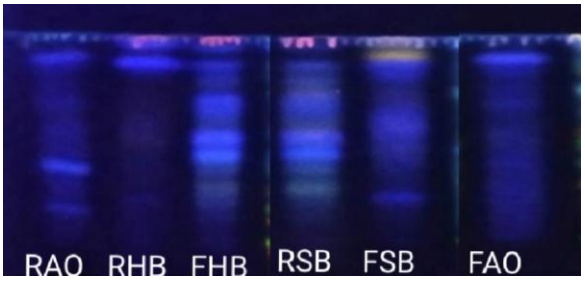


Figure 2: Chromatogram for detection of flavonoids.

Elution solvent: Ethyl acetate/Formic acid/Acetic acid/Water (100:11:11:26, v/v/v/v)
Developer: Neu reagent

➢ **Tannin content**

Figure 3 shows the chromatographic fingerprints of the tannins in the extracts. Spraying with 2% iron(III) chloride solution revealed some spots under UV light after 415 nm within 10 min of incubation. RSB, ESB extracts showed low intensity patches. Little or no staining was observed on the other RSB and BSE extracts.

Figure 3: Chromatogram for tannin detection.

Elution solvent: Chloroform/Ethyl acetate/Methanol/Water (6:18:2.4:2.4, v/v/v/v)
Developer: Iron trichloride (2%)

3. Acute toxicity

There were no signs of toxicity or death of the experimental rats during the 14 days of observations after oral administration of the ethanolic extract of S. birrea roots at a single dose of 5000 mg/kg. A more in-depth evaluation of toxicity carried out by observing the body weight gain did not reveal a significant difference ($p>0.05$) on the other hand, the extracts of 1000 and 2000 mg/kg of body weight compared to their control group have a significant reduction ($p < 0.05$).

4. Subchronic toxicity

➢ **Evaluation of body weight of male and female rats for 28 days**

We carried out weekly monitoring of body weight for a period of one month of experimentation. At the end of the statistical analysis, the average body weights showed a non-significant statistical increase ($P>0.05$) between the different batches.

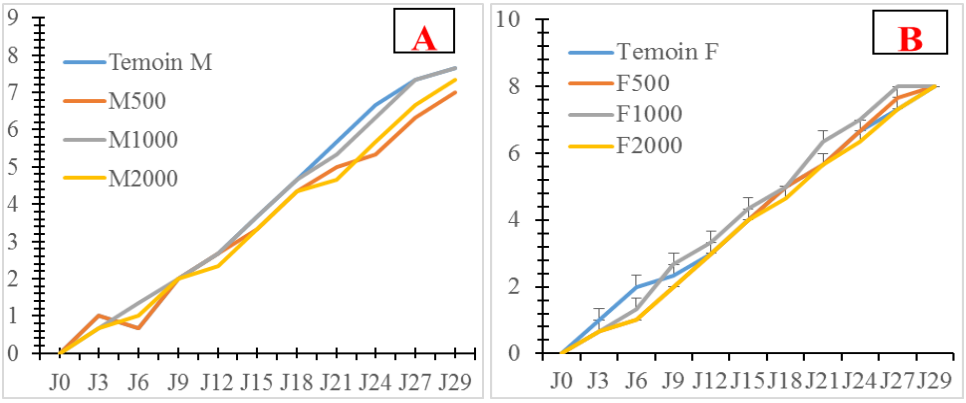
Table I shows us the statistical differences after 28 days of treatment.

After 28 days of treatment, we observed a non-significant increase in body weight of male rats;

- The control batch (168.3±18.23) and the male test batch 500 mg/Kg (175.4±21.61); i.e. an increase of 7.1g in weight in males.
- The control batch (168.3±18.23) and the male test batch 1000 mg/Kg (181.1±26.14); an increase of 12.8g in weight in males.
- The control batch (168.3±18.23) and the male test batch 2000 mg/Kg (190.5±29.79); an increase of 22.2g in weight in males.
- After 28 days of treatment, we observed a non-significant increase in body weight of female rats;
- The control batch (155.7±16.68) and the male test batch 500 mg/Kg (179.2±25.07); i.e. an increase of 23.5g in weight in females.
- The control batch (155.7±16.68) and the female test batch 1000 mg/Kg (185.1±26.55); i.e. an increase of 29.4g in weight in females.
- The control batch (155.7±16.68) and the female test batch 2000 mg/Kg (189.8±28.82) and an increase of 34.1g in weight in females.

Table I: Average weight mass values of male and female rats for 28 days.

Duration	Lot T Males	Lot 500	Lot 1000	Lot 2000
Day 0	160,5±15,58	168,9±17,65	177,6±24,57	184,6±25,59
Days 7	161,8±16,04	170,4±18,95	178,7±25,23	185,4±26,20
Days 14	163,4±17,33	171,9±19,63	179,5±25,96	186,8,±26,78
Days 21	165,9±17,89	173,2±20,36	181,1±26,14	188,2±27,96
Days 28	168.3±18,23	175,4±21,61	183,3±27,28	190,5±29,79
Duration	**Lot females**	**Lot 500**	**Lot 1000**	**Lot 2000**
Day 0	150,4±15,06	174,2±21,11	180,5±25,65	184,7±26,98
Days 7	151,7±15,29	175,7±21,75	181,2±26,05	185,5±26,31
Days 14	152,8±15,92	176,1±21,99	182,7±26,36	186,9±27,78
Days 21	153,7±16,16	177,9±22,56	183,6±27,98	188,3±28,11
Days 28	155.7±16,68	179,2±25,07	185,1±26,55	189,8±28,82



Legendre:

J0….. J29 = (day 0…. Day 29)

Témoin males = Male witness

M 500 = male dose of 500 mg/Kg

M 1000 = male dose of 1000 mg/Kg

M 2000 = male dose of 2000 mg/Kg

Témoin males = female witness

F 500 = female witness of 500 mg/Kg

F 1000 = female witness of 1000 mg/Kg

F 2000= female witness of 2000 mg/Kg

Fig 4 A and B: After the statistical analysis of the average body weights of the rats, the results represented in Table No. show on the one hand, a slight, insignificant increase (P>0.05) between the batch weight 500, 1000 and 2000 mg/kg compared to rats from the control group.

Table II: Weight mass of organs of male and female rats.

DOSE	LIVER	KIDNEYS	SPLEES
witness Lot	8.50 ± 0.48	1.42 ± 0.03	1.08 ± 0.02
Lot M500	5.98 ± 0.51	1.22 ± 0.01	0.61 ± 0.03
Lot M1000	6.34 ± 6.34	1.23 ± 1.23	0.74 ± 0.74
Lot M2000	8.36 ± 1.95	1.27 ± 0.20	1.19 ± 0.41

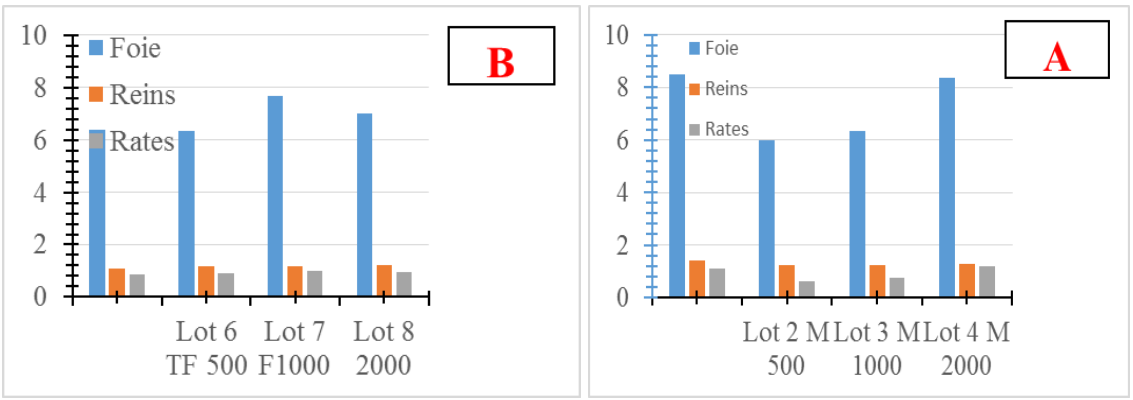
DOSE	LIVER	KIDNEYS	SPLEES
witness Lot	6.40 ± 0.04	1.07 ± 0.02	0.85 ± 0.09
Lot F500	6.32 ± 0.42	1.18 ± 0.08	0.89 ± 0.20
Lot F1000	7.66 ± 0.75	1.16 ± 0.10	1.02 ± 0.32
Lot F2000	6.98 ± 0.55	1.21 ± 0.14	0.96 ± 0.20

The statistical analyzes in Table II show us the following

For the control batch compared to the 500 mg/Kg dose, the values are expressed as mean ± standard deviation; using one-way analysis of variance with a total variance of 0.14%, the significant difference from the control is (P value =0.9913).

The control batch compared to the dose of 1000 mg/Kg, the values are expressed as mean ± standard deviation; using one-way analysis of variance with a total variance of 0.14%, the significant difference from the control is (P value = 0.9911).

The control batch in comparison with the batch of 2000 mg/Kg, the values are expressed as mean ± standard deviation; using one-way analysis of variance with a total variance of 0.84%, the significant difference from the control is (P value = 0.9498).



Legendre: Foie Rein Rates (liver kidneys, splees).

Témoin males = Male witness **Témoin male**s = female witness
M 500 = male dose of 500 mg/Kg **F 500** = female witness of 500 mg/Kg
M 1000 = male dose of 1000 mg/Kg F 1000 = female witness of 1000 mg/Kg
M 2000 = male dose of 2000 mg/Kg F 2000= female witness of 2000 mg/Kg

Figure 5 A and B: The analysis of the evolution of the relative weight of livers, kidneys and rats is made by evaluating the index of their masses, the results obtained show a non-significant decrease for these parameters studied (P>0.05), in particular between the relative weight of the liver in control rats (0.044±0.0035g) and rats treated with S. Birrea extracts (0.040±0.0056g). On the other hand, we note that there is no significant variation concerning the renal and pancreatic index for all the animals.

5. Biochemical parameters

➢ **Aspartate aminotransferase assay (ASAT/TGO)**

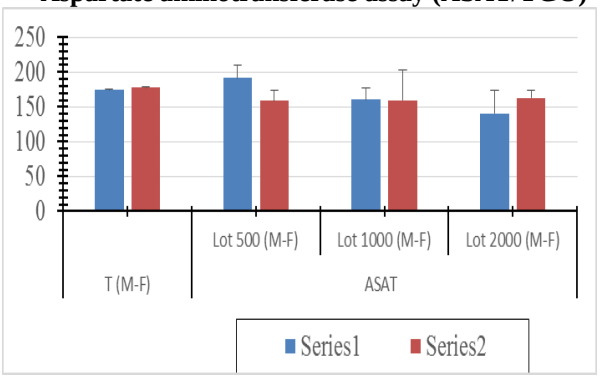


Figure 6: The ASAT content decreased significantly in the test rats than in the control rats; moreover, we noted an increase in the male rats in the 500 mg/Kg batch compared to the male control batch.

➢ **Dosage of alaline aminotransferase (ALT/TGP) in males and females**

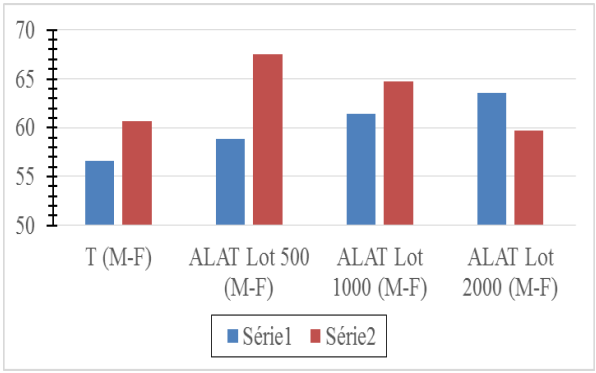


Figure 7: the ALT content increased significantly in all test batches in male rats, moreover in female rats an increase was observed in the 500 mg/Kg batch and tended to gradually decrease in the 1000 and 2000 mg/kg batches. Kg.

➢ **Urea dosage**

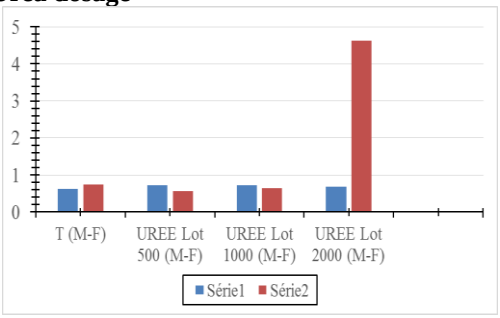


Figure 8: the urea content increased significantly in the 2000 mg/Kg test batch in male rats, otherwise the other batches without any change in the control groups.

➢ **Creatinine dosage**

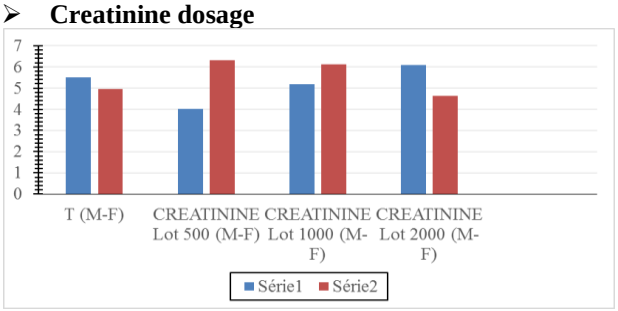


Figure 9: La teneur en créatinine est significativement augmentée dans tous les lots test 500 et 1ooo mg/Kg chez les rats mâles et femelles, par ailleurs on note une diminution de créatique dans le lot test 2000 mg/Kg chez les femelles.

6. Statistical analyzes

Subchronic toxicity: As shown in the table, rats in the group treated with the extract at a dose of 1000 and 2000 mg/kg/day presented hepatic and renal function indices (ASAT ALAT UREE and CREATINE) significantly (P value < 0.0001) higher than the control while no significant difference (p> 0.05) was observed at lower doses. It was also observed that UREE and CREATINETB were significantly lower only for rats given up to 2000 mg/kg/day of extract. For renal function indices (creatinine and urea) in rats having received ethanolic extracts of S. birrea roots), no significant difference was observed in the group having received 500 mg/kg/day compared to the control, while those administered with 2000 and 2000 mg/kg/day of the extract showed a significant increase (P value < 0.0001) in renal function parameters.

Les valeurs sont exprimées en moyenne ± écart type ; en utilisant l'analyse de variance à un facteur avec un total de variance de 98.74%, la différence significative par rapport au témoin est (P value < 0.0001).

Table III: Variation of biochemical parameters.

DOSE	ASAT	ALAT	UREA	CREATINE
witness Lot M	191.33±30.92	56.60 ± 0.85	0.63 ± 0.05	5.50 ±0.40
M500	191.33 ± 30.92	58.86 ± 7.63	0.71 ± 0.07	4.03 ± 1.60
M1000	160.66 ± 27.42	61.40 ± 4.95	0.73 ± 0.15	5.16 ± 0.23
M2000	140.66 ± 58.60	63.56 ± 6.79	0.68 ± 0.17	6.10 ± 1.56
DOSE	**ASAT**	**ALAT**	**UREA**	**CREATINE**
witness Lot F	177.66 ± 2.08	60.63 ± 6.28	0.73 ± 0.18	4.96 ± 0.05
F500	158.66 ± 26.63	67.50 ± 7.55	0.57 ± 0.14	6.30 ± 0.26
F1000	159.33 ± 76.37	64.76 ± 5.56	0.65 ± 0.12	6.13 ± 1.70
F2000	163.33 ± 19.50	59.70 ± 6.53	0.76 ± 0.11	4.63 ± 0.51

Values are expressed as mean ± standard deviation; using one-way analysis of variance with a total variance of 98.74%, the significant difference compared to the control is (P value < 0.0001).

Subchronic toxicity: As shown in the table, rats in the group treated with S. Birrea extract at the dose of 500, 1000 and 2000 mg/kg/day showed liver function indices with est (P value =0.9913) significantly higher than the control while no significant difference was observed at lower doses. Values are expressed as mean ± standard deviation; using one-way analysis of variance with a total variance of 0.14%, the significant difference from the control is (P value =0.991.

Table Iv: Statistical analysis of organ weight of male and female rats.

DOSE	LIVER	KIDNEYS	SPLEES
witness Lot	8.50 ± 0.48	1.42 ± 0.03	1.08 ± 0.02
M500	5.98 ± 0.51	1.22 ± 0.01	0.61 ± 0.03
M1000	6.34 ± 6.34	1.23 ± 1.23	0.74 ± 0.74
M2000	8.36 ± 1.95	1.27 ± 0.20	1.19 ± 0.41
DOSE	**LIVER**	**KIDNEYS**	**SPLEES**
witness Lot	6.40 ± 0.04	1.07 ± 0.02	0.85 ± 0.09
F500	6.32 ± 0.42	1.18 ± 0.08	0.89 ± 0.20
F1000	7.66 ± 0.75	1.16 ± 0.10	1.02 ± 0.32
F2000	6.98 ± 0.55	1.21 ± 0.14	0.96 ± 0.20

The Statistical of the evolution of the relative weight of livers, kidneys and spleens is made by evaluating the index of their masses, the results obtained show a non-significant decrease for these parameters studied (P>0.05), particularly between the relative weight of the liver in control rats (0.044±0.0035g) and rats treated with S. Birrea extracts (0.040±0.0056g). On the other hand, we note that there is no significant variation concerning the renal and pancreatic index for all the animals.

Values are expressed as mean ± standard deviation; using one-way analysis of variance with a total variance of 0.14%, the significant difference from the control is (P value = 0.0911).

7. Histology's of the organs of male rats at 28 days of treatment

Histological sections of the liver, kidneys and spleen of rats in both females and males are presented respectively in the figures below. Certain structural differences between the organs of treated rats and controls were observed in the sections produced.

The figure shows the effects of treatment on the microarchitecture of the kidney and liver and spleen in rats. It appears from this figure that normal rats as well as those receiving the plant at different doses present a typical renal parenchyma with a glomerulus, a urinary space, distinct distal and proximal convoluted tubules and a typical hepatic parenchyma with a bile canaliculus, a portal vein, a hepatic artery and distinct hepatocytes. It is the same for the parenchyma of the spleen which is normal with well differentiated white and red pulps.

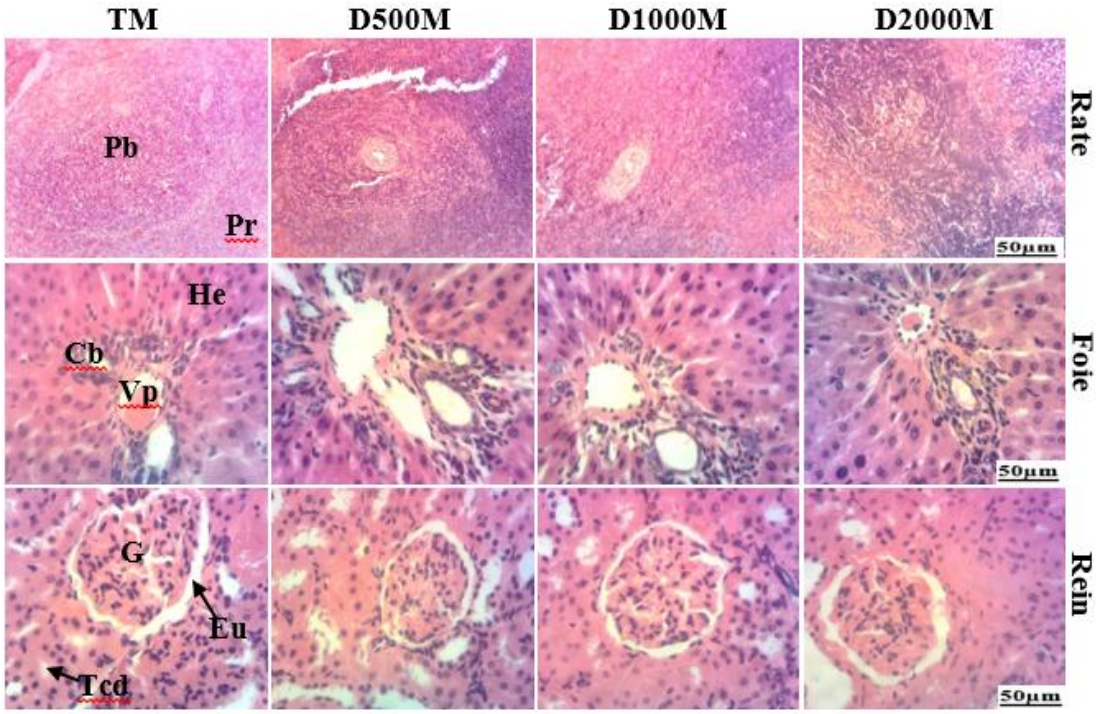


Figure 10: Histology's of the organs of males rats at 28 days of treatment.

Kidney: G = Glomerulus, Eu = Urinary space, Tcd = Distal convoluted tubule, Tcp = Proximal convoluted tubule. Liver: Vp = Portal vein, He = Hepatocyte, Cb = Bile canaliculus, Ah = Hepatic artery. Missed ; Pb = White pulp, Pr = Red pulp, Lung.

The figure shows the effects of the treatment on the microarchitecture of the kidney and liver and spleen in rats. It appears from this figure that normal rats as well as those receiving the plant at different doses present a typical renal parenchyma with a glomerulus, a urinary space, distinct distal and proximal convoluted tubules and a typical hepatic parenchyma with a bile canaliculus, a portal vein, a hepatic artery and distinct hepatocytes. The same is true of the parenchyma of the spleen which is normal with well-differentiated white and red pulps.

Kidney: G = Glomerulus, Eu = Urinary space, Tcd = Distal convoluted tubule, Tcp = Proximal convoluted tubule. Liver: Vp = Portal vein, He = Hepatocyte, Cb = Bile canaliculus, Ah = Hepatic artery. Missed; Pb = White pulp, Pr = Red pulp, Lung.

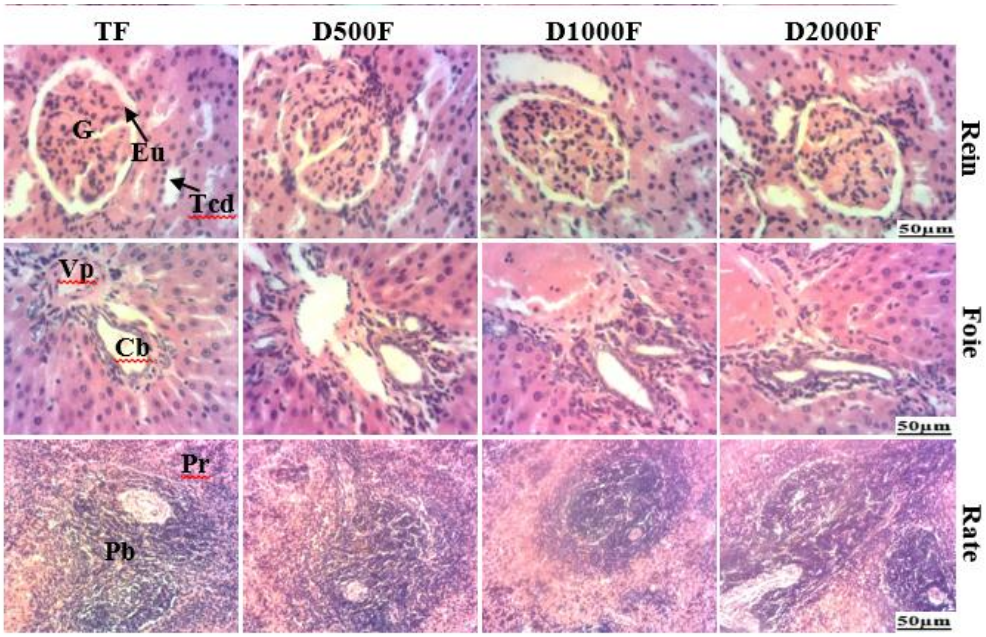


Figure 11: Histology's of the organs of females rats at 28 days of treatment.

Kidney: G = Glomerulus, Eu = Urinary space, Tcd = Distal convoluted tubule, Tcp = Proximal convoluted tubule. Liver: Vp = Portal vein, He = Hepatocyte, Cb = Bile canaliculus, Ah = Hepatic artery. Missed ; Pb = White pulp, Pr = Red pulp, Lung.

DISCUSSION

The ethanolic extract of the roots of S. birrea presents higher levels of phenolic compounds, flavonoids and tannins which corroborates with the studies done by Muhammad Salihu et al.[3,13]

Acute toxicity (LD50): An acute toxicity test at 5000 mg/kg body weight of ethanol extracts of Sclerocarya Birrea roots produced no mortality after 48 hours of observation nor change in behavior after 14 days, which indicates that the average lethal dose (LD50) of the extract would be beyond 5000 mg/kg of body weight.[4]

Subacute toxicity by daily oral administration over 28 days of the ethanolic extract of Sclerocarya Birrea at doses of 500, 1000 and 2000 mg/kg body weight did not result in any significant change in the body weight of rats treated with compared to those who received distilled water in both females and males. This would mean that at these doses and by this route of administration, the ethanolic extract of S. Birrea would have no effect on the normal growth of animals.[4,5] Changes in body weight have been observed as an indicator of adverse effects of extracts and foods. The relative weights of the liver, spleen and kidneys of animals in both females and males are virtually identical. This suggests that at these doses and by this route of administration, the ethanolic extract of Sclerocarya Birrea has no effect on these organs. No significant changes in serum ALT, AST, creatinine and urea levels were recorded the result is similar to that of Immaco-lata Faraone et al 2020 Tinotenda Shoko1 et al.[11,12]

This would mean that at these doses and by this route of administration, the ethanolic extract of S. Birrea would have no effect on renal function. Serum creatinine and urea levels help assess kidney function. In addition, histological sections of the kidney showed no tissue damage. In addition, histological sections of the liver showed no tissue damage. Alanine aminotransferase is a specific and cytological enzyme secreted in liver cells from where it is released into the blood in cases of hepatic cell necrosis. It is an important indicator in acute hepatic necrosis (Hussain et al., 2018). No significant difference was recorded in histological parameters.

CONCLUSION

In this study, our results indicated that the ethanolic extract of S. birrea roots presents higher levels of phenolic compounds, flavonoids and tannins.

This work allowed us to demonstrate the effect of oral administration of the ethanolic extract of Sclerocarya Birrea at a dose of (500, 1000 and 2000 mg/kg of body weight) for one month on the liver, spleen and kidney of white Rattus norvegicus rats.

At the end of the experiment, the results obtained showed that the administration of the ethanolic extract of Sclerocarya Birrea for one month caused non-significant changes in the weight of the animals compared to control rats. At the doses studied, the ethanolic extract of Sclerocarya Birrea had no effect on animal growth. The ethanolic extract of Sclerocarya Birrea also has no significant effect on renal, pancreatic and hepatic functions, on the biochemical and histological parameters of the liver, spleen and kidneys.

At the subchronic duration, Sclerocarya Birrea is not toxic at the doses studied.

REFERENCE

1. World Health Organization. WHO Strategy for Traditional Medicine 2014-2023. Geneva: World Health Organization, 2013.
2. Benmounah IH, Bensaad A, Imene E by: DB. Formulation of a Cough Syrup Based on Medicinal Plants. Ivy (Hedera helix L), Thyme (Thymus vulgaris L) and Myrtle (Myrtus com-munis L), 2020.
3. STUDY AND EVALUATION OF THE THERAPEUTIC PROPERTIES OF A MEDICINAL PLANT "Rhus tripartitum.
4. MAHOMET S1 HASSAN et al. ACUTE AND SUBCHRONIC TOXICITY STUDIES OF Sclerocarya birrea KERNEL EXTRACT IN RATS. (Hassan et al., 2010; Ojewole et al., 2011)
5. Mawoza T, Tagwireyi D, Nhachi C. Acute and sub-chronic toxicity studies of an aqueous stem bark extract of Sclerocarya birrea using a rat model, 2016; 7.
6. Windingoud i Rimwagna Christia n Ouedraog et al. Phytochemica l study, antioxidan t an d vasodilatation activities of leafy stem extracts of Flemingi a fagine a Guill. & Perr. (Barker), a medicina l plan t used fo r th e traditiona l treatmen t of arterial hypertension. Pharmacological Research - Modern Chinese Medicine xxx (xxxx) 100231.
7. Flore NJ, Abdoul KI, Ngotta BJB, Wouokoue TJB, Froumsia M, Gilbert T. Floristic diversity and regeneration of wild edible fruit species in the Reserve of Moutourwa and its surroundings (Far-North Cameroon). J Ecol Nat Environ, 2022.
8. Sene AL, Niang K, Faye G, Ayessou N, Sagna MB, Cisse M, et al. Identification of the uses of Scle-rocarya birrea (A. Rich) hoscht in the Ferlo zone (Senegal) and evaluation of the biochemical and nutritional potential of its fruit. Afr J Food Agric Nutr Dev. 201898. Moussa AH, Mahamadou EG, Rayanatou IA. Socio-economic characterization and identification of uses of Sclerocarya birrea (A. Rich.) Hochst. In Niger. J Appl Biosci, 2022.
9. Mguni S, Chiwara P, Gwate O. Exploring the utilisation and management of Sclerocarya birrea (A. Rich.) Hochst (Marula tree) in the Beitbridge district of southern Zimbabwe. Cogent Soc Scientifique du 19 oct 2023]
10. Nyaka AICN, Fadimatou SNMZ, Dzokouo CUD, Bourou S, Yaouba A. Antifungal effect of two plant extracts on pathogens identified on cashew fruits (Anacar-diumouest L.) in North Cameroon. Int J Biol Chem Sci., 2021.
11. Immacolata Faraone1,2, Daniela Russo1,2 & Luigi Milella1,2 Comparison of health-promoting metabolite antioxidant effects in Sclerocarya birrea (A. Rich.) Hochst extracts MEDICINAL PLANT COMMUNICATIONS Med Plant Commun, 2020; 3(4): 68-73.
12. Tinotenda Shoko1, Vinesh J. Maharaj1*, Dashnie Naidoo2, Malefa Tselanyane2, Rudzani Nthambeleni2, Eric Khorombi2 and Zeno ApostolidesAnti-aging potential of extracts from Sclerocarya birrea (A. Rich.) Hochst and its chemical profiling by UPLC-Q-TOF-MS (2018).
13. Muhammad Salihu Abdallah1,2, Muskhazli Mustafa1, Meenakshii A/P. Nallappan1, Sangho Choi 3, Jin-Hyub Paik 3 and Go Rusea1 *Determination of phenolic and flavonoid compounds of some useful medicinal plants and fractionation substances guided by bioassay of stem (bark) extract of Sclerocarya birrea (A. Rich) and their effectiveness against Salmonella typhi, July 27, 2021.