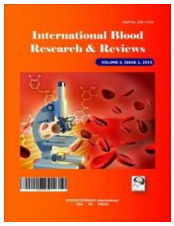

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Hematological Status and Organs/Body-weight Parameters in Wister Rats during Chronic Administration of *Cassia occidentalis*

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Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

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ABSTRACT

Background: *Cassia occidentalis* has been used in traditional medicine since prehistoric times and it's now known to be a natural medicine with many beneficial effects,
Objective: The present study aimed at evaluating the effects of methanol extract from this plant on haematological parameters in rats.
Methodology: A total of ten (10) white albino rats with average weight of 140±2.50 g were grouped into 2 groups of 5 animals each. Group 1 rats serve as control group, while group 2 was treated with 600 mg/kg of *Cassia occidentalis*, for 30 days. The haematological parameters were determined using the automated haematologic analyzer SYSMEX KX21, using standard techniques. The data

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were analyzed using ANOVA and the level of significance was at $P<0.05$.
Results: Showed that administration of the plant extract at dose of 600 mg/kg body weight did not produce any significant effect (P>0.05) on the RBC Hb, MCV, MCH, RCD width-cv and RCD width-sd but cause significant decrease and increase in HCT and MCH-C respectively compare to control rats. The extract also increased the WBC count and LY count but decrease the mid cell total count and granulocyte compared to the control rats. The extract also decrease platelate count and plateletecrit but had no significant effects on mean platelate volume and platelete distribution weight. However, the body weight gain and relative organ weight ratios were not significantly different from those of the control rats.
Conclusions: In conclusion, administration of methanol extracts of *C. occidentalis* at 600 mg/kg investigated has brought about alterations in the WBC and Ly among thrombocytic indices, platelate count and plateletecrit among leucocytic indices and cause alteration to only HCT and MCH-C among the erythrocytic lineage. This may be an indication of local systemic toxicity.

Keywords: Cassia occidentalis; haematology; erythrocyte; leucocyte, thrombocyte.

1. INTRODUCTION

The uses of medicinal plants are gaining popularity in developing countries as has been estimated that 80% of the world populations still relies on medicinal plants for their primary health care needs [1]. African traditional healer typically diagnoses and treats the psychological basis of an illness before prescribing medicines to treat the symptoms [2]. In sub-Saharan Africa, the ratio of traditional healers to the population is 1/500, while that for medical doctors to the population is 1/40 000 [3]. This is as a result of the high cost of Western pharmaceuticals and health care, or because the traditional medicines are more acceptable from a cultural and spiritual perspective [4]. The use of traditional medicine is not restricted to the developing countries. FAO [5], reported that at least 25% of drugs used in modern pharmacopoeia are derived from plants, while many others are synthetic analogues built on prototype compounds isolated from plants. The active ingredients of natural products are chemicals that are similar to those in purified medications, but natural products lack defined dose and potency data and have the same potential to cause serious adverse effects [6]. Emphasizes has therefore, been laid that safety should be overriding criteria in the selection of herbal medicine for use in health care [5].

Cassia occidentalis Linn belongs to Caesalpiniaceae family. It is an erect herb, commonly found by road sides, ditches and waste dumping sites. *C. occidentalis* has been widely used as traditional medicine. Entire parts of the plant have medicinal values [7]. The parts of the plant used are roots, leaves and seeds. The plant is used for fever, menstrual problems, tuberculosis, diuretic anemic, liver complaints, and as a tonic for general weakness and illness. The plant is also used to cure sore eyes, hematuria, rheumatism, typhoid, asthma, and disorder of haemoglobin and is also reported to cure leprosy. An infusion of the plant bark is given by the folklore in diabetes [8]. Biological activities of *C. occidentalis* including antimicrobial [9], antimalarial [10], antioxidant and hepatoprotective [11], antiinflamatory [12], immunosuppressive [13] and larvicidal [14] effect, have been reported.

Since *C. occidentalis* has been used in traditional medicine since prehistoric times and it's now known to be a natural medicine with many beneficial effects, it is therefore necessary to evaluate the safety of this plant for therapeutic purposes. Also, since the assessment of haematological parameters can be used to determine the extent of deleterious effect of foreign compound including plant extract on the blood [15]. It can also be used to explain blood relating functions of chemical compound/plant extract. The present study aimed at evaluating the effects of methanol extract from *C. occidentalis* on hematological parameters in rats.

2. MATERIALS AND METHODS

2.1 Plant Sample

Fresh leaves of *C. occidentalis* were obtained from Minna, Niger State Nigeria. Taxonomic authentications of the plants were carried out in National Institute of Pharmaceutical Research and Development (NIPRD) Abuja, Nigeria.

2.2 Sample Preparation and Extraction Procedure

The collected fresh leaves of *Cassia occidentalis* was destalked, washed with clean-water, dried at

room temperature and finally grounded using a grinder mill. Extraction of plant materials was performed by weighing 200 g of the powdered plants and extracted by soxhlet extraction using 300 ml each of methanol. The marc were filtered with muslin cloth and solvents removed under reduced pressure in a rotary evaporator. Green colored pastes were obtained, weighed and stored in a refrigerator at 4ºC until required.

2.3 Experimental Animals

A total of fifteen (10) white albino rats (*Rattus novergicus*) of both sex weighing between 120 and 200 g was obtained from the Small Animal Holding Unit of the Department of Biochemistry, Federal University of Technology Minna. The rats were kept in clean plastic cages and maintained under standard laboratory conditions (temperature: 22±3ºC; photoperiod: 12 h natural light and 12 h dark; humidity: 40-45%) [16].The animals were maintained on standard animal feeds (Bendel feeds and flour mills, Edo state, Nigeria) and tap water *ad libitum*. The principles governing the use of laboratory animals as laid out by the Federal university of Technology, Minna Committee on Ethics for Medical and Scientific Research and also existing internationally accepted principles for laboratory animal use and care as contained in the Canadian Council on Animal Care Guidelines and Protocol Review [17], were duly observed.

2.4 Animal Grouping and Extract Administration

Ten (10) Swiss Albino rats were randomly divided into two groups of 5 rats each Group 1 (control) were orally administered with distilled water (Vehicle for extract administration), while groups 2 was treated with 600 mg/kg body weight/day for 30 days.

2.5 Collection of Blood Sample

Collection of Blood sample for analyses was as described previously [18]. Prior to termination of the experiment on day 31, the rats were fasted overnight but distilled water was made available *ad libitum*. Blood samples were collected by cardiac puncture under ether anesthesia. The blood was collected in sample bottles containing EDTA for hematological analyses. The rats were quickly dissected and the whole liver, two kidneys heart, spleen and small intestine were excised, freed of fat, blotted with clean tissue paper and then weighed. The blood samples were analyzed immediately after collection at Center for Genetic Engineering and Biotechnology, Global Institute for Bioexploration Unit, Federal University of Technology, Minna.

2.6 Determination of Body Weight and Relative Organ Weight

The body weights of the rats were determined t and the weight gains were computed. Relative organ weights were computed by expressing the absolute weight of the organs to the body weight of the animals as described previously [1].

Weight gain= Final weight of rat (g)-Initial weight of rat (g)

Relative organ weight= organ weight (g)/body weight (g) x 100

2.7 Determination of Hematological Parameters

The hematological components including haemoglobin (Hb), haematocrite (HCT), red blood cells (RBC), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), white blood cells (WBC), Granulocyte count (GRA) lymphocytes (LY), platelate count (PLT) Mean platelate volume (PCT) Plateletecrit and platelete distribution weight were determined using the automated haematologic analyzer SYSMEX KX21, a product of SYSMEX Corporation, Japan employing the method described by Dacie and Lewis [19].

2.8 Statistical Analysis

Data were analyzed using statistical package for social science (SPSS) version 16 and presented as means±SEM. Comparisons between different groups was done using Analysis of Variance (ANOVA) and Duncan's Multiple Range Test (DMRT). Values of $P<0.05$ were considered as statistically significant as described by Yalta and Talha [20].

3. RESULTS

3.1 Hematological Parameters

The effects of 30 days administration of aqueous extract of *C. occidentalis* at 600 mg/kg body weight on the haematological parameters of albino rats are shown in Figs. 1-2 and Table 1, Administration of the plant extract at dose of 600 mg/kg body weight did not produce any significant effect ($P>0.05$) on the RBC Hb, MCV,

MCH, RCD width-cv and RCD width-sd but cause significant decrease and increase in HCT and MCH-C respectivey compare to control rats (Fig. 1). Administration of the extact also increased the white blood cells count and lymphocytic count but decrease the mid cell total count and granulocyte compared to the control rats (Fig. 2). The extract also decrease platelate count and plateletecrit but had no significant effects on mean platelate volume and platelete distribution weight (Table 1).

3.2 Body Weight and Relative Organ Weight

Administration of methanol extract of *C. occidentalis* had no significant (P>0.05) effects on body weight gain of albino rats (Table 2). The computed relative organ weight ratios indicated that the liver, spleen, kidney, small intestine and heart body weight ratios of the rats were not significantly different from those of the control rats (Table 3).

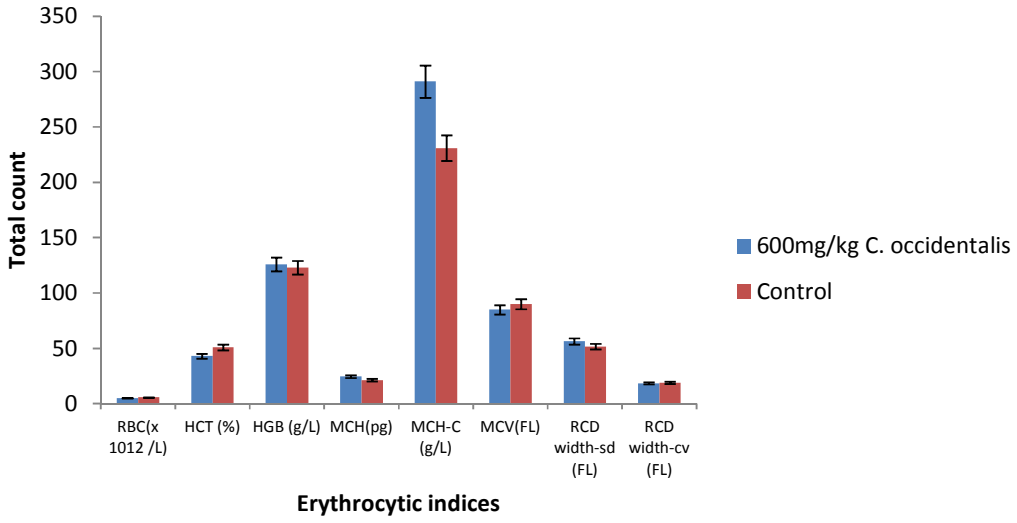

Fig. 1. Effect of methanol extract of *C. occidentalis* on erythrocytic indices of albino rats

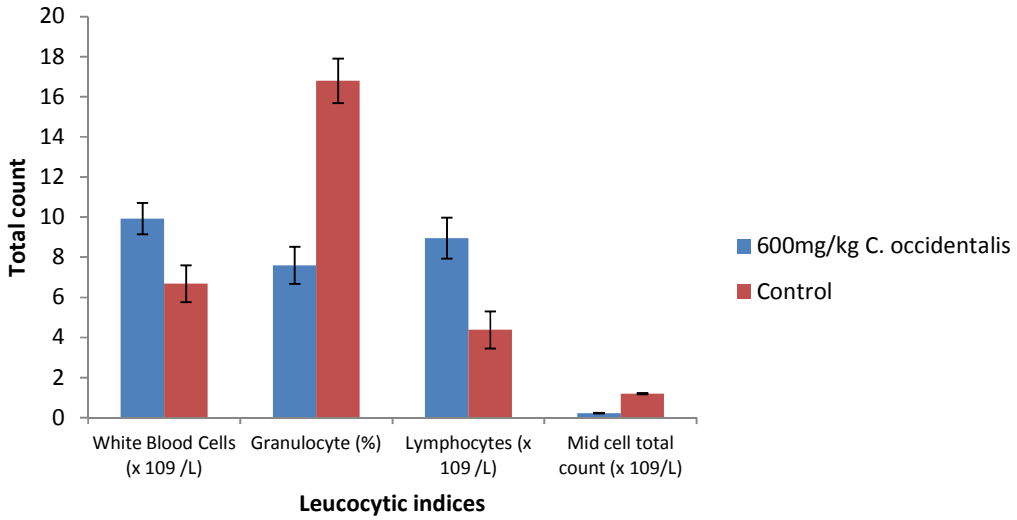

Fig. 2. Effect of methanol extract of *C. occidentalis* on leucocytic indices of albino rats

Table 1. Effect of methanol extract of *C. occidentalis* on thrombocytic indices of albino rats

Parameters	600 mg/kg *C. occidentalis*	Control
Platelate count (x 10^9 /L)	685±36.08[a]	815.5±6.83[b]
Mean platelate volume (FL)	13.70±0.09[a]	14.15±0.35[a]
Plateletecrit (L/L)	0.94±0.15[a]	1.15±0.15[b]
Platelete distribution weight (%)	18.60±0.41[a]	18.50±0.03[a]

Values are mean ± SEM of 5 determinations. The values along the same row with different superscripts are significantly different (p < 0.05)

Table 2. Effect of methanol extract of *C. occidentalis* on weight changes of albino rats

Groups	Initial weight (G)	Final weight (G)	Weight gain (G)
Control rats	183.46±9.51	199.70±8.82	16.24
600 mg/kg *C. occidentalis*	184.42±12.14	201.42±12.01	17.00

Values are mean ± SEM of 5 determinations

4. DISCUSSION

Evaluations of hematological indices provide useful information on the adverse effects of foreign components on the blood and also explain blood-related functions of chemical compounds [21]. The non-significant effect of the extract on the RBC, Hb, MCV, MCH, RCD width-cv and RCD width-sd throughout the experimental period is an indication that there was no destruction of matured RBC's and no change in the rate of production of RBCs (erythropoiesis). It further shows that the extract does not have the potential to stimulate erythropoietin release in the kidney, which is the humoral regulator of RBC production [22]. However, the decrease and increase in HCT and MCH-C respectively compare to control rats suggest the earlier explains selective toxicity of plant extract [23]. White blood cells defend the body against infections or any foreign body [1]. The significant increased the white blood cells count and lymphocytic count caused by the plant extract reflect leucopoetic and possible immunomodulatory effects of the extract which augmented the production of more WBC and Ly [1]. This will increase the animal's capability of generating antibodies in the process of phagocytocis and have high degree of resistance to diseases and enhance adaptability to local environmental and disease prevalent conditions [24]. The decrease in platelate count and plateletecrit caused by 30 days extract administration indicate anti-thrombopoetin potency of the extract and that the blood clotting mechanism of the animals will be inadequate with consequents effects of high loss of blood incase of injury [15].

The results of this study also show that treatment of the rats with the extract for 30 days does not affect the body weight gain of the animals (Table 2). Organ body weight ratios are normally investigated to determine whether the size of the organ has changed in relation to the weight of the whole animal. The absence of an effect on the computed organs/body weight ratios suggest that the extract did not cause any form of swelling, atrophy and hypertrophy on the organs [25].

Table 3. Effect of methanol extract of *C. occidentalis* on relative organ weight of albino rats

Parameters	600 mg/kg *C. occidentalis*	Control rats
Kidney	0.005±0.000[a]	0.005±0.001[a]
Liver	0.023±0.000[a]	0.024±0.005[a]
Heart	0.005±0.001[a]	0.005±0.001[a]
Spleen	0.004±0.002[a]	0.0042±0.000[a]
Small intestine	0.0021±0.001[a]	0.002±0.002[a]

Values are mean ± SEM of 5 determinations. The values along the same row with different superscripts are significantly different (p < 0.05)

5. CONCLUSION

In conclusion, administration of methanol extracts of *C. occidentalis* at 600 mg/kg has brought about alterations in the WBC and Ly among thrombocytic indices, platelate count and plateletecrit among leucocytic indices and cause alteration to only HCT and MCH-C among the erythrocytic lineage. This may be an indication of local systemic toxicity.

CONSENT

It is not applicable.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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