



## EVALUATION OF THE EFFECT OF AQUEOUS SEED EXTRACT OF *Ricinodendron heudelotii* ON THE BLOOD ELECTROLYTE AND HEMATOLOGICAL STATUS OF MALE WISTAR ALBINO RATS

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### ABSTRACT

Plants have widely being used for medicinal and therapeutic purposes, also for maintenance of a healthy biochemical system, the food intake a necessary factor. This study evaluates the effect of the intake of aqueous seed extract of *Ricinodendron heudelotii* on the blood electrolyte and hematological status of male wistar albino rats. Twenty five experimental rats were randomly grouped into five of five rats each; the control group fed with normal rat feed and water without the extract while groups 2, 3, 4 and 5 were in addition to normal feed and water *ad libitum* administered 400, 600, 800 and 1000mg/kg body weight respectively, 24 hourly for a period of 21 days. Blood samples were collected for the blood electrolyte assay; sodium (Na<sup>+</sup>), potassium (K<sup>+</sup>), chloride (Cl<sup>-</sup>), calcium (Ca<sup>++</sup>) and bicarbonate (HCO<sub>3</sub><sup>-</sup>) and hematological parameter; white blood cell (WBC), Hemoglobin (Hb), Red blood cell (RBC), Packed cell volume (PCV), Mean corpuscular hemoglobin concentration (MCHC), Mean corpuscular hemoglobin (MCH), Mean corpuscular volume (MCV), Neutrophils and lymphocytes. The results obtained revealed that administration of aqueous seed extract of *Ricinodendron heudelotii* did not have significant effect  $p \leq 0.05$  on the sodium, potassium, chloride and bicarbonate concentrations, however, a significant difference  $p \leq 0.05$  was observed for the calcium level. The aqueous seed extract caused a concentration dependent and statistical significant increase at  $p \leq 0.05$  in RBC and WBC concentrations. A concentration dependent increase in Hb, PCV, MCH was observed, although not statistically significant. The increase in the WBC, RBC, Hb, MCHC, PCV observed suggests that the aqueous seed extract of *Ricinodendron heudelotii* has the potency to stimulate the production of leucocytes and red blood cells and thereby serve as an immune booster during infections and prevent anemia hence maintain the hematological balance of the body.

**KEYWORDS:** *Ricinodendron heudelotii*, Blood electrolytes, Hematological parameters, medicinal plants.

### INTRODUCTION

An alteration of certain biochemical parameters in blood will destabilize the equilibrium and physiologic function of the blood. Certain disease conditions that stem from nutritional, social and lifestyle activities of humans may cause these changes.

Studies carried out by Odinga *et al.*, 2016 have shown the medicinal and therapeutic values of the seeds of *Ricinodendron heudelotii*, thus agrees with the statement of the father of medicine Hippocrates in 431 B.C. "Let food be thy medicine and medicine thy food". Food made available to all living creatures by nature exists in various forms such as plant seeds, leaves, roots, barks, herbs, trunks etc. These various forms of food have served and still in use due to their nutritive benefits and

their significant importance in the proper functioning of the body's metabolic system. (Sofowora, 1996) reported that foods available to humans in various forms are made up of components such as phytochemicals. Also, foods are easily accessible, less expensive with little or no side effects. *Ricinodendron heudelotii* is one of such food with medicinal and therapeutic potencies (Odinga *et al.*, 2016; Odinga *et al* 2018).

The Blood is a major fluid in the body that serves as a good indicator in determining the health status of an individual. The cellular constituents of the blood are used as important tools in immunotoxicology for the immunotoxic potential of a compound (Ladokun *et al.*, 2015). Blood is a tissue that consists of fluid plasma in which are suspended a number of formed elements. The

blood cells exist at fairly constant levels, suggesting the existence of feedback regulatory mechanisms (Guyton and Hall, 1996; Bowman and Rand, 1980).

Electrolytes are particles or solutes found throughout the body in fluids. They carry an electrical charge and are essential for fluid and acid base balance within the body. The cations (positively charged ions) are sodium ( $\text{Na}^+$ ), potassium ( $\text{K}^+$ ), magnesium ( $\text{Mg}^{++}$ ), and calcium ( $\text{Ca}^{++}$ ). The anions (negatively charged ions) are chloride ( $\text{Cl}^-$ ), bicarbonate ( $\text{HCO}_3^-$ ), sulfate ( $\text{SO}_4^{2-}$ ), and phosphate ( $\text{PO}_4^-$ ). The four major functions of electrolytes are: Regulation of Acid Base Balance, Maintenance of Fluid Balance and Osmolarity, Distribution of the Body Fluid and  $\text{H}_2\text{O}$  between the Compartments and Promoting Neuromuscular Function/Irritability. Fluid and electrolyte monitoring are an essential component of patient assessment. These factors regulate most physiological functions and the acid base balance. Abnormal levels of these electrolytes may result in a variety of pathological disorders (Ganong, 1991).

Haematological parameters are important in establishing the body's functional status as a result of exposure to toxicants, useful indices that can be employed to assess the toxic potentials of plant extracts in living systems (Joshi *et al.*, 2002). They can also be used to explain blood related functions of chemical compound/plant extract. Laboratory investigations involving haematologic parameters have been reported to be highly sensitive, accurate, and reliable and it remains the bedrock of ethical and rational research, disease diagnosis, prevention and treatment (Sunmonu and Oloyede 2002).

The assessment of hematological parameters provide information on inflammation, necrosis, various infections of visceral organs, presence of stress factors (Jurcik *et al.*, 2007; Melillo, 2007; Betancourt-Alonso *et al.*, 2011) as well as the extent of deleterious effect of foreign compound including plant extract on the blood (Yakubu *et al.*, 2007). It also plays a vital role in the physiological, nutrition and pathological status of an organism (Odeghe *et al.*, 2012). Functionally, white blood cells which are immune cells and its differentials fight infections, defend the body by phagocytosis against invasion by foreign organisms and to produce or at least transport and distribute antibodies in immune response (Lawal *et al.*, 2015b).

This study therefore investigates the effect of aqueous seed extract of *Ricinodendron heudelotii* on the blood electrolyte and hematological status of male wistar albino rats.

## MATERIALS AND METHODS

**Plant Material:** The dried seeds of *Ricinodendron heudelotii* were purchased from a local market in Port Harcourt, identified and authenticated in the Department

of Plant Science and Biotechnology, University of Port Harcourt. The clean seeds were pulverized into powder.

**Preparation of Extract:** The ground powder was subjected to extraction using 99% ethanol in the ratio of 1:3 ground powder to ethanol. After 72 hours, the mixture was filtered using whatman number 1 filter paper (Whatman qualitative filter paper No. 1, Camlab UK) and the extract was allowed to concentrate in a water bath to obtain a pure extract. The extract was stored at freezing temperature prior to its use for administration to the experimental animals.

A total of twenty-five (25) male wistar albino rats were purchased from the animal farm, Department of Biochemistry, University of Port-Harcourt and brought into the animal house, Department of Biochemistry, Rivers State University. The rats were weighed and allowed to acclimatize for 14 days.

**Administration procedure:** The twenty-five (25) albino rats were divided into five (5) groups of five (5) rats per group. The treatment design was as follows:

- Group 1: The rats were fed with normal feed and water only. It served as the control.
- Group 2: The rats were fed with normal feed, water and were administered with a dose of 400mg/kg/bw of aqueous seed extract.
- Group 3: The rats were fed with normal feed, water and were administered with a dose of 600mg/kg/bw of aqueous seed extract.
- Group 4: The rats were fed with normal feed, water and were administered with a dose of 800mg/kg/bw of aqueous seed extract.
- Group 5: The rats were fed with normal feed, water and were administered with a dose of 1000mg/kg/bw of aqueous seed extract.

Administration was carried out daily for a period of 21 days after which the rats were sacrificed and their blood collected for blood electrolyte and hematological parameters assay.

## Sample collection

Blood samples were collected from the animals via cardiac puncture with a 5.0ml syringe; this process was repeated for each of the rats. A part of the blood was collected into clean EDTA bottles and the other into plain bottles and taken to the laboratory for the estimation of hematological parameters and blood electrolytes respectively.

The blood electrolytes that were analyzed include; sodium ( $\text{Na}^+$ ), potassium ( $\text{K}^+$ ), chloride ( $\text{Cl}^-$ ), calcium ( $\text{Ca}^{++}$ ) and bicarbonate ( $\text{HCO}_3^-$ ) were measured by an automatic analyzer (MSLAB08, MSL, China).

Parameters that were analysed for haematological status include; white blood cell (WBC), Hemoglobin (Hb), Red blood cell (RBC), Packed cell volume (PCV), Mean

corpuscular hemoglobin concentration (MCHC), Mean corpuscular hemoglobin (MCH), Mean corpuscular volume (MCV), Neutrophils and lymphocytes using an automated hematological machine; Hematology Analyzer (Abacus Junior Vet 5, Austria).

### Statistical Analysis

Data were analyzed by one-way analysis of variance (ANOVA) followed by Student's t-tests using a commercially available statistics software package (SPSS for Windows, V. 15.0) program. Results were presented as means  $\pm$  SD. P values  $\leq 0.05$  were regarded as statistically significant.

## RESULTS AND DISCUSSION

**Table 1: Effect of *Ricinodendron heudelotii* on blood electrolytes of wistar albino rats.**

	Na <sup>+</sup>	K <sup>+</sup>	Cl <sup>-</sup>	Ca <sup>++</sup>	HCO <sub>3</sub> <sup>-</sup>
Group 1	102.4 $\pm$ 3.46 <sup>a</sup>	12.1 $\pm$ 1.30 <sup>a</sup>	77.6 $\pm$ 11.20 <sup>a</sup>	2.24 $\pm$ 3.46 <sup>a</sup>	27.6 $\pm$ 0.75 <sup>a</sup>
Group 2	84.6 $\pm$ 1.43 <sup>a</sup>	11.82 $\pm$ 0.3 <sup>a</sup>	76.2 $\pm$ 9.81 <sup>a</sup>	3.9 $\pm$ 3.46 <sup>c</sup>	26.0 $\pm$ 0.89 <sup>a</sup>
Group 3	103.2 $\pm$ 2.76 <sup>a</sup>	12.88 $\pm$ 1.47 <sup>a</sup>	69.0 $\pm$ 2.93 <sup>a</sup>	3.08 $\pm$ 3.46 <sup>bc</sup>	26.0 $\pm$ 1.41 <sup>a</sup>
Group 4	109 $\pm$ 18.78 <sup>a</sup>	11.82 $\pm$ 0.4 <sup>a</sup>	63.6 $\pm$ 10.83 <sup>a</sup>	2.06 $\pm$ 3.46 <sup>ab</sup>	26.4 $\pm$ 1.33 <sup>a</sup>
Group 5	112.2 $\pm$ 1.28 <sup>a</sup>	10.96 $\pm$ 1.23 <sup>a</sup>	60.4 $\pm$ 2.65 <sup>a</sup>	1.47 $\pm$ 3.46 <sup>e</sup>	29.2 $\pm$ 1.02 <sup>a</sup>

- Values are expressed as Mean  $\pm$  Standard deviation
- Values with different superscripts show significant difference at  $p \leq 0.05$  level.
- Values with the same superscripts show no significant differences.

Electrolytes are minerals found in body fluids that carry electric charge and are essential to keep the heart, nails and muscles functioning very well. As such it is important to maintain a precise and constant electrolyte balance to stay healthy. Having an excess or insufficiency of electrolytes can be dangerous or in some cases, fatal (Gulati, 2016).

From the results obtained as shown in table 1, there was no statistical significant difference ( $P \leq 0.05$ ) in the concentration for Sodium, Potassium, Chloride and Bicarbonate when compared with the control group. However, a statistical significant increase in concentration of calcium was seen. A concentration dependent increase was observed for Sodium, and bicarbonate, also a concentration decrease was observed for chloride and potassium. These increase and decrease in concentration were however not statistically significant  $p \leq 0.05$  when compared with the control group.

The relationship between Sodium and hypertension is based on the capacity of this mineral to "attract" water. When the blood sodium level increase, the body retains water, thereby decreasing the sodium concentration. Blood pressure, which depends in part on blood volume, increases as retained water increases. Reverse is the case when the serum sodium level decreases. In this way, serum levels of sodium can affect blood pressure (Kruetler, 1980).

The result obtained for the serum Potassium levels showed a decrease in potassium levels for groups 2, 3 and 5 when compared to the control group. Kruetler, (1980) suggested that *Ricinodendron heudelotii* has hypokalemic potency and can be used to reverse the consequences of hyperkalemia which are muscle weakness and cardiac arrhythmias that lead to heart

failure. However, this can only be determined when clinical trials are performed on humans.

The results for Chloride ion showed a reduction across all administered groups compared to the control. Chloride levels have been reported to be related to sodium levels.

The results obtained for bicarbonate showed a decrease in bicarbonate levels for groups 2, 3 and 4 compared to the control. The highest dose of 800mg/kg/bw gave the highest bicarbonate level which was slightly higher than the control. The results show little effect of *Ricinodendron heudelotii* seed extract on serum bicarbonate level and thus indicate that it has the ability to stabilize bicarbonate level which is essential for the buffering system of the body.

The results obtained for serum calcium levels showed significant increase for the group administered with 400mg/kg/bw and a significant decrease for the group administered with 600mg/kg/bw. A decreasing trend was observed with increase in concentration for all administered groups. This may be attributed to the influence of the extracts on the influx and efflux of calcium in the extract cellular compartment.

**Table 2: Effect of *Ricinodendron heudelotii* on hematological parameters of wistar albino rats.**

Parameters	Group 1	Group 2	Group 3	Group 4	Group 5
Hb g/l	13.60 ± 1.856 <sup>a</sup>	11.90 ± 1.516 <sup>a</sup>	13.80 ± 1.640 <sup>a</sup>	13.86 ± 0.378 <sup>a</sup>	14.48 ± 0.598 <sup>a</sup>
PCV (%)	40.80 ± 5.630 <sup>a</sup>	35.80 ± 4.550 <sup>a</sup>	41.70 ± 4.876 <sup>a</sup>	41.60 ± 1.140 <sup>a</sup>	43.40 ± 1.817 <sup>a</sup>
RBC (x10 <sup>12</sup> /L)	6.20 ± 1.351 <sup>a</sup>	4.72 ± 1.071 <sup>a</sup>	5.86 ± 0.945 <sup>a</sup>	5.96 ± 0.241 <sup>a</sup>	6.50 ± 0.412 <sup>b</sup>
WBC (x10 <sup>9</sup> /L)	3.74 ± 0.397 <sup>a</sup>	3.740 ± 1.309 <sup>a</sup>	7.58 ± 0.608 <sup>b</sup>	9.04 ± 1.569 <sup>b</sup>	10.02 ± 4.013 <sup>b</sup>
MCHC (g/L)	32.60 ± 0.548 <sup>a</sup>	32.00 ± 0.707 <sup>a</sup>	32.80 ± 0.447 <sup>a</sup>	33.00 ± 0.000 <sup>a</sup>	33.00 ± 0.000 <sup>a</sup>
MCH (pg)	22.40 ± 2.702 <sup>a</sup>	25.40 ± 2.302 <sup>a</sup>	23.20 ± 1.095 <sup>a</sup>	23.80 ± 0.837 <sup>a</sup>	22.20 ± 0.837 <sup>a</sup>
MCV (fl)	67.00 ± 7.450 <sup>a</sup>	77.00 ± 7.280 <sup>a</sup>	69.80 ± 3.271 <sup>a</sup>	69.20 ± 5.070 <sup>b</sup>	66.60 ± 1.673 <sup>a</sup>
N	23.20 ± 4.147 <sup>a</sup>	30.00 ± 10.075 <sup>a</sup>	32.60 ± 3.975 <sup>a</sup>	43.00 ± 3.464 <sup>a</sup>	29.40 ± 5.639 <sup>a</sup>
L	76.80 ± 4.147 <sup>a</sup>	70.00 ± 10.075 <sup>a</sup>	67.40 ± 3.975 <sup>a</sup>	57.00 ± 3.464 <sup>a</sup>	70.60 ± 5.639 <sup>a</sup>

- Values are expressed as Mean ± Standard deviation
- Values with different superscripts show significant difference at p ≤ 0.05 level.
- Values with the same superscripts show no significant differences.

Hematological parameters are important health indices and are of diagnostic significance in routine clinical evaluation of the states of health (Patrick *et al.*, 2014). Table 2 shows results for WBC, Hb, RBC, PCV, MCHC, MCH, MCV, Neutrophils, and lymphocytes.

A significant increase at (P ≤ 0.05) was observed in the concentration of White blood cell level when compared to the control group. The increase in WBC concentration was peak at group 5 which received the high dose of *Ricinodendron heudelotii* aqueous seed extract, this could be attributed to the effect of the Saponins which is a phytochemical constituent and Saponin are reported to be suitable immunostimulator (James *et al.*, 1988), changes the immune response inducing Lymphocytes and Neutrophils (Ramanaviciene *et al.*, 2004). There was an increase in neutrophils compared to the control group which is beneficial because neutrophils are the major white blood cell that fight pathogens and this will confer further immunity to humans (Mayadas *et al.* 2014). However, this study showed decreasing lymphocyte status with increasing concentrations of *R. heudelotii*.

The RBC concentration in group 5 increased significantly at p ≤ 0.05. Increase in Red blood cells could consequently lead to an increase in Hemoglobin (Hb) concentration, hence, *Ricinodendron heudelotii* can therefore be a treatment in anemia since it boosts red blood cell status.

Packed Cell Volume (PCV) which is also called Hematocrit is the percentage (%) of red blood cell in blood. An increase in PCV level of groups 5 could be a sign of polycythemia vera. Polycythemia vera is a proliferative disorder in which the bone marrow produces excessive number of red blood cells. This could lead to increase in blood volume and a consequent increase in PCV.

There was a concentration dependent increase in group 4 and 5 in Mean Cell Volume MCV when compared to the control group. A concentration dependent decrease was observed in the Mean Cell Hemoglobin in group 5

compared to control group. MCH is the hemoglobin amount per red blood cell.

Table 2 shows that the aqueous extract of *Ricinodendron heudelotii* seed had some positive effect on the hematological status of the experimental rats.

Our study did not access the MCV (Mean Corpuscular Volume), MCHC (Mean Corpuscular Haemoglobin Concentration and MHCH (Mean Corpuscular Haemoglobin Concentration) which may suggest the existence of a pathology. Further studies should involve chronic administration of *Ricinodendron heudelotii* with the estimation of these red cell indices to know whether it causes significant pathology to the red blood cells with chronic use.

## CONCLUSION

The study revealed that administration of aqueous seed extract of *Ricinodendron heudelotii* did not have significant effect p ≤ 0.05 on the sodium, potassium, chloride and bicarbonate concentrations, however, a significant difference p ≤ 0.05 was observed in calcium level. It also suggests that the aqueous seed extract of *Ricinodendron heudelotii* has the potency to stimulate the production of leucocytes and can serve as an immune booster, It further showed that *Ricinodendron heudelotii* increases red blood cell levels and could be used in treating anemia. The study therefore recommends that *Ricinodendron heudelotii* should be consumed only in moderate doses to ensure normal regulation of blood levels.

## Conflict of interest

The authors hereby declare no conflict of interest.

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