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Hepatotoxic and nephrotoxic activities of aqueous root extracts of *Dichrostachys cinerea* in male Wistar rats

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ABSTRACT**:** Root extracts of *Dichrostachys cinerea* are used locally for the treatment of various medical conditions such as abdominal disorders, malaria, liver disorders, catarrh, bronchitis, pneumonia, asthma, tuberculosis, oedema, blennorrhoea, orchitis, venereal diseases, pains, anaemia, infertility, gynaecological disorders and as an aphrodisiac. This study seeks to evaluate the toxicity of hot aqueous root extract of *Dichrostachys cinerea* (HAREDC) on liver and kidney function indices in wistar rats. The extract was subjected to secondary metabolite analysis. Forty male rats were randomly divided into groups A – D. Group A served as control and received distilled water while groups B, C and D received 50, 100 and 200 mg/kg body weight (BW) of HAREDC respectively. The animals were treated for 1 and 21 days and sacrificed 24 h after the last treatment administration. Their livers, kidneys and blood were obtained and used for analysis of selected parameters of liver and kidney functions. The extract contained alkaloids, tannins, flavonoids and steroids to the tune of 7.35, 13.88, 15.10 and 3.06 %/2 g sample respectively while saponins, terpenoids, anthraquinones and cardiac glycosides were not detected. Extract significantly increased liver and serum aspartate aminotransferase (AST), serum alanine aminotransferase (ALT), kidney and serum gammaglutamyl transferase (GGT) and serum urea levels while it decreased the levels of liver ALT, serum total protein, creatinine, sodium, calcium and phosphate. In conclusion, acute doses of the HAREDC elicited deleterious effect on rat liver and hampered kidneys excretory functions. It is however relatively safer after extended usage. Its use in ethnomedicine should be done with caution.

Keywords: *Dichrostachys cinerea*, Hepatotoxicity, Nephrotoxicity, Extract, Ethnomedicine

Introduction

Plants are the chief source of bioactive compounds, many of which have proven useful for treating various health conditions as well as for various other uses. Medicinal plants are a rich source of ingredients which can be used in drug development and synthesis (Singh, 2015). Traditional medicine is known as the preferred primary health care system in many communities, over 60 % of the world's population and about 80 % in developing countries depend on medicinal plants for their medical needs. This is due to its

affordability, accessibility and low cost (Mintah, 2019). The use of traditional medicine and medicinal plants in most developing countries, as a basis for the maintenance of good health, has been widely observed (Singh, 2015). However, the bioactive compounds in medicinal plants despite having healing properties, also have the potential to cause harm. It is this tendency to be injurious that necessitates toxicity evaluation**.**

Dichrostachys cinerea is commonly known as Sickle bush, Bell mimosa, Chinese lantern tree or Kalahari Christmas tree. It is identified by different names by the ethnic groups in Nigeria. Hausas call it Dundu, the Yorubas call it Kara, and the Igbos call it Ami-ogwu (Quattrocchi, 2012). *Dichrostachys cinerea* is a semi-deciduous to deciduous fast growing tree of the family Mimosaceae and typically grows up to 7 meters in height. It is characterized by strong alternate smooth spines (up to 8 cm long), dark grey-brown fractures on old branches and stems and bark on younger branches (El-Sharawy *et al*., 2017). It is also characterized by compound and pinnate leaves and bicoloured fragrant flowers (Mudzengi *et al.*, 2014; Sabo *et al*., 2017). It is native to Africa and Asia, where it is common in the tropics of Africa, the Sahelian and Sudanian ecozones, and the South Arabian Peninsula (Mudzengi *et al.*, 2014). Traditionally, the leaves are used as anti-inflammatory and in the treatment of pneumonia and leprosy. The dried bark is used to treat diarrhea and postpartum pain. The root is used in the treatment of syphilis and leprosy and as an aphrodisiac (Sabo *et al*., 2017).

Leaf, root and bark extracts of *Dichrostachys cinerea* have been found to exhibit antibacterial and anticandidal activities, effective against *Staphylococcus aureus, Bacillus subtilis, Escherichia coli, Pseudomonas aeruginosa*, and *Candida albicans* (Neondo *et al*., 2012). Dose dependent relaxation effect of *Dichrostachys cinerea* extract was demonstrated on trachea preparations of guinea pigs (Aworet-Samseny *et al.*, 2011). El- Sharawy *et al.* (2017) isolated ten phenolic compounds namely; apigenin-7-O-apiosyl (1-2) glucoside, chrysoeriol-7-O- apiosyl (1-2) glucoside, Clovamide (N- caffeoyl-L-DOPA), Quercetin-3-O-rhamnopyranoside, myricetin-3-O-glucopyranoside, quercetin-3-O-glucopyranoside, myricetin, apigenin and kaempferol from aqueous and ethanolic leaf extracts of *Dichrostachys cinerea*, of which clovamide was found to be the major constituent responsible for antiplasmodial and antiviral activity of crude extracts of *Dichrostachys cinerea*.

Studies have shown *Dichrostachys cinerea* extracts to exhibit significant antimalarial activity (Kweyamba *et al*., 2019), antidiarrhoeal activity (Jayakumari *et al.*, 2011), analgesic and anti-inflammatory activities (Susithra and Jayakumari, 2018), and anticancer activity on brain tumor cell line and liver carcinoma cell line (Zeid *et al.*, 2008). Methanolic extract of *Dichrostachys cinerea* leaf was shown to have significant hepatoprotective effect and has a lethal dose above 3500 mg/kg (Babu *et al.*, 2011). Methanolic extract of *Dichrostachys cinerea* root was found to show positive effect of nullifying the viper venom toxicity in mice (Mishal, 2002). Brine shrimp toxicity assay of leaf, stem bark and roots showed that the leaf has the highest toxicity with LC_{50} of 2000 ppm (Neondo *et al*., 2012). Due to the vast medicinal applications of *Dichrostachys cinerea* root, it is necessary to study its safety or otherwise in mammals. In light of this, this study was conducted to determine the toxicity potential of aqueous root extract of *Dichrostachys cinerea* on liver and kidney function parameters in Wistar rats.

Materials and Methods

Materials

Plant collection and authentication

Dichrostachys cinerea roots were collected from National Electric Power Authority (NEPA), Mile 7, Zaria road, Jos, Plateau State, Nigeria. It was identified and authenticated at the Herbarium Unit of the Department of Plant Science and Biotechnology, Faculty of Natural Sciences, University of Jos, where a voucher specimen was deposited (JUHN20000303).

Experimental Animals

Forty (40) male Wistar rats of average weight of 150.54 ± 15.05g were obtained from the Animal House Unit, Department of Pharmacology, Faculty of Pharmaceutical Sciences, University of Jos, Jos, Plateau State, Nigeria. The rats were housed in plastic cages and fed standard rat pellets (Top Feeds Ltd, Ogorode Industrial Estate, Sapele, Delta State, Nigeria) and tap water *ad libitum* during the experiment.

Ethical approval

This study was approved by the University of Jos Institutional Ethical Committee on Animal Care and Use, and was assigned approval number UJ/FPS/F17-00379.

Preparation of extracts

Roots of *Dichrostachys cinerea* were washed, chopped to pieces and then air-dried at room temperature (25 ^{o}C) for 3 weeks. The chopped roots were further oven-dried at 40 ^{o}C (Carbolite PF 200, Keison Products, Essex, United Kingdom) to constant weight. The dried roots were ground into fine powder using a hammer mill (Model PC 200 x 300, DEWO Machinery Company Ltd, Zhengzhou, Henan, China). Hot aqueous extract of *Dichrostachys cinerea* root was prepared by adding 600 g of fine root powder to 2500 ml of boiled distilled water. The mixture was left to stand for 48 h in a refrigerator at 4 ^{o}C, after which it was filtered using clean white muslin fabric. The extract was then concentrated in an oven at 40 ^{o}C (Carbolite PF 200, Keison Products, Essex, United Kingdom) and the yield calculated to be 10.59 % (w/w). Extract was reconstituted in distilled water to give the doses of 50, 100 and 200 mg/kg BW used in this study.

Animal grouping and treatment administration

Forty (40) male Wistar rats were randomly assigned into two groups (alpha and beta) consisting of 20 rats each. Each of the two groups was further divided into 4 groups (i.e. A - D). Group A served as the control and was administered with distilled water only while groups B, C and D received 50, 100 and 200 mg/kg body weight doses of the extract respectively. The first group (alpha) received treatment for 1 day (acute dose), and the second group (beta) received treatment for 21 days (sub-acute dose). Extract was administered orally using an oral cannula. Rats were anaesthetized using diethyl ether and sacrificed 24 h after the last treatment administration. Their blood, liver and kidney were collected into plain tubes for determination of liver and kidney function indices.

Assay kits and reagents

Assay kits used for total protein, urea, sodium ion, calcium ion, potassium ion, phosphate, alanine aminotransferase, aspartate aminotransferase, albumin and creatinine were products of Fortress diagnostics, United Kingdom. Kits used for alkaline phosphatase and γ-glutamyltransferase were products of Pointe Scientific, Belgium.

Preparation and storage of serum

Blood sample for each animal was collected into a sterile vacutainer bottle and left to stand for 10 min to clot. They were thereafter centrifuged at 9000 rpm for 5 min (Allegra X-30, Beckman Coulter Life Sciences, Indianapolis, United States). Serum samples obtained were then collected into plain tubes using Pasteur's pipette. Serum was stored in a refrigerator (4^{o}C) until required.

Preparation and storage of tissue

Liver and Kidney samples were collected into sterile bottles containing cold 0.25 M sucrose solution as soon as they were harvested. They were then homogenized mechanically (Mixer homogenizer 115V, Thomas Scientific, Swedesboro, NJ 08085, USA) in ice cold 0.25 M sucrose in the ratio 1:5 (1g of tissue to 5 ml sucrose) in an ice bath. Homogenates were then centrifuged at 7000 rpm for 5 min (Mixer homogenizer 115V, Thomas Scientific, Swedesboro, NJ 08085, USA).

Determination of biochemical parameters

All analysis were carried using a microplate reader (SpectraMax 340PC384 Absorbance Microplate Reader, Molecular Devices, San Jose, California, USA) according to the test kit manufacturer's prescribed protocol. The methods described by Tietz (1995) were used for the analysis of aspartate aminotransferase, alanine aminotransferase, total protein, creatinine, phosphate and calcium. Sodium, potassium, alkaline phosphatase, gammaglutamyl transferase, urea and albumin were analysed by the methods described by Trinder (1951), Henry (2001), Wright et al. (1972), Szasz (1976), Fawcett and Scott (1960) and Tietz (1976) respectively.

Secondary Metabolite Analysis

Secondary metabolites screening

Crude hot aqueous extract of *Dichrostachys cinerea* root was screened for the presence of alkaloids, tannins, anthraquinones and cardiac glycosides using the methods of Trease and Evan (1978). Flavonoids was determined using the method of Trease and Evan (1984). Saponins, steroids and terpenoids were determined using the methods of Ayoola *et al*. (2008), Sofowora (1982) and Bot *et al*. (2007) respectively.

Secondary metabolites quantification

The quantitative secondary analysis of hot aqueous extract *Dichrostachys cinerea* root was determined gravimetrically. Alkaloid and flavonoids were quantified using to the method of Ezeonu and Ejikeme (2016). Tannins and steroids were quantified using to the methods of Makkar *et al*. (1993) and Osuagwu and Ihenwosu (2014) respectively.

Statistical Analysis

Laboratory-generated data were subjected to statistical analysis using the IBM® Statistical Package for Social Sciences (SPSS) Software. All significant differences were determined by one way Analysis of Variance (ANOVA) and Post Hoc multiple comparisons were done using Duncan's multiple range test. The significance level was set at $p < 0.05$. Values are presented as mean ± SEM.

Results

Secondary metabolite composition

Secondary metabolites analysis revealed the presence of alkaloids, tannins, flavonoids and steroids. Flavonoids had the highest concentration (15.10 %/2g sample) while steroids had the lowest (3.06 %/2g sample). Saponins, terpenoids, anthraquinones and cardiac glycosides were not detected (Table 1).

Liver function indices

There was a dose-dependent increase ($p < 0.05$) in liver aspartate aminotransferase (AST) activity in the extract-treated animals when compared with the control animals after the first extract administration. Similarly, at day 21, the activity of AST in animals administered 100 and 200 mg/kg BW of the extract also significant increased ($p < 0.05$) when compared with the control animals (Table 2). In the serum, AST activity was significantly increased ($p < 0.05$) in the animals treated with the 100 and 200 mg/kg BW of the extract at day 1 of treatment while only the animals that received the 200 mg/kg BW of the extract for 21 days showed a significantly higher ($p < 0.05$) serum AST activity when compared with the control group (Table 2).

The activity of alanine aminotransferase (ALT) in the liver was significantly decreased ($p < 0.05$) in the animals treated with all the doses of the extract at day 1. The recorded decrease was also dose-dependent. However, there was no significant difference ($p > 0.05$) in liver ALT of all the extract-treated animals when compared with the control group after 21 days of treatment. In the serum, ALT activity was significantly

increased in a dose dependent manner at day 1. There was however no significant difference ($p > 0.05$) in serum ALT activity of the extract-treated animals when compared with the control group (Table 2).

Table 1: Secondary metabolite composition of hot aqueous root extracts of *Dichrostachys cinerea*

Secondary Metabolites	**Concentration (% per 2g sample)**
Alkaloids	7.35
Saponins	ND
Tannins	13.88
Flavonoids	15.10
Steroids	3.06
Terpenoids	ND
Anthraquinones	ND
Cardiac glycosides	ND

ND = Not detected

Table 2: Aspartate Aminotransferase (AST) and Alanine Aminotransferase (ALT) activities in rats administered hot aqueous root extracts of *Dichrostachys cinerea*

Treatment Duration (Days)	**Groups**	**Liver AST (U/L)**	**Serum AST (U/L)**	**Liver ALT (U/L)**	**Serum ALT (U/L)**
1	A (Distilled Water)	250.90 ± 8.25^{a}	152.11 ± 6.85^{a}	429.74 ± 7.14^{a}	70.00 ± 2.29^{a}
	B (50 mg/kg BW HAREDC)	269.74 ± 3.19^{b}	150.79 ± 5.93^{a}	411.58 ± 1.52^{b}	79.32 ± 1.26^{b}
	C (100 mg/kg BW HAREDC)	272.63 ± 5.02^{b}	162.11 ± 6.58^{b}	361.58 ± 22.63^{c}	97.02 ± 5.78^{c}
	D (200 mg/kg BW HAREDC)	301.84 ± 6.53^{c}	176.32 ± 4.86^{c}	260.79 ± 4.47^{d}	139.74 ± 5.40^{d}
21	A (Distilled Water)	257.11 ± 0.15^{a}	124.74 ± 7.89^{a}	349.74 ± 1.37^{a}	69.08 ± 5.39^{a}
	B (50 mg/kg BW HAREDC)	252.81 ± 5.76^{ac}	122.11 ± 2.63^{a}	344.56 ± 2.28^{a}	65.40 ± 4.32^{a}
	C (100 mg/kg BW HAREDC)	277.37 ± 3.65^{b}	127.54 ± 3.56^{a}	356.05 ± 8.68^{a}	66.84 ± 2.99^{a}
	D (200 mg/kg BW HAREDC)	264.47 ± 2.28^{c}	165.00 ± 6.53^{b}	347.37 ± 8.08^{a}	67.13 ± 8.01^{a}

All values are expressed as mean ± SEM (n=5). Means with different superscripts down the columns are significantly different ($P < 0.05$). HAREDC = Hot Aqueous root extract of *Dichrostachys cinerea.* BW = Body Weight.

Administration of the extract caused significant reduction ($p < 0.05$) in total protein concentration in the treated animals at the highest dose (200 mg/kg BW) on day 1 (Table 3). There was however no significant difference ($p < 0.05$) in the total protein concentration of the animals treated with 50 and 100 mg/kg BW of the extract when compared with control group. At day 21, the extract at all dose levels did not induce any alteration in total protein concentration when compared with control animals. Similarly there

was no significant difference ($p < 0.05$) in albumin concentration of the extract-treated animals at days 1 and 21 when compared with control.

Kidney function indices

Administration of HAREDC at all those levels did not induce any alteration in kidney alkaline phosphatase (ALP) activity at days 1 and 21. Similar result was also recorded in the serum (Table 4). However, there was a significant increase ($p < 0.05$) in kidney gammaglutamyl transferase (GGT) activity in the animals that received the highest dose of HAREDC (i.e. 200 mg/kg BW) at day 1 when compared with the control group (Table 4). At day 21 of HAREDC administration, kidney GGT activity was significantly increased ($p < 0.05$) by all the doses administered. Serum GGT activity was similarly increased significantly ($p < 0.05$) by HAREDC at all the doses at day 1 while only the 200 mg/kg BW of HAREDC significantly increased ($p < 0.05$) serum GGT activity at day 21 (Table 4).

Table 3: Serum total protein and albumin concentrations in rats administered hot aqueous root extracts of *Dichrostachys cinerea*

Treatment Duration (Days)	Groups	Total Protein (g/dl)	Albumin (g/dl)
1	A (Distilled Water)	8.19 ± 0.46^{a}	5.13 ± 0.24^{a}
	B (50 mg/kg BW HAREDC)	7.81 ± 0.29^{a}	4.92 ± 0.86^{a}
	C (100 mg/kg BW HAREDC)	7.67 ± 0.11^{ab}	5.21 ± 0.46^{a}
	D (200 mg/kg BW HAREDC)	7.27 ± 0.59^{b}	5.12 ± 0.38^{a}
21	A (Distilled Water)	8.31 ± 0.13^{a}	4.30 ± 0.61^{a}
	B (50 mg/kg BW HAREDC)	8.96 ± 0.44^{ac}	4.41 ± 0.38^{a}
	C (100 mg/kg BW HAREDC)	8.10 ± 0.47^{a}	4.60 ± 0.56^{a}
	D (200 mg/kg BW HAREDC)	8.19 ± 0.54^{a}	4.78 ± 0.24^{a}

All values are expressed as mean ± SEM (n=5). Means with different superscripts down the columns are significantly different ($P < 0.05$). HAREDC = Hot Aqueous root extract of *Dichrostachys cinerea*. BW = Body Weight

Serum creatinine was significantly decreased ($p < 0.05$) at day 1 in a dose dependent manner following administration of HAREDC, however, at day 21, there was no significant difference ($p > 0.05$) in serum creatinine concentration of HAREDC-treated animals when compared with the control group (Table 5). The 100 and 200 mg/kg BW doses of HAREDC significantly increased ($p < 0.05$) serum urea concentration at day 1 whereas ay day 21, there was no significant difference in serum urea concentration of HAREDC-treated animals when compared with the control group (Table 4).

Serum sodium ion concentration was significantly decreased ($p < 0.05$) by HAREDC administration at all doses and both treatment durations (Table 6). Only the 200 mg/kg BW dose of HAREDC significantly increased ($p < 0.05$) serum potassium ion concentration at day 21 (Table 6). Serum calcium ion concentration was significantly reduced ($p < 0.05$) by all doses and treatment duration of HAREDC while serum phosphate ion concentration was significantly reduced ($p < 0.05$) by all HAREDC doses at day 1 and by 100 and 200 mg/kg BW doses at day 21 (Table 6).

Table 4: Alkaline phosphatase and gammaglutamyl transferase activities in rats administered hot aqueous root extract of *Dichrostachys cinerea*

Treatment duration (Days)	Groups	Kidney ALP (U/L)	Serum ALP (U/L)	Kidney GGT (U/L)	Serum GGT (U/L)
1	A (Distilled water)	72.62 ± 1.36[a]	53.43 ± 0.17[a]	30.85 ± 0.06[a]	1.06 ± 0.12[a]
	B (50 mg/kg BW HAREDC)	73.09 ± 0.43[a]	52.91 ± 0.19[a]	31.27 ± 1.53[a]	2.97 ± 0.12[b]
	C (100 mg/kg BW HAREDC)	72.55 ± 0.16[a]	55.30 ± 0.59[a]	31.91 ± 2.51[a]	3.39 ± 0.12[b]
	D (200 mg/kg BW HAREDC)	69.66 ± 1.11[a]	55.86 ± 0.34[a]	36.47 ± 0.25[b]	2.97 ± 0.25[b]
21	A (Distilled water)	69.93 ± 0.89[a]	51.09 ± 0.06 [a]	22.37 ± 0.43[a]	2.01 ± 0.24[a]
	B (50 mg/kg BW HAREDC)	67.14 ± 1.34 [a]	52.86 ± 0.83 [a]	38.59 ± 2.08[b]	2.48 ± 0.12[ab]
	C (100 mg/kg BW HAREDC)	67.74 ± 1.33 [a]	51.39 ± 0.98 [a]	43.25 ± 2.51[c]	2.41 ± 0.35[ab]
	D (200 mg/kg BW HAREDC)	65.55 ± 1.68 [a]	51.07 ± 0.61 [a]	38.16 ± 1.22[b]	2.76 ± 0.13[b]

All values are expressed as mean ± SEM (n=5). Means with different superscripts down the columns are significantly different (P < 0.05). HAREDC = Hot Aqueous root extract of *Dichrostachys cinerea.* BW = Body Weight

Table 5: Serum creatinine and urea concentrations in rats administered hot aqueous root extract of *Dichrostachys cinerea*

Treatment duration (Days)	Groups	Creatinine (µmol/l)	Urea (mg/dl)
1	A (Distilled water)	58.13 ± 3.06[a]	34.93 ± 2.96[a]
	B (50 mg/kg BW HAREDC)	52.44 ± 3.47[a]	33.39 ± 1.97[a]
	C (100 mg/kg BW HAREDC)	31.47 ± 3.93[b]	43.81 ± 0.59[b]
	D (200 mg/kg BW HAREDC)	23.60 ± 2.27[c]	45.10 ± 1.54[b]
21	A (Distilled water)	59.77 ± 9.83[a]	36.41 ± 0.89[a]
	B (50 mg/kg BW HAREDC)	59.00 ± 3.93[a]	35.37 ± 2.13[a]
	C (100 mg/kg BW HAREDC)	57.03 ± 1.14[a]	39.07 ± 4.70[a]
	D (200 mg/kg BW HAREDC)	60.56 ± 3.47[a]	37.53 ± 0.51[a]

All values are expressed as mean ± SEM (n=5). Means with different superscripts down the columns are significantly different (P < 0.05). HAREDC = Hot Aqueous root extract of *Dichrostachys cinerea.* BW = Body Weight

Table 6: Concentrations of selected serum electrolytes in rats administered with hot aqueous root extract of *Dichrostachys cinerea*

Treatment duration (Days)	Groups	Sodium (mmol/l)	Potassium (mmol/l)	Calcium (mmol/l)	Phosphate (mmol/l)
1	A (Distilled water)	119.82 ± 1.55^{a}	5.55 ± 0.78^{a}	4.13 ± 0.30^{a}	2.95 ± 0.23^{a}
	B (50 mg/kg BW HAREDC)	84.82 ± 1.30^{b}	5.49 ± 0.53^{a}	2.91 ± 0.46^{b}	2.07 ± 0.10^{b}
	C (100 mg/kg BW HAREDC)	88.86 ± 3.67^{bc}	5.14 ± 0.31^{a}	2.99 ± 0.13^{b}	2.16 ± 0.33^{b}
	D (200 mg/kg BW HAREDC)	93.65 ± 2.52^{c}	5.50 ± 0.19^{a}	3.15 ± 0.20^{c}	2.19 ± 0.14^{b}
21	A (Distilled water)	117.90 ± 0.22^{a}	5.41 ± 0.04^{a}	4.45 ± 0.05^{a}	2.85 ± 0.22^{a}
	B (50 mg/kg BW HAREDC)	101.71 ± 2.94^{b}	5.04 ± 0.43^{a}	2.43 ± 0.30^{b}	2.59 ± 0.21^{a}
	C (100 mg/kg BW HAREDC)	96.72 ± 1.08^{b}	5.64 ± 0.18^{a}	3.22 ± 0.53^{c}	1.63 ± 0.24^{b}
	D (200 mg/kg BW HAREDC)	81.36 ± 2.98^{c}	6.33 ± 0.42^{b}	3.95 ± 0.25^{c}	2.05 ± 0.19^{c}

All values are expressed as mean ± SEM (n=5). Means with different superscripts down the columns are significantly different ($P < 0.05$). HAREDC = Hot Aqueous root extract of *Dichrostachys cinerea.* BW = Body Weight

Discussion

Orally administered drug-induced toxicity often manifest as injury to the liver and kidneys, this is because these organs play important roles in the metabolism and excretion of foreign compounds as well as due to their portal location within the circulatory system (Yakubu and Omoniwa, 2012). Biochemical parameters investigated in this study, are routine markers of liver and kidney function. Enzyme pattern in these tissues may be used to assess organ dysfunction and serum enzyme levels may be used to further verify the physiological state of these organs (Yakubu and Omoniwa, 2012). The survival of all the albino rats used in this study following administration of HEREDC at maximum dose of 200 mg/kg body weight implies that the extract is not lethal at this dose, and corroborates findings that have pegged its lethal dose at above 2000 mg/kg (Jayakumari *et al.,* 2011).

The concurrent elevation of AST in the liver and serum recorded in this study suggests damage to liver cell membranes occasioned by HAREDC administration. Derangement of hepatocytes membranes allow leakage of intracellular enzymes into the serum (Yakubu and Omoniwa, 2012). This effect was found to be dose-dependent at day 1 of extract administration. The animals however were able to recover from the assault posed by the lower doses of the extract, since only in animals treated with the 200 mg/kg BW was the anomaly sustained after 21 days. Similar effect was recorded for ALT at day one, however, at day 21, the animals showed total recovery from the initial assault caused by HAREDC at day 1. Alterations in ALT and AST levels will impact on transamination reactions and thus hamper effective amino acid metabolism.

Total protein and albumin are proteins synthesized in the liver and as such are used to assess the state of liver synthetic functions (Kaplan *et al*., 1979; Yakubu and Omoniwa, 2012). Acute exposure to HAREDC in this study had a dampening effect on liver protein synthesis, especially at the higher doses. This effect was however reversed by day 21 of HAREDC treatment. Since albumin was not altered at day 1 of extract administration, it can therefore be inferred that globulin synthesis might have been temporarily suppressed following acute exposure of the animals to HAREDC. Reduced total protein associated with a decrease in globulin level has been linked with nephrotic syndrome, where the kidneys excrete too much

protein from the body. Nephrosis is the most common cause of acute kidney failure and has been reported to occur as a result of animal exposure to nephrotoxins such as those from plants (Constable *et al*., 2017).

In this study, the level of kidney and serum ALP was not altered following HAREDC treatment. The recorded increase in kidney GGT level especially after prolonged treatment indicates HAREDC ability to induce excessive production of kidney GGT beyond what is normally required. It is possible that the extract contains an activator of GGT or an inducer of the GGT synthetic pathway. In addition, since kidney and serum ALP were not altered in this study, it is logical to imply that the recorded increase in serum GGT originated from tissues other than the kidney such as the liver or pancreas. These data may suggest that HAREDC does not compromise the integrity of renal cell membranes since both ALP and GGT are membrane enzymes. Therefore, the slightly elevated level of serum GGT may be explained as likely resulting from injury in other tissues such as liver, bone, heart, intestine, and spleen (Ghadban, 2019; Vroon and Israili, 1990). It also seem to strongly corroborate the fact that acute doses of HAREDC inflicted hepatic injury. Increases in serum GGT levels are associated with all forms hepatobiliary disorders. Moderate elevation (2 to 5 times reference) is often associated with hepatic cell injury due to toxic or infectious hepatitis. Cholestasis due to intrahepatic or extrahepatic biliary obstruction results in higher serum gamma GGT levels (5 to 30 times reference) (Vroon and Israili, 1990). Interestingly, result for serum gamma glutamyl transferase (GGT) assay in this study shows the ratio of GGT activity in the groups that received plant extract to be of 2 – 5 times that of the control group, further pointing to hepatic cell injury due to toxicity and not cholestasis as the cause of hepatic disturbance.

Creatinine is a product of irreversible, nonenzymatic dehydration and dephosphorylation of creatine phosphate in the muscles (Murray *et al*., 2003). One of the excretory functions of the kidneys is to excrete excess creatinine through urine by glomerular filtration. (Murray *et al*., 2003). The dose-dependent reduction in serum levels of creatinine recorded in this study suggests either reduced production of creatine phosphate in the muscles and brain, reduced utilization of creatine phosphate in ATP generation or increased filtration of creatinine by the glomerulus. Since creatinine production from creatine phosphate is a non-enzymatic reaction, then it means that extract might have inhibited of repressed the synthesis of other enzymes catalyzing earlier reactions leading to creatinine production. However, the recorded reduction in serum creatinine levels was transient as animals adjusted to the assault posed by acute exposure to the extract.

The kidneys are responsible for excreting urea which are formed in the liver by deamination of amino acids that are in excess of requirements. Assessment of blood or urine concentration of urea is therefore used to ascertain the integrity of kidneys excretory ability. In this study acute exposure to HAREDC resulted in elevation of serum urea which was however reversed following extract treatment for 21 days. Increased serum levels of urea can be caused by dehydration, excessive protein catabolism, urinary tract obstruction, gastrointestinal bleeding and certain medications (e.g. some antibiotics). It is possible that the extract might elicit a temporary diuretic effect leading to excessive loss of water, which was however not monitored in this study. In addition, tannins present in *Dichrostachys cinerea* root, which possess proven antibiotic activity might induce elevation of blood urea levels (Banso and Adeyemo, 2007).

The recorded reduction in the levels of serum sodium, calcium and phosphate indicates loss of these electrolytes probably through excessive loss of body fluids. This result corroborates the earlier suggestion that HAREDC may possess diuretic or even laxative activities. Electrolytes imbalances are reportedly inducible by drugs that cause excessive loss of body fluids. For example, thiazide diuretics, which are used to treat hypertension and oedema inhibit the sodium-chloride transporter and causes the body to release sodium chloride and water into the lumen, thereby increasing the amount of urine produced each day (Akbari and Khorasani-Zadeh, 2019).

Conclusion

The result of this study showed hot aqueous extract of *Dichrostachys cinerea* root altered liver and kidney function parameters of Wistar rats especially after acute exposure. Research on the toxicity of this extract on other mammalian tissues is highly recommended to confirm the results in this work. The use of aqueous extract of *Dichrostachys cinerea* root in tradomedicine should be with caution.

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