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Short term effect of aqueous extracts of root, pod, and stem of *Telfairia occidentalis* on some biochemical parameters in rats

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KEYWORDS: *Telfairia occidentalis*, inorganic composition, hepatotoxicity, nephrotoxicity.

ABSTRACT: This study investigates the effect of fourteen (14) -day administration of *Telfairia occidentalis* root, pod and stem aqueous extracts on rats. Sixty four (64) Wister albino rats of both sexes were assigned to sixteen (16) groups of 4 animals per group. Different groups received distilled water and, root, stem, and pod extracts at the doses of 250, 750, 1500, 2250, and 3000mg/kg of body weight. All animals were treated for 14 days and sacrificed on the 15th day. The biochemical assay results show that the root extract caused significant decreases in the activities of alanine amino transferses (ALT) and aspartate amino transferase (AST) at lower concentrations (250mg/kg and 750mg/kg), while the stem extract showed significant increase in their activities at 3000mg/kg. The pod extract had no effect on ALT and AST. Also the stem extract showed significant increase in the activities of alkaline phosphatase (ALP) and lactate dehydrogenase (LDH) and reduction in the concentration of serum sodium ion. Serum creatinine (not urea) was elevated when the pod extract was administered at the lowest dose (250mg/kg). Conversely, the stem extract caused a reduction in the concentration of creatinine at 250-, 750- and 1500mg/kg. There was no significant effect on serum total protein and albumin by all the extracts. The stem extract showed a significant increase in the liver- body weight ratio when administered at 750mg/kg and 1500mg/kg dosage. Thus, while *Telfairia occidentalis* root, pod, and stem extracts may have varying protective and toxic effects on liver and kidney function parameters, their effects may be dosage- and duration- dependent.© JASEM

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Telfairia occidentalis (Family: *Curcubitaceae*) is a popular leafy vegetable grown in Nigeria and other West African countries for its edible leaves and seed. Apart from their nutritional importance, the leaves of *Telfairia occidentalis* have been reported for the treatment of convulsion, anaemia, artherosclerosis, cardiovascular disease, hypertension, malaria and impotence (Iwu, 1983**;** Odoemene and Onyeneke, 1988; Sofowora, 1996**;** Obute and Adubor, 2007).

The *T. occidentalis* leaf extract has been demonstrated to possess a dose-dependent hepatoprotective effect (Oboh, 2005). The ethanolic fruit extract has also been shown to have a dose dependent hypercholesterolemic, hyperproteinemic, hypertriglyceridemic and hyper- conjugated bilirubinemic effect on rats, suggesting that the fruit may not be safe for consumption (Olorunfemi *et al.,* 2006). Adaramoye *et al.* (2007) reported a hypolipidemic effect of *Telfairia occidentalis* (Fluted Pumpkin) in rats fed cholesterol-rich diet, which they suggested might be mediated through reduction of oxidative stress and cholesterol levels.

Eseyin *et al*. (2006), investigating the effect of the root ethanol extract of the plant on glucose level of normoglycemic rats observed that, unlike the leaf extract, the root of *T. occidentalis* did not possess hypoglycemic activity. Their work also could not confirm the claim of the toxicity of the root. Also the co- administration of the leaf extract of *T. occidentalis* before or simultaneously with chloroquine affected the pharmacokinetics of the drug (Eseyin *et al.,* 2007).

Telfairia occidentalis leaf extract was also reported to have a regenerative effect on the destroyed testicular

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histology induced by quinine therapy (Nwangwa *et al*., 2007).

Apart from the leaves and the seed of *T. occidentalis* there is little or no known use for the root, stem and pod. The high consumption of the plant in this part of the world has led to the generation of enormous bye products which are usually wasted. The uncertainty about the toxicity or otherwise of the root has led to abandonment of this part of the plant which is usually uprooted during land cultivation or allowed to rot underground. This study tries to ascertain the short term effect of the *T. occidentalis* root, pod and stem extracts on some liver and kidney function parameters.

MATERIALS AND METHODS

Chemicals and Reagents Chemicals and reagents from Sigma-Aldrich Laborchemikalien and BDH Laboratory, Poole England were used in this investigation. Randox (UK) reagent kits were used for estimation of serum alanine aminotransferase (ALT), aspartate aminotransferase (AST), and alkaline phosphatase (ALP).

Collection and Preparation of Plant Materials: Telfairia occidentalis roots stem, and pods were collected from Port Harcourt, Nigeria. The samples were washed, cut into smaller bits and dried under shade. Plant samples were given mild heat treatment prior to blending in mechanical grater mill. The plant samples were ground into fine powder and each sample (200g) was macerated in adequate distilled water for 24 hours to obtain the crude extracts. The extract solutions were filtered and filtrates concentrated and reduced to constant weight at 50^{o}C. The extracts were preserved in a refrigerator until used.

Experimental Design: Sixty-four (64) Wister albino rats were used in this study. Animals were acclimatized for two weeks in the animal house of Biochemistry Department, University of Port Harcourt, with unlimited access to food and water. Animals were divided into 16 groups of 4 animals per group. Two control groups received distilled water alone. Five (5) groups received 250mg/kg body weight, 750mg/kg, 1500mg/kg, 2250mg/kg and 3000mg/kg of the root extract; four (4) groups received received 250mg/kg, 1500mg/kg, 2250mg/kg and 3000mg/kg of the pod extract; while the remaining five (5) groups received 250mg/kg, 750mg/kg, 1500mg/kg, 2250mg/kg and 3000mg/kg of the stem extract. The animals were treated for fourteen (14) days and sacrificed on the 15th day by jugular laceration. Blood was collected into appropriate sample bottles for analysis.

Biochemical analysis: Serum alanine aminotransferase (ALT), aspartate aminotransferase (AST), serum alkaline phosphatase (ALP), and lactate dehydrogenase (LDH) were estimated colorimetrically using Randox reagent enzyme kits based on the methods of Reitman and Frankel (1957), Kind and King (1954) and Rec GSCC (1970) respectively. Total protein assay was based on Biuret method as described by George and O'Neill (2001) and determination of serum albumin, based on bromocresol green (BCG) method as described by Tietz (1990). Estimation of urea was carried out by diacetyl monoxime method (Rosenthal, 1955), creatinine by Jaffe method as described by Wen-Sheng et al. (2012) and serum sodium ion, by ion selective electrode method as described by Chacko et al. (2011).

Statistical analysis: Data obtained were analysed statistically using Analysis of Variance (ANOVA). Post-hoc comparisons were made using the Bonferroni's test. $P<0.05$ was considered statistically significant.

RESULTS AND DISCUSSION

Table 1.0 shows the results of 15-day exposure of experimental animals to *Telfairia occidentalis* root and pod extracts on serum ALT, AST, creatinine and urea. The serum ALT and AST activities were reduced significantly ($P<0.05$) by the root extract at the doses of 250mg/kg body weight and 750mg/kg, while the other dose levels had no effect. The reduction in activities of serum ALT and AST by root extract at the doses of 250mg/kg bw and 750mg/kg bw may be an indication that short term administration of increased dose of the root extract on ALT and AST activities as reported by Ogbonnaya and Uadia (2013) correlates with a lower dose administration over a longer period**.** The creatinine and urea levels were unaffected at all the doses of the root extract administered. The insignificant effect of the root extract on these and other biochemical parameters suggests that the extract may not cause any liver or kidney dysfunction. The variation in responsive doses may be as a result of bioaccumulation and/or antagonism of responsible principles. The root extract also did not affect the organ weights.

The pod extract showed no significant change in ALT, AST, creatinine (except for significant increase

at 250mg/kg bw) and urea. Although insignificant, ALP activity decreased with increase in dose. There were also no significant changes in the other parameters: LDH, total protein, albumin, sodium and animal organ weights. Thus, the extract may not cause any liver or kidney dysfunction within the duration of the administration.

In Table 2.0, the stem extract increased the ALT and AST activities significantly ($P<0.05$) at 3000mg/kg. It also significantly ($P<0.05$) lowered the level of creatinine at dose values of 250mg/kg, 750mg/kg, and 1500mg/kg, compared to normal, while the rest of the doses had no significant effect. Although insignificant, the same trend as creatinine was observed for the effect of stem extract on serum urea.

The results of the effect of 15-day exposure to *Telfairia occidentalis* root and pod extracts on the activities of alkaline phosphatase (ALP) and lactate dehydrogenase (LDH), and serum total protein (TP), albumin, and sodium levels are shown in Table 3.0. The extracts had no significant ($P>0.05$) effect on all the parameters, although the root showed a dose-dependent increase in the activity of ALP, and the pod, a dose-dependent decrease.

Table 4.0 shows the effect of stem extract on serum ALP, LDH, total protein, albumin and sodium. The stem extract at all the doses administered caused a significant ($P<0.05$) increase in the ALP activity compared with control. The increase margin dipped as extract dose increased from 250mg/kg through 1500mg/kg, and rises with the rest of the dose levels. Serum ALP level is related to the function of the hepatic cell and increase in serum level of ALP has been associated with increased synthesis of the enzyme, in presence of increasing biliary pressure (Toda *et al*., 1980). The stem extract also caused a significant ($P<0.05$) increase in LDH activity at dose levels of 750mg/kg through 3000mg/kg. This increase is reduced for the group administered 3000mg/kg stem extract. Lactate dehydrogenase (LDH) is a cytoplasmatic enzyme present in essentially all major organ systems. Its increased presence outside the cell has been associated with damage or cell death resulting from ischaemia, excess heat or cold, starvation, dehydration, injury, exposure to bacterial toxins, ingestion of certain drugs, and from chemical poisonings (Lott and Nemensanszky, 1987; Moss and Henderson, 1986). Thus the significant rise in LDH activity caused by the stem extract at dose levels of 750mg/kg through 3000mg/kg may be associated with its effect on the liver cells. Certain flavonoids such as quercetin, has been reported to prevent lactate dehydrogenase leakage in mouse liver (Molina *et al*., 2003), and this is absent in the stem extract (Ogbonnaya and Uadia, 2013).

Also administration of the stem extract caused a significant reduction in the serum sodium concentration. The significant reduction in the serum sodium concentration by the stem extract may indicate the ability of the extract to prevent or control electrolyte (such as sodium)-induced hypertension. Abnormal concentration of sodium and/or potassium in serum has been noted to affect the osmotic pressure of the body fluid which is related to blood pressure. Sodium attracts water and increase in serum sodium level causes the body to retain water. Thus, as serum level of sodium can affect blood pressure (Cheesbrough, 2002), the stem extract may be used in the control of electrolyte (sodium)-induced hypertension. This may explain the ethno-medicinal use of *Telfairia occidentalis* in the treatment of hypertension (Weiner, 1992).

The exposure of *Telfairia occidentalis* root and pod extracts to experimental animals had no significant ($P>0.05$) effect on the organ: body weight ratios (Table 5.0). However, the stem extract caused a significant increase in the liver: body weight ratio when administered at doses of 750mg/kg and 1500mg/kg (Table 6.0). Moore and Dalley (1999) reported that increase in the liver-body weight ratio was an indication of swollen organ, atrophy or hypertrophy. Increase in liver size/weight has been associated with hepatocellular hypertrophy, necrotic foci, vacuolization, and increased eosinophilia (National Academy of Sciences, 1994). Hepatocyte vacuolation may result from plasma influx into the cytoplasm (Li *et al*., 2003). Thus, the increase in the liver-body weight ratio caused by the stem extract at 750- and 1500mg/kg bw may also be associated with the its effect on electrolyte redistribution leading to hepatocyte vacuolation.

Thus, while *Telfairia occidentalis* root, pod, and stem extracts may have varying protective and toxic effects on liver and kidney function parameters, these effects may be dosage- and duration- dependent.

Table 1: Result of Fifteen (15) - Day Exposure to *Telfairia occidentalis* Root and Pod Extracts

Group	Treatment	ALT(U/L) Mean± SEM	AST(U/L) Mean± SEM	Creatinine (µmol/L) Mean± SEM	Urea (mmol/L) Mean± SEM
1	Distilled Water	28.33 ± 2.33	101.50 ± 0.5	17.00± 1.00	3.00± 0.46
2	Root Extract(250mg/kg)	22.75± 2.14	74.00± 3.4 [a]	18.75± 1.25	4.35± 0.54
3	Root Extract(750mg/kg	22.00± 0.82	77.03±3.02 [a]	17.67± 0.88	4.33± 0.48
4	Root Extract(1500mg/kg)	30.00±1.47	93.63± 8.0	19.25± 0.95	4.58± 0.15
5	Root Extract2250mg/kg)	28.00± 1.00	96.60± 3.9	19.00± 1.73	4.43± 0.12
6	Root Extract(3000mg/kg)	30.75± 2.46	93.10± 1.35	22.25± 0.63	4.50± 0.26
7	Pod Extract(250mg/kg)	28.00± 4.00	96.87± 5.77	25.33± 2.33[a]	4.70± 0.30
8	Pod Extract(1500mg/kg)	32.00± 3.5	107.85± 8.35	18.67± 1.76	3.97± 0.91
9	Pod Extract(2250mg/kg)	30.00±1.00	96.17±4.82	23.67± 1.45	3.60± 0.53
10	Pod Extract(3000mg/kg)	25.33± 1.67	101.00± 0.00	19.67± 1.86	4.10± 0.15

n=4; [a]Significant difference (P< 0.05) compared with Group 1.

Table 2: Result of Fifteen (15) - Day Exposure *to Telfairia occidentalis* Stem Extract

Group	Treatment	ALT(U/L) Mean± SEM	AST(U/L) Mean± SEM	Creatinine (µmol/L) Mean± SEM	Urea (mmol/L) Mean± SEM
1	Distilled Water	24.00± 0.58	75.50 ± 3.50	77.50 ± 3.23	5.00± 1.07
2	Stem Extract (250mg/kg)	18.75± 1.11	90.00± 4.52	60.00± 2.04 [a]	4.98 ± 0.21
3	Stem Extract (750mg/kg)	18.00 ± 2.55	91.50 ± 4.57	52.50 ± 1.44 [a]	4.48 ± 0.23
4	Stem Extract (1500mg/kg)	30.5 ± 1.32	94.00± 2.65	50.00 ± 2.04 [a]	3.63 ± 0.15
5	Stem Extract (2250mg/kg)	31.75 ± 0.75	99.50 ± 0.50	67.50 ± 3.23	5.25 ± 0.16
6	Stem Extract (3000mg/kg)	36.00 ± 1.73 [a]	110.25 ± 2.69 [a]	71.25 ± 2.39	5.05 ± 0.12

n=4; [a]Significant difference (P< 0.05) compared with Group 1.

Table 3: Result of Fifteen (15) - Day Exposure to *Telfairia occidentalis* Root and Pod Extracts

Group	Treatment	ALP(U/L) Mean± SEM	LDH(U/L) Mean± SEM	Total Protein(g/l) Mean± SEM	Albumin(g/l) Mean± SEM	Sodium(mmol/L) Mean± SEM
1	Distilled Water alone	616.7 ±28.4	1771.0±24.0	63.7±0.9	28.3±1.5	139.0±2.5
2	Root Extract (250mg/kg)	649.3± 51.2	1620.0±241.0	70.0±1.5	29.3±0.9	139.0±0.4
3	Root Extract (750mg/kg	654.3± 48.2	1437.0±95.3	65.3±2.0	27.5±0.6	140.3±0.8
4	Root Extract (1500mg/kg)	701.3± 44.7	1235.0±85.5	70.5±1.8	29.0±0.7	141.5±0.3
5	Root Extract 2250mg/kg)	700.3±60.6	1985.0±37.5	70.3±3.0	30.0±1.2	139.3±2.2
6	Root Extract (3000mg/kg)	747.0± 72.7	1255.0±152.3	69.0±2.3	30.0±1.5	141.8±1.1
7	Pod Extract (250mg/kg)	606. ± 28.5	1441.0±202.8	66.8±1.8	28.3±0.8	140.8±0.6
8	Pod Extract (1500mg/kg)	554.5±32.5	2102.0±53.0	66.0±1.0	27.3±1.7	138.0±0.6
9	Pod Extract (2250mg/kg)	495.3±2.2	1492.7±197.8	68.8±0.5	28.8±0.9	139.0±1.5
10	Pod Extract (3000mg/kg)	437.0±32.0	1915.5±48.5	68.7±2.7	28.7±0.3	137.3±0.7

n=4

Table 4: Result of Fifteen (15)-Day Exposure to *Telfairia occidentalis* Stem Extract

Group	Treatment	ALP(U/L) Mean± SEM	LDH (U/L) Mean± SEM	Total Protein(g/l) Mean± SEM	Albumin(g/l) Mean± SEM	Sodium (mmol/L) Mean± SEM
1	Distilled Water	199.7 ±3.2	1771.0±24.0	65.0±2.3	29.5 ± 3.1	153.5±1.2
2	Stem Extract (250mg/kg)	383.3 ± 5.2[a]	1567.3 ± 73.8	59.3 ± 4.4	38.0 ± 1.5	142.0± 0.9 [a]
3	Stem Extract (750mg/kg)	287.7 ± 9.4 [a]	3451.0 ± 77.2 [a]	68.0 ± 2.1	38.8 ± 2.4	142.8 ± 0.8 [a]
4	Stem Extract (1500mg/kg)	250.7 ± 5.2 [a]	3532.0 ± 64.1 [a]	53.8 ± 6.3	38.0 ± 1.6	139.3 ± 0.6 [a]
5	Stem Extract (2250mg/kg)	442.7 ± 3.7 [a]	3766.8 ± 56.5 [a]	69.5 ± 1.0	39.5 ± 2.9	138.8 ± 0.9 [a]
6	Stem Extract (3000mg/kg)	476.7 ± 12.0 [a]	2565.0 ± 75.2 [a]	68.0 ± 4.4	39.5 ± 1.7	144.5 ± 1.0 [a]

n=4; [a]Significant difference (P< 0.05) compared with Group 1.

Table 5: Tissue Weight Result of Fifteen (15)- Day Exposure to *Telfairia occidentalis* Root and Pod Extracts

Group	Treatment	Tissue Weight (g/100g Body Weight)		
		Kidney Mean± SEM	Liver Mean± SEM	Heart Mean± SEM
1	Distilled Water	0.31± 0.02	2.76 ± 0.28	0.36 ± 0.01
2	Root Extract (250mg/kg)	0.29 ± 0.03	2.82 ± 0.40	0.35 ± 0.01
3	Root Extract (750mg/kg	0.33 ± 0.02	3.30 ± 0.21	0.34 ± 0.04
4	Root Extract (1500mg/kg)	0.31 ± 0.02	3.12 ± 0.27	0.37 ± 0.01
5	Root Extract 2250mg/kg)	0.29 ± 0.03	3.00 ± 0.75	0.36 ± 0.01
6	Root Extract (3000mg/kg)	0.32 ± 0.01	2.90 ± 0.17	0.35 ± 0.01
7	Pod Extract (250mg/kg)	0.32 ± 0.02	3.43 ± 0.15	0.35 ± 0.01
8	Pod Extract (1500mg/kg)	0.35± 0.01	3.13 ± 0.26	0.39 ± 0.05
9	Pod Extract (2250mg/kg)	0.29 ± 0.02	2.77 ± 0.19	0.28 ± 0.01
10	Pod Extract (3000mg/kg)	0.29± 0.03	2.96 ± 0.06	0.33 ± 0.01

n=4

Table 6: Tissue Weight Ratio Result of Fifteen (15)-Day Exposure to *Telfairia occidentalis* Stem Extract

Group	Treatment	Tissue Weight (g/100g Body Weight)		
		Kidney Mean± SEM	Liver Mean± SEM	Heart Mean± SEM
1	Distilled Water	0.42 ± 0.03	2.73± 0.25	0.39 ± 0.02
2	Stem Extract (250mg/kg)	0.38 ± 0.02	3.27 ± 0.21	0.33 ± 0.00
3	Stem Extract (750mg/kg)	0.45 ± 0.02	3.86 ± 0.12[a]	0.39 ± 0.02
4	Stem Extract (1500mg/kg)	0.47 ± 0.04	3.97 ± 0.29[a]	0.41 ± 0.04
5	Stem Extract (2250mg/kg)	0.42 ± 0.02	3.51 ± 0.13	0.36 ± 0.01
6	Stem Extract (3000mg/kg)	0.37 ± 0.01	3.34 ± 0.29	0.33 ± 0.01

n=4; [a]Significant difference (P< 0.05) compared with Group 1.

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