

Haematinic Effect of Raw and Boiled Leaf Juice of *Cnidoscolus aconitifolius* using Cyclophosphamide-Treated Adult Male Albino Rats

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Abstract

This study evaluated the haematinic effect of raw and boiled leaf juice of *Cnidoscolus aconitifolius* using cyclophosphamide treated adult male albino rats. Fresh leaves of *Cnidoscolus aconitifolius* were harvested from Aku, Enugu state, Nigeria. The leaves were used to prepare fresh and boiled leaf juices which were subjected to chemical analysis using standard analytical methods. Thirty-two adult male albino rats weighing 150 - 250g were allotted into 8 groups of 4 rats each. Seven groups were induced with anaemia. Six groups out of the seven anaemia-induced groups received 5 ml, 10 ml and 15 ml per kg body weight of the raw leaf juice and 5 ml, 10 ml and 15 ml per kg body weight of the boiled leaf juice. The remaining anaemia-induced group was the negative control group. The eighth group was the normal control group. Blood samples were obtained from the rats through the retro-orbital venous sinus for assessment of haematological parameters after acclimatization, after anaemia induction and at the end of the study. Statistical Product for Service Solution (SPSS) for windows version 17 was used to analyse data obtained. $P < 0.05$ was accepted as a cut-off for significant level. Group 6 and 8 (experimental group) had the highest PCV ($45.00\% \pm 2.58$) after treatment while group 7 (negative control group) had the least PCV ($22.50\% \pm 3.42$) level after treatment. Group 7 (negative control group) had the lowest RBC (26.75 ± 6.99) after treatment while group 6 (experimental group) had the highest RBC (260.00 ± 18.26) count after treatment. Significant ($p < 0.05$) increases in haemoglobin, packed cell volume, red blood cell and white blood cell of the anaemic rats treated with raw and boiled *Cnidoscolus aconitifolius* leaf juices were observed. This reveals that the raw and boiled *Cnidoscolus aconitifolius* leaf juices possessed haematinic property.

Keywords: *Cnidoscolus aconitifolius*; Haemoglobin; Packed Cell Volume; Red Blood Cell; White Blood Cell

Introduction

Anaemia, defined as a condition in which the number of red blood cells or their oxygen-carrying capacity is insufficient to meet physiologic needs, develops when the rate of bone marrow red cell production fails to keep pace with destruction or loss of cells [1]. It can result in impaired cognitive development, reduced physical work capacity and in severe cases, increased risk of mortality [2]. The incidence of anaemia is higher in third world than in developing countries due to the presence of many aggravating factors such as poor nutrition, high prevalence of blood parasites eg. plasmodium, trypanosomes and helminthes infestation [3]. Rural populations in various parts of the world do not have access to high quality drugs for the treatment of anaemia, so they depend heavily on plants and herbal products for the treatment of diseases and anaemia [3]. According to Akah, Okolo and Ezike [4], a good number of medicinal plants are traditionally employed to alleviate anaemia. Some of these plants include *Brillantasia nitens* [4], *Telferia occidentalis* [5], *Hibiscus sabdariffa* [6] and *Hygrophila spinosa* [7].

In Nigeria, the rural dwellers are known to use natural herbs and their extracts to treat various blood deficiencies. One of such plant is *Cnidoscolus aconitifolius* which belongs to the *Euphorbiaceae* family and is commonly known as tree spinach (English), *efoyanaipaja*

or *efo* Jerusalem (Yoruba). *Cnidoscolus aconitifolius* is an ornamental evergreen, drought deciduous shrub of 3 - 5m tall [8]. It is a widely distributed annual plant ranging temperate to tropical zones and has a long history of use as both a medicinal and an edible plant [9]. There are claims by people in Aku, Igbo-Etiti LGA of Enugu state, Nigeria that the raw leaf juice of *Cnidoscolus aconitifolius* boosts blood when consumed. This study was therefore undertaken to evaluate the potential haematinic effect of raw and boiled leaf juice of *Cnidoscolus aconitifolius* using cyclophosphamide-treated adult male albino rats.

Materials and Methods

Collection of plant samples

Fresh leaves of tree spinach (*Cnidoscolus aconitifolius*) were collected from a residential home garden in Eziosigo` Aku, Igbo-Etiti L.G.A, Enugu state, Nigeria. It was botanically identified at the Department of Plant Science and Biotechnology, University of Nigeria, Nsukka.

Preparation of plant juice

Four hundred gram (400g) of tree spinach (*Cnidoscolus aconitifolius*) leaves were washed under tap water, placed in a colander for 10minutes to drain out water. The leaves were squeezed thoroughly and severally in a large bowl in order to produce juice from the leaves. Four hundred millilitres (400ml) of water was added to the leaves. The mixture was stirred and the leaf particles were removed. A muslin cloth was used to sieve *Cnidoscolus aconitifolius* leaf juice so as to remove the tiny leaf particles in the juice. The residue was discarded and the juice obtained. The leaf juice was divided into two (A and B). Juice A was boiled for 5 minutes and allowed to cool prior to use while juice B was used in its raw form.

Proximate analysis: Duplicate samples of the raw and boiled leaf juices were analysed for moisture, ash, crude fibre, crude fat and crude protein using the standard method of AOAC [10]. Carbohydrate was obtained by difference.

Micronutrient determination: Duplicate samples of the raw and boiled leaf juices were analysed for micronutrients. Pro-vitamin A determination was done using the procedure of Jakutowicz [11]. Ascorbic acid (Vitamin C) determination was done using the DCIP titrimetric method of AOAC [10] for ascorbic acid determination. Riboflavin (vitamin B₂) determination was done using the method of Koche [12]. Vitamin B₆ was determined by the modified method of Stroebecker and Henning [13]. The method described by the Association of Vitamin Chemists [14] was used for folate determination. Vitamin B₁₂ (cyanocobalamine) determination was done using the colorimetric method described by Karmi, Zayed, Baragethi, Qadi and Ghanem [15]. Iron determination was done using the standard phenanthroline method of AOAC [10]. The dithizone method described by AOAC [10] was used for zinc determination. Determination of copper was done using the Neocuproinine method as described by AOAC [11].

Rat study

Animal source:

Thirty two (32) adult male albino rats weighing between 150 - 250g were purchased from the Faculty of Veterinary Medicine, Department of Veterinary Pathology, University of Nigeria, Nsukka, Enugu state, Nigeria.

Housing and grouping of rats

The rats were housed in the metabolic cages of the laboratory animal facility of the Department of Home Science, Nutrition and Dietetics, Faculty of Agriculture, University of Nigeria, Nsukka. The rats were weighed and allotted into eight (8) groups of four rats each such that the difference in weight of the rats in each group were not more than five grams (5g).

Induction of anaemia

Anaemia was induced by administering cyclophosphamide (CPX) interperitoneally at a dose of 50 mg/kgBW [17]. This was monitored every other day until the level of hemoglobin (Hb), packed cell volume (PCV), and red blood cell (RBC) went below normal on the fifth day. Anaemia was induced in all the groups except the normal control group (Group 8).

Feeding of the animals

The rats were fed rat chow and water *ad-libitum* during the 5-day acclimatization period and throughout the study period. After acclimatization, groups 1-6 were induced with anaemia and were given the leaf juices orally using orogastric tubes and syringes in varying levels according to their groups and body weight. Groups 1 - 3 received 5 ml/kgBW, 10 ml/kgBW and 15 ml/kgBW of the raw leaf juice of *Cnidoscopus aconitifolius*, respectively, while groups 4 - 6 received 5 ml/kgBW, 10 ml/kgBW and 15 ml/kgBW of the boiled leaf juice of *Cnidoscopus aconitifolius*. Group 7 (negative control group) and group 8 (normal control group) received rat chow and water *ad-libitum* throughout the study period. The rat study lasted for 24 days (5 days of acclimatization, 5 days for anaemia induction and 14 days for the experimental study).

Haematological parameter determination: Five millilitres of blood was obtained after acclimatization, after anaemia induction and at the end of the study from the rats through the retro-orbital venous sinus into heparinized bottles for haematological parameter determination. Packed cell volume (PCV) was determined by the microhaematocrit method, haemoglobin (Hb) concentration was done spectrophotometrically cyanomethaemoglobin method, red blood cells(RBCs) count determination was done using the Thoma (manual counting) method, white blood cells (WBCs) count determination was done using manual WBC counting method. MCV was calculated from the RBCs count and PCV using the following formula:

$$MCV = \frac{PCV}{RBC} \times 10 \text{ as described by [18]}$$

MCHC was calculated from the haemoglobin and PCV using the following formula:

$$MCHC = \frac{Haemoglobin}{PCV} \times 100 \text{ as described by [19]}$$

Statistical analysis

Data obtained was analysed using the computer program Statistical Product for Service Solution(SPSS) for windows version 17. Main analysis included means and standard deviation. Analysis of variance (ANOVA) was used to separate means and Duncans new multiple range test was used to compare means. P < 0.05 was accepted as the level of significance.

Results

Table 1 shows the haemoglobin (Hb) concentration of rats fed raw and boiled leaf juice of *Cnidoscopus aconitifolius*. Group 6 (experimental group) had the highest haemoglobin concentration (14.93 g/dl ± 0.83) after treatment while group 7 (negative control group) had the least haemoglobin concentration (6.68 g/dl ± 1.36) after treatment. The Hb concentration of some experimental groups (groups 1 and 3) and negative control group (group 7) did not increase above the baseline value while the haemoglobin concentration of the rest of the experimental group (groups 2, 4, 5 and 6) and normal control group (group 8) increased above the baseline value.

Groups	Baseline (g/dl)	After induction (g/dl)	After treatment (g/dl)	Percentage increase/ decrease (%)
Group 1	13.83 ± 1.49 ^{ab}	6.88 ± 0.15 ^a	13.35 ± 0.97 ^b	-3.59
Group 2	12.90 ± 0.36 ^{ab}	8.45 ± 0.58 ^{cd}	13.35 ± 0.53 ^b	+3.37
Group 3	13.30 ± 1.90 ^{ab}	8.73 ± 0.19 ^c	13.20 ± 0.47 ^b	-0.76
Group 4	11.65 ± 2.69 ^a	7.60 ± 0.36 ^{abc}	13.13 ± 0.53 ^b	+11.27
Group 5	12.63 ± 0.53 ^{ab}	7.48 ± 0.88 ^{ab}	14.70 ± 0.54 ^c	+14.08
Group 6	14.35 ± 0.40 ^b	8.78 ± 0.67 ^d	14.93 ± 0.83 ^c	+3.88

Group 7	14.40 ± 1.22 ^b	8.20 ± 0.70 ^{bcd}	6.68 ± 1.36 ^a	-115.57
Group 8	13.90 ± 2.18 ^{ab}	14.10 ± 0.61 ^e	14.88 ± 0.91 ^c	+6.59

Table 1: Haemoglobin concentration (Hb) of rats fed with raw and boiled leaf juice of *Cnidoscopus aconitifolius* (g/dl).

Mean ± S.D

Mean value in the same row with different superscripts are significantly different at $p < 0.05$

Key: Groups 1-3 were induced with anaemia and treated with 5 ml/kgBW, 10 ml/kgBW and 15 ml/kgBW of the raw leaf juice of *Cnidoscopus aconitifolius* respectively.

Groups 4-6 were induced with anaemia and treated with 5 ml/kgBW, 10ml/kgBW and 15ml/kgBW of the boiled leaf juice of *Cnidoscopus aconitifolius* respectively.

Group 7 (negative control group) was induced with anaemia without treatment with either of the juices.

Group 8 (normal control group) was not induced and was not treated with either of the juices.

Table 2 shows the packed cell volume (PCV) of rats fed raw and boiled leaf juice of *Cnidoscopus aconitifolius*. Group 6 and 8 (experimental group) had the highest PCV (45.00% ± 2.58) after treatment while group 7 (negative control group) had the least PCV (22.50% ± 3.42) level after treatment. The PCV of some experimental group (groups 1 and 3) and the negative control group (group 7) did not increase above the baseline while the PCV of the rest of the experimental group (groups 2, 4, 5 and 6) and the normal control group (group 8) increased above the baseline.

Groups	Baseline (%)	After induction (%)	After treatment (%)	Percentage increase / decrease (%)
Group 1	41.50 ± 4.51 ^{ab}	21.75 ± 1.71 ^a	39.50 ± 2.52 ^b	-5.06
Group 2	38.75 ± 0.96 ^{ab}	26.50 ± 1.30 ^b	40.75 ± 2.22 ^{bc}	+4.91
Group 3	40.00 ± 5.72 ^{ab}	24.75 ± 2.22 ^{ab}	39.75 ± 1.71 ^b	-0.63
Group 4	35.00 ± 8.08 ^a	22.25 ± 1.26 ^a	40.00 ± 2.83 ^b	+12.5
Group 5	38.00 ± 1.63 ^{ab}	23.50 ± 2.52 ^{ab}	44.00 ± 1.64 ^{cd}	+13.6
Group 6	43.00 ± 1.15 ^b	26.00 ± 2.16 ^b	45.00 ± 2.58 ^d	+4.44
Group 7	43.25 ± 3.59 ^b	24.75 ± 2.50 ^{ab}	22.50 ± 3.42 ^a	-92.22
Group 8	41.75 ± 6.65 ^{ab}	42.25 ± 1.71 ^c	45.00 ± 2.58 ^d	+7.22

Table 2: Packed cell volume (PCV) of rats fed raw and boiled leaf juice of *Cnidoscopus aconitifolius* (%).

Mean ± S.D

Mean value in the same row with different superscripts are significantly different at $p < 0.05$

Key: Groups 1-3 were induced with anaemia and treated with 5 ml/kgBW, 10 ml/kgBW and 15 ml/kgBW of the raw leaf juice of *Cnidoscopus aconitifolius* respectively.

Groups 4-6 were induced with anaemia and treated with 5 ml/kgBW, 10 ml/kgBW and 15 ml/kgBW of the boiled leaf juice of *Cnidoscopus aconitifolius* respectively.

Group 7 (negative control group) was induced with anaemia without treatment with either of the juices.

Group 8 (normal control group) was not induced and was not treated with either of the juices.

Table 3 shows the red blood cell (RBC) count of rats fed raw and boiled leaf juice of *Cnidoscopus aconitifolius*. Group 7 (negative control group) had the lowest RBC (26.75 ± 6.99) after treatment while group 6 (experimental group) had the highest RBC (260.00± 18.26) count after treatment. The RBC count of some experimental group (groups 1 and 3) and negative control group (group 7) did not increase above the baseline value while the RBC of the rest of the experimental group (groups 2, 4, 5 and 6) and normal control group (group 8) increased from the baseline.

Groups	Baseline (10 ⁶ /ul)	After induction (10 ⁶ /ul)	After treatment (10 ⁶ /ul)	Percentage increase / decrease (%)
Group 1	137.00 ± 15.03 ^a	97.50 ± 8.66 ^a	130.25 ± 7.32 ^b	-5.18
Group 2	126.25 ± 7.14 ^{ab}	96.00 ± 10.20 ^a	132.50 ± 17.08 ^b	+4.72
Group 3	127.25 ± 16.52 ^{ab}	93.00 ± 6.00 ^a	112.50 ± 12.58 ^b	-13.11
Group 4	110.50 ± 21.81 ^a	93.25 ± 4.11 ^a	138.50 ± 5.97 ^b	+20.2
Group 5	120.50 ± 9.98 ^{ab}	93.00 ± 12.06 ^a	125.00 ± 34.16 ^b	+3.6
Group 6	140.25 ± 1.71 ^a	92.25 ± 7.41 ^a	260.00 ± 18.26 ^c	+46.06
Group 7	137.00 ± 11.60 ^a	93.50 ± 8.06 ^a	26.75 ± 6.99 ^a	-412.15
Group 8	128.50 ± 23.40 ^{ab}	169.25 ± 6.40 ^b	242.50 ± 26.30 ^c	+47.01

Table 3: Red blood cell (RBC) count of rats fed with raw and boiled leaf juice of *Cnidoscolus aconitifolius* (10⁶/ul).

Mean ± S.D

Mean value in the same row with different superscripts are significantly different at *p* < 0.05

Key: Groups 1-3 were induced with anaemia and treated with 5 ml/kgBW, 10 ml/kgBW and 15 ml/kgBW of the raw leaf juice of *Cnidoscolus aconitifolius* respectively.

Groups 4-6 were induced with anaemia and treated with 5 ml/kgBW, 10 ml/kgBW and 15 ml/kgBW of the boiled leaf juice of *Cnidoscolus aconitifolius* respectively.

Group 7 (negative control group) was induced with anaemia without treatment with either of the juices.

Group 8 (normal control group) was not induced and was not treated with either of the juices.

Table 4 shows the white blood cell count (WBC) of rats fed raw and boiled juice of *Cnidoscolus aconitifolius*. The WBC of some experimental group (groups 1, 3, 4, 5 and 6) and normal control group (group 8) did not increase above the baseline after treatment. Group 7 (negative control group) had the highest WBC count (11700 mcg ± 258.20) and group 2 (experimental group) had a slight increase (7700 mcg ± 739.37) in WBC after treatment.

Groups	Baseline (mcg)	After induction (mcg)	After treatment (mcg)	Percentage increase / decrease (%)
Group 1	6625 ± 434.93 ^{bcd}	10450 ± 1535.14 ^{ab}	5400 ± 294.39 ^b	-22.69
Group 2	7575 ± 330.40 ^d	11400 ± 2540.34 ^b	7700 ± 739.37 ^c	+1.62
Group 3	7350 ± 998.33 ^{cd}	10750 ± 838.65 ^{ab}	7300 ± 1732.05 ^c	-0.68
Group 4	6075 ± 596.52 ^{ab}	10775 ± 1255.32 ^{ab}	4250 ± 387.30 ^a	-42.94
Group 5	7500 ± 1437.59 ^{cd}	12500 ± 886.94 ^b	5050 ± 341.57 ^{ab}	-48.51
Group 6	5300 ± 258.20 ^a	12250 ± 718.80 ^b	4100 ± 258.20 ^a	-29.27
Group 7	6250 ± 1147.46 ^{abc}	11150 ± 822.59 ^{ab}	11700 ± 258.20 ^e	+46.58
Group 8	9300 ± 258.20 ^e	9200 ± 516.40 ^a	8900 ± 529.15 ^d	-4.49

Table 4: White blood cell (WBC) count of rats fed with raw and boiled leaf juice of *Cnidoscolus aconitifolius* (mcg).

Mean ± S.D

Mean value in the same row with different superscripts are significantly different at *p* < 0.05

Key: Groups 1-3 were induced with anaemia and treated with 5 ml/kgBW, 10 ml/kgBW and 15 ml/kgBW of the raw leaf juice of *Cnidoscolus aconitifolius* respectively.

Groups 4-6 were induced with anaemia and treated with 5 ml/kgBW, 10 ml/kgBW and 15 ml/kgBW of the boiled leaf juice of *Cnidoscolus aconitifolius* respectively.

Group 7 (negative control group) was induced with anaemia without treatment with either of the juices.

Group 8 (normal control group) was not induced and was not treated with either of the juices.

Table 5 shows the mean cell volume (MCV) of rats fed raw and boiled leaf juice of *Cnidoscolus aconitifolius*. Group 7 (negative control group) had the highest MCV (8.61fL ± 1.14). The MCV of some experimental group (groups 1, 4 and 6) and normal control group (group 8) did not increase above the baseline while the MCV of the rest of the experimental group (groups 2, 3 and 5) increased above the baseline value.

Groups	Baseline (fL)	After induction (fL)	After treatment (fL)	Percentage increase / decrease (%)
Group 1	3.03 ± 0.02 ^a	2.24 ± 0.23 ^a	3.03 ± 0.08 ^b	0
Group 2	3.07 ± 0.10 ^a	2.78 ± 0.26 ^{bc}	3.17 ± 0.49 ^b	+3.15
Group 3	3.14 ± 0.15 ^{ab}	2.66 ± 0.19 ^{abc}	3.57 ± 0.50 ^b	+12.04
Group 4	3.15 ± 0.16 ^{ab}	2.39 ± 0.24 ^{ab}	2.88 ± 0.14 ^b	-9.38
Group 5	3.16 ± 0.13 ^{ab}	2.54 ± 0.17 ^{abc}	3.77 ± 1.25 ^{abc}	+16.18
Group 6	3.06 ± 0.07 ^a	2.84 ± 0.49 ^c	1.74 ± 0.19 ^a	-75.86
Group 7	3.15 ± 0.09 ^{ab}	2.65 ± 0.19 ^{abc}	8.61 ± 1.14 ^c	63.41
Group 8	3.26 ± 0.10 ^b	2.49 ± 0.13 ^{ab}	1.86 ± 0.10 ^a	-75.27

Table 5: Mean cell volume MCV of rats fed with raw and boiled leaf juice of *Cnidoscolus aconitifolius* (fL).

Mean ± S.D

Mean value in the same row with different superscripts are significantly different at $p < 0.05$

Key: Groups 1-3 were induced with anaemia and treated with 5 ml/kgBW, 10 ml/kgBW and 15 ml/kgBW of the raw leaf juice of *Cnidoscolus aconitifolius* respectively.

Groups 4-6 were induced with anaemia and treated with 5 ml/kgBW, 10 ml/kgBW and 15 ml/kgBW of the boiled leaf juice of *Cnidoscolus aconitifolius* respectively.

Group 7 (negative control group) was induced with anaemia without treatment with either of the juices.

Group 8 (normal control group) was not induced and was not treated with either of the juices.

Discussion

The haemoglobin concentration of the anaemic rats treated with raw and boiled leaf juices of *Cnidoscolus aconitifolius* were significantly ($p < 0.05$) increased. Haemoglobin is a protein used by red blood cells to distribute oxygen to other tissues and cells in the body [20]. According to [21] haemoglobin is composed of an iron-containing substance called haem. Increase in haemoglobin might be due to the iron and vitamin C content of raw and boiled leaf juices. The iron was, however, of plant origin (non-haem) and as a result was closely bound to organic molecules in the juice as ferric acid. Vitamin C which is known to enhance iron absorption by reducing ferric iron to ferrous iron may have contributed to the increased haemoglobin concentration observed in the anaemic rats treated with the raw and boiled *Cnidoscolus aconitifolius* leaf juices.

Significant ($p < 0.05$) increases in PCV of the anaemic rats treated with the raw and boiled *Cnidoscolus aconitifolius* leaf juices was observed and this is indicative of the normal functioning of the bone marrow in the process of erythropoiesis [22]. According to Qasim [23] PCV is the proportion of blood volume that is occupied by red blood cells.

Red blood cells (RBCs) whose main function is to carry oxygen which it picks up in the lungs to the cells of the body and to transport CO₂ from the cells to the lungs for excretion [24], was significantly ($p < 0.05$) increased in the anaemic rats treated with raw and boiled *Cnidoscolus aconitifolius* leaf juices. This shows that the raw and boiled leaf juices both have the ability to stimulate erythropoietin release into the kidney which according to [24] is the humoral regulator of RBCs production. This process is known as erythropoiesis. During erythropoiesis, renal erythropoietic factor (an enzyme) is secreted in response to peritubular cell hypoxia. This factor interacts with a

plasma protein to form erythropoietin [24]. According to [25] erythropoietin increases the number of erythropoietin-sensitive committed stem cells in the bone marrow that are converted to RBCs and subsequently to mature erythrocytes.

The significant ($p < 0.05$) increase in the haematological parameters (haemoglobin, PCV and RBCs) of the anaemic rats treated with the raw and boiled *Cnidoscopus aconitifolius* leaf juices could be attributed to the vitamins and minerals composition of the leaf juices. These vitamins and minerals are well-known haemopoietic factors that have direct influence on blood production. Therefore, the possible mechanism responsible for the haematinic activity of the raw and boiled leaf juices of *Cnidoscopus aconitifolius* could be the combined haematinic effects of iron, copper, zinc, folate, vitamin C, vitamin B₆ as well as vitamin B₁₂.

Leucocytosis, which is an elevation in total WBCs count ($> 11,000/\mu\text{L}$) (George-Gay & Parker, 2003) was observed in the group of anaemic rats which were not treated with either of the leaf juices (negative control group). WBCs are cells of the blood that the body uses to fight infections and react to foreign bodies. According to Ode and Nwaehujor [26] leucocytosis is a normal body response to an underlying pathophysiological condition which in this case happens to be anaemia. Leucocytosis was also observed after anaemia induction in some experimental group (groups 2, 4, 5 and 6) and the negative control group. White blood cells (WBCs) of the group of anaemic rats treated with the raw and boiled leaf juices of *Cnidoscopus aconitifolius* decreased significantly ($p < 0.05$). The decrease observed could be as a result of suppression of leucocytosis by the extract or from suppression of the production of WBCs in the bone marrow.

Conclusion

Raw and boiled *Cnidoscopus aconitifolius* leaf juices reversed anaemia in cyclophosphamide-induced anaemic male albino rats. This affirms the traditional use of *Cnidoscopus aconitifolius* leaf juice as blood booster by Aku people of Enugu State, Nigeria.

Recommendation

Further studies are needed on the phytochemical and anti-nutrient composition of the raw and boiled *Cnidoscopus aconitifolius* leaf juices, acute toxicity of the leaf juices and the toxicological effect of the leaf juices on the liver and kidney.

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