ABSTRACT

Introduction: Medicinal plants have become increasingly useful as a form of alternative therapy. *Cnidoscolus aconitifolius* is a medicinal plant applied in folklore remedies in the treatment and prevention of various diseases.

Aim: The aim of this study is to assess the effects of *Cnidoscolus aconitifolius* on some hematological parameters of male Wistar rats.

Methodology: A total of 15 male Wistar rats weighing between 100-250 g were randomly placed in groups. Group 1 served as control and received distilled water only; group 2 received 200 mg/kg and group 3 received 400 mg/kg of the hydromethanolic leaf extract of *Cnidoscolus aconitifolius*. Extract was administered once daily using oro-gastric cannula for 30 days. Blood samples were collected by direct cardiac puncture into appropriate sample tubes for estimation of hematological parameters including red blood cell count (RBC), packed cell volume (PCV), haemoglobin (Hb) concentration, white blood cell count (WBC) and platelet count.
Results: Results showed a significant \((P<0.05)\) increase in RBC, PCV, Hb and platelet count with the higher dose of 400 mg/kg body weight of the extract compared to control group. However, the WBC count was not significantly \((P>0.05)\) altered.

Conclusion: Oral administration of the leaf extract of *Cnidoscolus aconitifolius* increases red blood cell count, packed cell volume, hemoglobin level and platelet count at 400 mg/kg body weight.

Keywords: *Cnidoscolus aconitifolius*; hematological parameters; hydromethanolic; wistar rats.

1. INTRODUCTION

There is an increasing dependence on medicinal plants for the prevention and treatment of various illnesses around the world. It has been estimated that, a large number of the population in developing countries rely heavily on medicinal plants and the services of traditional medicine practitioners to meet their health needs [1]. The importance attached to the use of medicinal plants is derived from its affordability, minimal side effects, and accessibility compared to modern medicines [2].

Specific compounds found in most medicinal plants are effective in the treatment and prevention of diseases. These compounds are frequently extracted and used as raw materials in the synthesis of different drugs [1].

Blood indices particularly, red and white cells and hemoglobin concentration, are mostly useful clinical indicators of disease state. These indicators are controlled in healthy persons [3]. Any gene mutation that affect hematological parameters have significant phenotypic consequences, not excluding multiple variants that impact susceptibility to diseases. However, most variability in blood is continuous and is influenced by multiple effects [4].

*Cnidoscolus aconitifolius* is propagated normally by wooden stem cuttings about 6-12 inches long, as seeds are produced only rarely. Early growth is slow as roots are slow to develop on the cutting, so leaves are not harvested until the second year. Parts of *Cnidoscolus aconitifolius* such as shoots and leaves are used; as laxative and circulating stimulant to improve digestion, for treating diabetes and stimulation of lactation. The plant reportedly has high fiber content and also possesses antibacterial activities [5]. The edible parts of the plant, which taste like spinach when cooked serve as important nutritional source of protein, vitamin (A and C), minerals (calcium, iron and phosphorus), niacin, riboflavin and thiamine [5]; some of which are factors necessary for erythropoiesis. There are anecdotal reports showing that *Cnidoscolus aconitifolius* is used to boost blood parameters. This study therefore, aims to determine the effects of *Cnidoscolus Aconitifolius* on some hematological parameters in male Wistar rats.

2. MATERIALS AND METHODS

2.1 Plant Materials Extraction and \(LD_{50}\)

Leaves of *Cnidoscolus aconitifolius* were obtained from our institution Garden and were identified by Dr. Ekeke Chimezie. of the Department of Plant Science and Biotechnology, University of Port Harcourt, Nigeria. Voucher specimen of the plant was deposited in the herbarium with the reference number: UPH/P/219. Our study was conducted in accordance with the guidelines for the care and use of laboratory animals issued by the United States Institute for Laboratory and Animal Research [6].

Fresh leaves of *Cnidoscolus aconitifolius* were air dried for a minimum of 2 weeks. The dried leaves were pulverized with a grinding machine into pieces. Extraction was carried out with Soxhlet extractor using hydromethanol as solvent. The solution was filtered using Whatman filter paper. The filtrate was concentrated under reduced pressure in vacuum at 45°C using rotary evaporator (Gallen Kamp UK). The extract yield was stored at 4°C until used for animal feeding experiments. Our choice of extract doses was predicated on the lethal toxicity test \((LD_{50})\) results determined by Akachukwu et al., 2014 [7].

2.2 Experimental Animals

A total of 15 male Wistar rats weighing between 100-250 g were procured and housed at the animal house, Department of Human Physiology, Faculty of Basic Medical Sciences, University of Port Harcourt, Nigeria. The animals were fed with normal rat pellet and tap water *ad libitum* and subsequently acclimatized for 14 days after
which they were grouped. This study was carried out in the animal house of the Department of Human Physiology, Faculty of Basic Medical Sciences, College of Health Sciences, University of Port Harcourt, Nigeria.

2.3 Experimental Design

The rats were randomly placed in three groups of 5 rats each. Group 1 served as control and received distilled water only. Group 2 received 200mg/kg body weight of the extract and group 3 received 400mg/kg body weight of extract. The extract was administered once daily using an oral cannula. On completion of treatment, the animals were made unconscious with chloroform inhalation (cotton wool soaked in 3.5% chloroform) and blood samples were collected by direct cardiac puncture using a 5-ml syringe attached to a needle (21 SWG); the blood was collected into plain capped bottles containing ethylenediaminetetraacetate (EDTA) for the determination of some hematological parameters.

2.4 Determination of Hematological Parameters

Hematological parameters and indices were determined using the practicability of a quick diagnostic laboratory test with the aid of Reflotron manufactured by Boehringer Mannheim as described by Kurner and Grimm [8].

2.5 Statistical Analysis

This was done using Statistical Package for Social Sciences (SPSS). Results are expressed as mean ± standard error of mean. Significant differences were determined using one-way ANOVA. The P-value of less than 0.05 was considered statistically significant.

3. RESULTS

Results are presented in Tables 1-2.

The mean RBC count of the control, low dose (200 mg/kg) [group 2] and high dose (400 mg/kg) [group 3] were 5.62±0.13, 5.46±0.13 and 6.58±0.21 respectively. The high dose group had significantly (P<0.05) higher values compared to control. On the other hand, the values obtained for RBC count in group 2 showed no significant difference.

The mean PCV was found to be 39.80±0.86% in the control group, 39.40±0.51% in the low dose group and 43.40±1.03% in the high dose group respectively. The PCV of the high dose group was significantly (P<0.05) higher compared to the control group.

The mean Hb concentrations were 13.18±0.27, 13.14±0.17 and 14.44±0.34 g/dl for control, low dose and high dose groups respectively. Significant difference (P<0.05) was observed in the mean value of rats in group 3 compared to group 1.

Table 1. Effect of hydromethanolic extract of *Cnidoscolus aconitifolius* on RBC count, PCV and Hb

<table>
<thead>
<tr>
<th>Groups/Extracts (mg/kg)</th>
<th>RBC count (x10^6/mm³)</th>
<th>PCV (%)</th>
<th>Hb (g/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>5.62±0.13</td>
<td>39.80±0.86</td>
<td>13.18±0.27</td>
</tr>
<tr>
<td>200</td>
<td>5.46±0.13</td>
<td>39.40±0.51</td>
<td>13.14±0.17</td>
</tr>
<tr>
<td>400</td>
<td>6.58±0.21*</td>
<td>43.40±1.03*</td>
<td>14.44±0.34*</td>
</tr>
</tbody>
</table>

Values expressed as Mean ± SEM. n=5. Significant at [*P<0.05] when compared to control group.

Table 2. Effect of hydromethanolic extract of *Cnidoscolus aconitifolius* on WBC count and platelet count

<table>
<thead>
<tr>
<th>Groups/extracts (mg/kg)</th>
<th>WBC count (x10^9/mm³)</th>
<th>Platelets (x10^9/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>9.98±0.87</td>
<td>228.40±5.53</td>
</tr>
<tr>
<td>200</td>
<td>11.30±1.15</td>
<td>260.40±9.22</td>
</tr>
<tr>
<td>400</td>
<td>10.84±0.61</td>
<td>279.00±22.12*</td>
</tr>
</tbody>
</table>

Values expressed as Mean ± SEM. n=5. Significant at [*P<0.05] when compared to control group.
The WBC count for the control group was 9.98±0.87 cells/mm$^3$, while the values for the low dose and high dose groups were 11.30±1.15 and 10.84±0.61 cells/mm$^3$ respectively. The values of the low and high doses were not significantly different compared to control.

The mean platelet values were found to be 228.40±5.53, 260.40±9.22 and 279.00±22.12 $\times 10^9$ L for the control, rats administered with 200 mg/kg and those that received 400 mg/kg of the extract respectively. Animals in group 3 [400 mg/kg] were found to have significantly higher values for platelet count compared to animals in control group.

4. DISCUSSION

The effect of Cnidoscolus Aconitifolius on some hematological parameters in male Wistar rats was investigated in this study. The extract (at 400 mg/kg body weight) caused an increase in the RBC count. This was confirmed by the increased hematocrit (Hct) and Hb in the high-dose recipient group. Normally, local tissue anoxia apparently leads to the formation of a glycoprotein called erythropoietin, which stimulates increased production of erythrocytes [9]. It is very likely that some of those factors necessary for erythropoiesis which are found in Cnidoscolus aconitifolius leaves [5]; may have contributed to the increased production of erythrocytes.

Similar results were obtained for platelets. There was increased platelet formation in the group that received 400 mg/kg body weight of the extract. Hence, it would seem likely that the extract also contains some compounds that are capable of causing the release of thrombopoietin [10]. Platelets play an important role in the maintenance of normal homeostasis and platelet aggregation; release of thromboxane $A_2$, platelet factor 4, and beta-thromboglobulin; and expression of glycogen 1b and glycogen IIb/IIIa receptors are indicators of platelet function [11][12].

The total WBC counts were not significantly altered following extract administration. However, a close look at the different doses of the extract administered revealed marginal increases in groups 2 and 3 but the total WBC count remained largely unaltered. WBCs are activated when the body is invaded by bacteria and they provide the first line of defence against invading microorganisms [13]. The important role of white cells in offering first line defence against infection and damage to tissues is well established. This finding is in agreement with previous reports that Cnidoscolus aconitifolius and some commonly prescribed medicinal plants contains agents that encourages white cell production [14,15]. This suggests that the extract may not have significant immune boosting properties. Immune boosters are usually recommended to strengthen and harmonize degenerative body functions and assist the immune system to fight invading organisms [14,16].

5. CONCLUSION

The oral administration of Cnidoscolus aconitifolius leaf extract increases red blood cell count, packed cell volume, hemoglobin concentration and platelet count at 400 mg/kg body weight of the extract.

ETHICAL APPROVAL

Animal Ethic committee approval has been taken to carry out this study. The research was approved by the Ethics Committee of the Centre for Research Management and Development of our University.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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5. Awoyinka AO, Balogun IO, Oggunnowo AA. Phytochemical screening and in vitro bioactivity of Cnidoscolus aconitifolius


