Effects of aqueous extract of *Ocimum gratissimum* on haematological parameters of Wistar rats

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Abstract

The effect of aqueous extract of *Ocimum gratissimum* on hematological parameters of Wister rats was studied. Twenty five rats with weight ranging between 100-160g were used. The rats were divided into five groups; with group one as the control group. Increasing doses (0.2, 0.4, 0.8, 1.6g kg⁻¹ body weight) of the extract were administered orally to the other four groups for a period of four weeks. Significant (P< 0.05) decreases in the level of hemoglobin (Hb), packed cell volume (PCV), red blood cells (RBC), white blood cells (WBC) and other hematological parameters were observed.

Keywords: *Ocimum gratissimum*, haematologic effects, Wistar rats

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INTRODUCTION

Nature has provided a very beautiful environment for us to live and survive. Man through the ages has depended on his immediate environment for solving his numerous problems. Man has learnt to depend on plants in order to provide solutions to the myriad of health problem plaguing him.\(^1\)

The plant being investigated is popularly called African basil. It is a shrub commonly found in gardens and around village huts.\(^2\) Scientifically, it is called *Ocimum gratissimum*. It is widely distributed in tropical and warm temperature regions.

It is used commonly in folk medicine to treat different diseases such as upper respiratory tract infections, diarrhea, headache, pneumonia and also as a treatment for cough, fever and conjunctivitis.\(^3,4\)

*Ocimum* is a genus of about thirty five species of aromatic annual and perennial herbs and shrubs in the family Lamiaceae, native to the tropical and warm temperate region of the old world. *Ocimum gratissimum* is one of the species from the genus. It is commonly called African basil or shrubby basil. It is Efinrin in Yoruba, Ebavbokho in Bini, Aai doya ta gida in Hausa and Nchonwu in Igbo.\(^5\)

The chemical composition of *Ocimum gratissimum* are α-pinene, β-pinene, 1,8-cineole, Terpineole, Eugenol, (E)-β-Caryophyllene, Muurolere, Sehirene and β-selinerel (Sainsbury and Sofowora.\(^6\)).

Erythrocytes (red blood cell) which are anucleate are packed with the oxygen carrying proteins hemoglobin. The normal concentration of erythrocytes in blood is approximately 3.9-5.5 million per micro liter in women and 4.1-6 million per micro liter in men.

Human erythrocytes survive in the circulation for about 120 days, worn out erythrocytes are removed from the circulation by macrophages of the spleen and bone marrows. The signal for removal seems to be the appearance of defective complex oligosaccharides attached to integral membrane protein of the plasmalemma.\(^7\)

Leukocytes (white blood cells) migrate to the tissue, where they perform multiple functions and most die by apoptosis. Leukocytes are involved in the cellular and humoral defense of the organism against foreign material. The number of leukocytes in the blood varies according to age, sex and physiological conditions. In normal adults they are roughly 6,000-10,000 Leukocytes per micro liter of blood.

Blood platelets (thrombocytes) are nonnucleated disk like cell fragments 2-4µm in diameter. Platelets originate from the fragmentation of giant polypoid megakaryocytes that reside in the bone marrow. Platelets count range from 200,000 to 400,000 per micro liter of blood. Platelets have a life span of about 10 days. Platelets function: the role of platelets in controlling hemorrhage can be summarized as primary aggregation, secondary aggregation, blood coagulation, clot retraction and clot removal. Due to the widespread consumption of *Ocimum gratissimum*, it is necessary to study its effect on blood, the tissue that transports substances in the body.

MATERIALS AND METHODS

Animals

Twenty-five Wistar rats weighing between 100-160g were used for the study. The rats were purchased from the animal house of Biochemistry Department of University of Ilorin. They were bred for weeks in the animal house of Anatomy department of University of Ilorin. The rats were fed with pellets grower mash obtained from Bendel feed mill, Yoruba road, Ilorin and with water *ad libitum* during the breeding period designed to acclimatize the rats.

The rats were picked at random and grouped into five. A control group, while groups 1-4 were the experimental groups. Each of the rats were marked at the tail with different colours of pen marker and put into different segments of the cage, according to their group.

The rats were sacrificed after four weeks of extract administration using cervical dislocation. Blood from each rat was collected into labeled heparinized bottle to prevent coagulation of the blood.
Preparation of extracts
Fresh leaves of the plant *Ocimum gratissimum* were bought from Oja-Oba market in Ilorin. Botanical identification of the plants was done at the herbarium of the Botany Department of University of Ilorin.

The fresh leaves were kept in the oven at 80°C for ten minutes to stop any enzyme activity and then at 60°C for 30 minutes. They were collected from the oven, air dried and ground into coarse power. 50g of the powdered leaves was stirred into 450 ml of boiling distilled water.

Boiling was allowed to continue for five minutes, the mixture was then kept aside for thirty minutes to allow it to infuse. It was then filtered using a filter paper. The filtrate was concentrated to 200ml (1ml of the extract being equivalent to 0.25g of the starting material). The extract was kept in a refrigerator until it was time to use.

Administration of extract
Administration of the aqueous extract was done orally by means of calibrated syringe with attached rubber cannula. The animals received their doses daily for four weeks.

Control group received a quantity of distilled water equivalent to the volume in group 4. Groups 1, 2, 3 and 4 received the aqueous extract of *Ocimum gratissimum* at doses of 0.2, 0.4, 0.8 and 1.6gkg⁻¹ respectively.

Hematological parameters
Evaluation of the hematological parameters was carried out using automated hematological Analyzer K-X- 21 made by Symex, Kobe, Japan.

Sample of blood from the wistar rats in heparinized bottle were analyzed using this machine for accuracy. Data obtained were analysed using t-test and chi square. Level of significance was predetermined as p< 0.05.

RESULTS
The results of the effect of different doses of aqueous extract of *Ocimum gratissimum* on the haematological parameters are shown in Figs 1-4.

The machine analyzed blood samples to give about ten parameters, which include hemoglobin PCV, MCHC, RBC, MCH, MCV, WBC, platelets, Neutrophil and lymphocytes.

![Figure 1: Effect of aqueous extract of *Ocimum gratissimum* on Haemoglobin](image1)

![Figure 2: Effect of aqueous extract of *Ocimum gratissimum* on PCV](image2)

![Figure 3: Effect of aqueous extract of *Ocimum gratissimum* on MCHC](image3)

![Figure 4: Effect of aqueous extract of *Ocimum gratissimum* on lymphocytes](image4)
The mean Haemoglobin decreased significantly ($p=0.04$) as compared with the control group as shown in Fig. 1. The mean packed cell volume (PCV) value in the treated animals was significantly reduced ($p=0.031$) at the end of the treatment period as compared with the control group (Fig. 2).

The mean corpuscular haemoglobin concentration also decreased as compared with the control. However this was not significant ($p=0.066$). The mean RBC count decreased significantly ($p =0.0217$) at the end of the treatment period as compared with the control group (Table 1). Both the mean corpuscular haemoglobin and the mean corpuscular volume decreased significantly as compared with the control (Table 1).

**Table 1:** Effect of aqueous extract of *Ocimum gratissimum* on RBC count, MCV and MCH

<table>
<thead>
<tr>
<th>Groups</th>
<th>Values (x10^12/L)</th>
<th>Values (pg)</th>
<th>Values (F1)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>7.59</td>
<td>20.67</td>
<td>57.67</td>
</tr>
<tr>
<td>0.2 g/kg</td>
<td>7.08</td>
<td>19.75</td>
<td>55.25</td>
</tr>
<tr>
<td>0.4 g/kg</td>
<td>6.92</td>
<td>19.59</td>
<td>53.25</td>
</tr>
<tr>
<td>0.8 g/kg</td>
<td>6.33</td>
<td>19.5</td>
<td>51.75</td>
</tr>
<tr>
<td>1.6 g/kg</td>
<td>5.57</td>
<td>18.4</td>
<td>50.8</td>
</tr>
</tbody>
</table>

**Table 2:** Effect of aqueous extract of *Ocimum gratissimum* on WBC and platelets

<table>
<thead>
<tr>
<th>Groups</th>
<th>White cell count (x10^9/L)</th>
<th>Platelet count (x10^9/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>10.03±0.031</td>
<td>798.7±2.30</td>
</tr>
<tr>
<td>0.2 g/kg</td>
<td>8.03±0.024</td>
<td>660±1.42</td>
</tr>
<tr>
<td>0.4 g/kg</td>
<td>6.65±0.041</td>
<td>581.5±2.51</td>
</tr>
<tr>
<td>0.8 g/kg</td>
<td>5.58±0.015</td>
<td>524±3.01</td>
</tr>
<tr>
<td>1.6 g/kg</td>
<td>4.08±0.024</td>
<td>584±2.08</td>
</tr>
</tbody>
</table>

**Table 3:** Effect of aqueous extract of *Ocimum gratissimum* on neutrophil

<table>
<thead>
<tr>
<th>Groups</th>
<th>Values (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>32</td>
</tr>
<tr>
<td>0.2 g/kg</td>
<td>30</td>
</tr>
<tr>
<td>0.4 g/kg</td>
<td>28.75</td>
</tr>
<tr>
<td>0.8 g/kg</td>
<td>25.25</td>
</tr>
<tr>
<td>1.6 g/kg</td>
<td>19.6</td>
</tr>
</tbody>
</table>

The white blood cell and platelets counts decreased in the experimental groups compared with the control group (Table 2). This was not statistically significant. There was no statistically significant decrease in the neutrophil count as compared with control group (Table 3). The Lymphocytes count also decreased ($p=0.067$) when compared to the control (Table 4).

**DISCUSSION**

This work tested the effect of aqueous extract of *Ocimum gratissimum* on hematological parameters of wistar rats. The results of the study show that, the leaf extract of *Ocimum gratissimum* administered at the dosages used and for the duration of the experiment suppress the haemopoetic system.

The hemoglobin values at the end of the experiment were 14.70 g/dl on the average for the control group and 12.95, 12.48, 12.15 and 10.72 g/dl on the average for the experimental groups. This shows a reduction in the hemoglobin level, this is in consonance with the finding of Ephraim *et al* in which the hemoglobin value decrease significantly after administration of aqueous extract of *Ocimum gratissimum* to rabbits.

The packed cell volume at the end of the experiment for the control group is 42.7% on the average and 36.5, 34.75, 30.5, 27.2 % on the average for the experimental groups. This shows a decrease in the PCV value as compared to the control, this also agrees with the finding of Ephraim *et al*.

The mean corpuscular hemoglobin MCH at the end of the experiment is 20.67pg on the average for the control group and 19.75, 19.50, 19.50, 18.40 for the experimental group on the average. The MCH value decreases at the end...
of the experiment as compared to the control group value. This is also similar to the findings of Ephraim et al. The mean corpuscular volume MCV at the end of the experiment is 57.67 fl on the average for the control group and 55.25, 53.25, 51.75, 50.80 on the average for the experimental groups. These values show a decrease in MCV value as compared to the control group; this is supported by the findings of Ephraim et al.

The white blood cell count give a mean value of 10.03x10^9/L for the control group at the end of the experiment and a mean values of 8.03, 6.65, 5.58, 4.08x10^9/L for the experimental groups, a significant decrease in the WBC count was observed in the experiment group as compared to the control. Ephraim et al. reported a decrease in WBC count by their work.

The platelets count at the end of the experiment for the control group is 798.7x10^10/L on the average and 660, 581.5, 524.0, 384.0x10^10/L on the average for the experimental groups. These values show a decrease in the platelets count as compared to the control group; this is similar to the findings of Ephraim et al.

The neutrophil level at the end of the experiment for the control group is 32.0% on the average and 30.0, 28.75, 25.25, 19.6% for the experimental groups on the average, a significant decrease on the neutrophil level was observed in the experimental groups as compare to the control. This is supported by the findings of Ephraim et al.

The lymphocyte level at the end of the experiment is 81.0% on the average for the control group and 77.0, 69.5, 66.6% for the experimental groups on the average. The lymphocyte level decreased at the end of the experiment as compared to the control group. This is supported by the findings of Ephraim et al.

In conclusion, the study shows that the leaf extract of Ocimum gratissimum administered at the dosages used and for the duration of the experiment suppress the haemopoetic system. The reduction may have occurred due to lysis of blood cells and probably suppression of blood cell synthesis by saponins found in the leaf extract. Saponins are known to be toxic to body systems. Despite of the popularity of the plant as a condiment and herbal medicine, the extract has been observed to suppress the haemopoetic system. It is therefore suggested that chronic usage of the leaf is not advisable.

REFERENCES