## RIGINAL ARTICLE

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# Determination of the inorganic composition and short term effect of aqueous extracts of root, pod, and stem of *Telfairia occidentalis* on some hematological parameters in rats

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# ABSTRACT

Aims and Objectives: This study is to investigate the inorganic composition and the effect of fourteen (14) -day oral administration of aqueous extracts of root, pod and stem of Telfairia occidentalis in rats. Materials and Methods: Plant samples were analyzed for K, P, Mg, Ca, S, Mn, Fe, Cu, Zn, Pb, and Crusing atomic absorption spectrophotometry (AAS). Wister rats of both sexes were assigned to sixteen (16) groups of 4 animals per group. Different animal 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. Results: The inorganic composition result showed that potassium (K) was the highest in abundance in all the samples (root, pod, and stem), compared to the other mineral elements. The root contained high levels of potassium and manganese; the stem had magnesium, manganese and iron while the pod contained high amount of potassium, manganese and iron. Hematological assay results showed reduction in the values of white blood cell (WBC) count for higher doses of root extract and all doses (except for 1500 mg/kg bw) of pod extract. Platelet count (PLT) was significantly high for root extract at 750 mg/kg bw. The stem extract showed a consistent decrease in the red blood cell (RBC) count for all dose administered. Conclusion: Telfairia occidentalis root, pod, and stem may be good sources of inorganic elements. The extracts have varying effects on hematological parameters which may be dosage- and duration- dependent.

**Key words:** *Telfairia occidentalis*, Inorganic composition, Hematological parameters, RDA, Root, Pod, Stem

# INTRODUCTION

In a world of close to 600 million people below \$1.25 a day and increasing hunger and sickness, there is the need for alternative and cheaper sources of food and medicine.<sup>1,2</sup> The focus on non-conventional sources for food and medicine will enable the utilization of, hitherto, waste materials, in the generation of products that are of both nutritional and medicinal importance. Plant materials are composed of mineral elements and chemicals which can be harnessed for animal and human nutrition, and as remedy for ailments. The mineral elements in plants comprise of nitrogen, phosphorus, potassium, sodium, magnesium, calcium, manganese, iron, copper, zinc, lead and chromium. These are required in macro- or micro- level for cell metabolism. Their deficiency in the body may result in one ailment or the other.

*Telfairia occidentalis* belongs to the family Curcubitaceae. It is a food crop grown in some West African countries including Nigeria and used as vegetable. In ethno-medicine, the leaves are useful in the treatment of convulsion, anemia, diabetes, artherosclerosis, cardiovascular disease, hypertension, malaria and impotence.<sup>3-9</sup> The aqueous root,

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stem and pod extracts, on short term investigation has been reported to have dose dependent effects on the liver and the kidney.10

There have been many investigations into the leaves and seed part of the plant and very few on the root, stem and pod. There is also uncertainty about the toxicity or otherwise of the root extract which has contributed to absolutely no nutritional use for this part of the plant although it may contain considerable level of nutrients and phytomedicines.11

In this study we investigated the quantitative elemental composition of the root, stem and pod of Telfairia occidentalis and also the effect of their extracts on some hematological parameters in rats.

# **MATERIALS AND METHODS**

## **Plant materials**

Telfairia occidentalis root stem and pod were collected from a farmland in obigbo, Port Harcourt, Nigeria. Samples were identified at the Plant Science and Biotechnology Department of University of Port Harcourt, Port Harcourt, Nigeria. The samples were washed, cut into bits and dried under shade.

#### Animals

Sixty four (64) Wistar albino rats, weighing (75-175g) were obtained from the animal house of Physiology Department, University of Nigeria, Enugu Campus. The animals were kept in the animal house of Biochemistry Dept, University of Port Harcourt and acclimatized for two (2) weeks with unlimited access to water and normal rat chow.

#### Extraction of plant

Plant samples were given mild heat treatment before blending in mechanical grater mill. The plant samples were ground into fine powder, and part of the powdered samples kept for elemental composition determination. Each sample (200 g) was macerated in adequate aqueous solvent for 24 hours to obtain crude extracts. The extract solutions were filtered and filtrates concentrated and reduced to constant weight at 30°C. The extracts were stored in the refrigerator until used.

#### Inorganic composition analysis

This was carried out using Buck Scientific 205 Atomic Absorption/Emission Spectrophotometer.

The elements such as K, Mg, Ca, Mn, Fe, Cu, Zn, Pb, and Cr were analysed using atomic absorption spectrophotometry (AAS). Phosphorus and sulphur were

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analysed as phosphate and sulphate using a modified method.12

Contribution to recommended dietary allowance (RDA) This was calculated according to NRC.13

$$RDA (\%) = \frac{Concentration of the elements}{RDA} \times 100$$

Where RDA is recommended dietary allowance.13

## **Animal treatments**

Wistar albino rats of both sexes were assigned to 16 groups of 4 animals per group. Different groups received distilled water (2 groups), root-, stem-, and pod extracts at doses of 250, 750, 1500, 2250, and 3000 mg/kg of body weight. All animals were treated for 14 days and sacrificed on the 15<sup>th</sup> day.

Animals were placed under mild chloroform anesthesia, and blood collected by jugular laceration. Blood was collected into sample bottles containing potassium salt of ethylene diamine tetra acetic acid (EDTA) for analysis.

#### Hematological analysis

Packed Cell volume (PCV) was determined using microhaematocrit reader; hemoglobin by LED modified method; red blood cell (RBC) count was determined by formal citrate, formaldehyde and counting chamber method; white blood cell (WBC) count, using Turks solution and improved Neubauer counting chamber; and platelet by method of 1% ammonium oxalate solution and counting chamber.14-16

#### **Statistical analysis**

The data 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

The elemental composition analysis showed that potassium (K) is the most abundance in all the Telfairia occidentalis samples (root, pod, and stem), compared to other inorganic components. This was followed by magnesium and calcium. Also potassium is concentrated more in the root and fruit pod, than in the stem. The Telfairia occidentalis root has a relatively high abundance of sodium, calcium and zinc. The pod has the highest concentrations of potassium, iron, zinc and lead. The stem has relatively high iron content, while chromium was absent in all the samples (Table 1).

Table 2 shows the mineral elements' contribution of Telfairia occidentalis root, stem and pod to recommended

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dietary allowance (RDA). The root and the pod showed higher contribution of potassium compared with stem. The stem showed the highest percentage contribution of magnesium. While manganese was highly contributed by root, stem and pod, the stem and the pod contributed more iron than the root.

The hematological assay result (Table 3) shows that the root and pod extracts had no significant effect on packed cell volume (PCV), hemoglobin count (Hb) and red blood cell count (RBC) for all the groups that received the extracts. A significant increase in platelet (PLT) level was observed for root at 750mg/kg bw. There were significant reduction in the white blood cell count (WBC) for root extract (2250- and 3000mg/kg bw) and all the pod extract groups (except 1500mg/kg bw group).

The stem extract caused no significant change in the PCV, Hb, WBC, and PLT irrespective of dose administered (Table 4). However, it showed a significant reduction in the RBC value for all the doses compared to control.

## DISCUSSION

The mineral composition and contribution to dietary intake indicated that the root and pod are good sources of potassium and manganese, while the stem is rich in magnesium, manganese and iron. The pod may also be a good source of iron. Potassium is known to be the most abundant cation in plant cells and plays essential roles in maintaining the membrane potential and ion homeostasis, and in enzyme activation, signal transduction, and many other physiological processes.<sup>18</sup>

Potassium was reported to lower blood pressure and reduces salt sensitivity.<sup>19,20</sup> Research also suggested its effect on the structure and mechanical function of the heart, which can lead to improvements in many cardiovascular risk factors.<sup>21</sup> Similar to *Telfairia occidentalis* leaf extract which was reported to be rich in iron, potassium and magnesium among others, and used in the treatment of anemia, high blood pressure and cardiovascular disorders,<sup>8,22</sup> *Telfairia occidentalis* root, stem and pod extract may possess similar antianemic and antihypertensive properties.

Magnesium and manganese are involved in enzyme activation leading to energy production and protein metabolism.<sup>23,24</sup> Manganese also plays a cofactor role in some antioxidative enzymes and is usually taken up by liver and kidney.<sup>25-28</sup> High dose of manganese cause oxidative injury and low dose have antioxidant effect.<sup>29,30</sup> Manganese is of importance to avian metabolism as it is

Elements	Root	Stem	Pod
Phosphorus (mg/100 g)	5.58±0.52	1.11±0.01	3.97±0.93
Potassium (mg/100 g)	1653.00±23.00	880.00±20.00	1995.50±25.50
Sulphur (mg/100 g)	15.00±1.00	30.00±5.00	24.00±3.00
Sodium (mg/100 g)	53.63±3.53	4.11±0.11	12.43±1.44
Chlorine (mg/100 g)	0.28±0.01	0.07±0.01	0.60±0.11
Magnesium (mg/100 g)	118.00±9.00	486.50±16.50	155.00±15.00
Calcium (mg/100 g)	293.50±6.50	120.00±8.00	46.80±4.00
Manganese (mg/100 g)	4.11±0.11	6.75±0.45	3.47±0.47
Iron (mg/100 g)	<0.10	7.84±0.35	8.15±1.00
Copper (mg/100 g)	0.00±0.00	0.06±0.00	0.03±0.00
Zinc (mg/100 g)	0.82±0.11	2.09±0.09	1.016±0.02
Lead (mg/100 g)	0.34±0.04	0.40±0.10	0.47±0.02
Chromium (mg/100 g)	0.00±0.00	0.00±0.00	0.00±0.00

Values represented as mean±standard error of mean (SEM) of duplicate determination

Table 2: Contributions of mineral elements of 7	<i>Felfairia occidentalis</i> root, stem and pe	od to RDA
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Elements	RDA* (Mg)	Root contribution to RDA (%)	Stem contribution to RDA (%)	Pod contribution to RDA (%)
Phosphorus (mg/100 g)	1200	0	0	0
Potassium (mg/100 g)	2000	83	44	100
Sodium (mg/100 g)	500	11	1	2
Magnesium (mg/100 g)	350	34	139	44
Calcium (mg/100 g)	1200	24	10	4
Manganese (mg/100 g)	2-5	82-206	135-338	69-174
Iron (mg/100 g)	10-15	1	52-78	54-82
Copper (mg/100 g)	1.5-3	0	2-4	1-2
Zinc (mg/100 g)	12-19	4-7	11-17	5-8

Group	Treatment	Mean±SEM					
		Pcv (%)	Hb (g/dl)	RBC (×10 <sup>9</sup> )	WBC (×10 <sup>9</sup> )	PLT (×10°)	
1	Distilled water	30.7±1.8	10.4±0.6	3.8±0.2	6.9±0.7	343.7±48.2	
2	Root extract (250 mg/kg bw)	35.3±0.9	11.9±0.4	4.3±0.1	5.7±0.3	317.3±40.2	
3	Root extract (750 mg/kg bw	39.0±2.0	12.4±0.0	4.5±0.00	5.0±0.2	652.0±18.0ª	
4	Root extract (1500 mg/kg bw)	33.0±3.0	11.5±1.5	3.7±0.2	6.7±0.4	291.5±8.5	
5	Root extract (2250 mg/kg bw)	29.5±1.5	12.0±2.0	3.6±0.1	1.8±0.4ª	389.0±16.0	
6	Root extract (3000 mg/kg bw)	32.0±3.0	12.5±2.5	3.1±0.3	3.1±0.3ª	292.0±9.0	
7	Pod extract (250 mg/kg bw)	33.0±2.0	11.0±1.1	3.6±0.2	1.7±0.2ª	409.0±11.0	
8	Pod extract (1500 mg/kg bw)	33.0±1.0	11.3±0.3	3.8±0.1	5.8±0.6	307.0±53.0	
9	Pod extract (2250 mg/kg bw)	31.5±2.5	11.5±0.5	4.1±0.3	3.0±1.6ª	353.0±52.0	
10	Pod extract (3000 mg/kg bw)	30.0±0.0	10.6±0.5	3.6±0.1	2.2±0.4ª	323.0±19.0	

n=4; °Significant difference (P<0.05) compared to Group 1

Table 4: Effect of Telfairia occidentalis stem extract on hematological parameters

Group	Treatment					
		Pcv (%)	Hb (g/dl)	RBC (×10 <sup>9</sup> )	WBC (×10 <sup>9</sup> )	PLT (×10 <sup>9</sup> )
1	Distilled water alone	47.0±1.0	15.4±0.5	10.8±0.5	9.5±2.1	716.5±14.5
2	Stem extract (250 mg/kg bw)	43.7±0.9	14.5±0.2	8.5±0.0ª	8.3±1.3	809.5±2.5
3	Stem extract (750 mg/kg bw)	42.7±0.7	14.5±0.5	8.2±0.3ª	8.7±0.6	759.0±11.5
4	Stem extract (1500 mg/kg bw)	44.0±0.0	14.5±0.2	7.6±0.5ª	6.5±1.0	682.7±32.9
5	Stem extract (2250 mg/kg bw)	44.8±0.5	14.5±0.2	8.6±0.2ª	10.1±1.7	748.0±14.2
6	Stem extract (3000 mg/kg bw)	44.8±1.0	14.3±0.2	8.4±0.2ª	9.6±1.4	826.7±20.5

n=4; "Significant difference (P<0.05) compared to Group 1

involved in the synthesis of choline and cholesterol. It has been reported to have hypoglycemic effect.<sup>31,32</sup> It was also reported that magnesium may substitute for manganese as enzyme cofactor without compromising enzyme activity.<sup>31</sup> Magnesium has been noted to affect glucose, calcium and iron homeostasis; stimulate vasodilatation and lower blood pressure.<sup>33,34</sup> These elements may also contribute to antihypertensive and hypoglycemic tendencies of the extracts.

Iron has been associated with hemoglobin synthesis and erythrocyte production and plants are known to be good sources of non-heme iron.<sup>35</sup> It has been noted that absorption of non-heme iron was influenced by iron status; dietary factors such as phytate, tannins and excess of some mineral elements such as calcium, phosphorus, manganese, zinc and copper.<sup>36</sup> Thus the level of manganese in the plant extracts may have some influence in the iron homeostasis as indicated by the RBC count results.

The root extract caused no significant change in the hematological parameters except for the WBC. Reduction in the level of WBC has been attributed to immunosuppressant activity and may also result from reduced production and redistribution from peripheral blood into tissues or rapid destruction of WBC.<sup>37,38</sup> A greater proportion of WBC is composed of lymphocytes and reduction in T-lymphocyte number, blastogenesis and mitogenesis have been reported for iron deficiency.<sup>39</sup> Increase in circulating platelets (thrombocytosis) has been attributed to initial response to toxic effect and a decrease resulting from decreased haematopoiesis.<sup>40,41</sup>

Except for WBC which was significantly (P<0.05) reduced: also indicating immunosuppressant activity, the pod extract had no affect on the peripheral blood.<sup>37</sup> The expected increase in the level of RBC due to significant concentration of elemental iron may have been reversed due to the high level of manganese in the pod. Manganese has been suggested to compete with iron for binding site in the specific iron-binding  $\beta_1$ -globulin, transferrin, in the plasma. Also, the degree of saturation of transferrin affects iron deposition in the liver and its release to red blood cell precursors.<sup>42</sup>

The stem extract caused a significant (P<0.05) reduction in the RBC value for all the doses, compared to control. Iron is a principal component of RBC and was found to have 52-78% contribution to RDA (Table 2). The reduction in RBC may be related to the presence of some factors that influence absorption of non-heme (plant) iron. These include dietary factors such as glycosides-especially cyanogenic glycosides, phytate, tannins and manganese.<sup>36,43</sup> But with the levels of PCV and Hb unaffected, the reduction in RBC may also arise from other sources including increased water intake, hemolysis or suppression of erythropoietin production.<sup>44,45</sup> The reduction of WBC by extract is suggestive of the effect of some of the constituents on WBC circulation or formation. Thus the aqueous extract of root, pod and stem of *Telfairia* occidentalis are rich sources of potassium, magnesium, manganese and iron. The antihypertensive property ascribed to *Telfairia occidentalis* may be contributed by the potassium and magnesium content of the plant. The availability of the elemental iron may be dependent on other factors that influence iron metabolism, including antinutritional factors and competing elements. Their effects on WBC may be attributed to their antimicrobial or immunosuppressive effects.

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#### Authors Contribution:

EAO - Initiated, designed and carried out the research, and compiled manuscript; POU - Supervised research design and experimentation, revised manuscript.

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