Biochemical and Haematological Effects of Aqueous Leaf Extract of*Sida* cordifoliain *Plasmodium beighei* infected rat wistar albino rats

**Abstract**

Assessment of haematologicaland biochemical parameters can be predictive of the adverse effects resulting from the ingestion of a foreign substance.Freshly, harvested leaves of *Sidacordifolia* were washed and dried at room temperature, after which they were ground to powder and subsequently extracted. Twenty-five adult wistar rats were divided into five groups of five rats each. Group I was the normal control and was administered with 2 ml of distilled of water. Group II was infected without treatment, while Group III and IV were infected and afterwards administered with 200 and 400 mg/kg of aqueous extract of *Sida acuta* respectively.Group V was administered with the standard drug. The Packed Cell Volume reported for group II (negative) control was significantly ($P<0.05$) lower than that reported for the normal control. However, oral administration of 200 and 400 mg/kg of *Sidacordifolia* leaf extract significantly ($P<0.05$) increased it though to a level which was significantly ($P<0.05$) lower than that reported for the normal control. Similar observation was made on Haemoglobin Concentration (Hb) and Red Blood Cell(RBC). However, a contrary observation was made on the white blood cell. Urea and creatinine reported for the negative control were significantly ($P<0.05$) higher than those reported for the normal control. However, the aforementioned parameters were significantly ($P<0.05$) reduced following oral administration of the said extract. In conclusion, it can be deduced from this study that*Sidacordifolia* leaf extract has the ability to restore a distorted haematological and biochemical states resulting from *P. berghei* infection.

**Keywords:**Sida *cordifolia*, Blood, Cell, Haemoglobin, Leaf

**Introduction**

*Sidacordifolia* a notorious specie in the genus (Malvaceae) is commonly recognized for its therapeutic potentials[1]. It is commonly known in India as Bala and mallow country one of the ingredients used in making Ayurvedic formulations[2]. *Sidacordifolia* is reportedly used as antioxidant, anticancer, and antidiabetic [3]. Other activities of *Scordifolia*include analgesic and anti-inflammatory activities among others [3].

The wrongly heldimpression that plant based medicinal preparations wield minimal or no side effects has undermined approach to cautious application of this category of therapy in the

treatment of diverse human diseases and its attendant consequences [4].Medicinal plants being endowed with diverse arrays of biological compounds with great complexity tend to have several broad and actions on the physiological and biochemical systems [5].

Assessing the haematological parameters can be relied upon to diagnoseunpleasant effects resulting from the usage of foreign compounds on the blood constituents of an animal [6]. Furthermore, ingestion of chemical compounds at toxic doses can orchestrate alterations in blood parameters that are suggestive of hematological disorders [7]. The role of biochemical markers in performing accurate diagnosis as well as in the assessment of risk and adoption of therapy that improve clinical outcomes [8] cannot be overemphasized thereby informing the imperativeness of this study.



## Materials and Methods

### Collection of plant sample

Mature leaves of *Sidacordifolia*were harvested from a bush in Uturu, Isikwuato Local Government of Abia State. The leaves were conveyed in a dark polythene bag to the herbarium unit of the Department of Forestry, Michael Okpara University of Agriculture Umudike, Southeastern Nigeria.

### Experimental Animals

Twenty-five (25) adult mice were gotten from the animal House of the Department of Science Laboratory Technology, AkanuIbiam Federal Polytechnic, UwanaAfikpo. The animals were handled in accordance with ethics governing the handling and use of laboratory animals for

research purposes. They were kept in transparent plastic cages and were fed guinea grower feed for two weeks to acclimatize them before the experiment.

**Malaria parasite**

Four mice passaged with the parasite *Plasmodium berghei*used for this study were bought from the Department of parasitology, University of Nigeria Nsukka.

**Extraction**

Clean tap water was used to wash leaves of *Sidacordifolia*to get rid of dirt. The leaves were subsequently dried at room temperature after which they were ground into fine powder. Exactly 500 g of powdered plant sample was subsequently soaked in 3 L of distilled water for 24 h. The mixture was filtered with a clean sieve and was concentrated to dryness in a water bath for 3 days at $50^{o}$C.

**Acute Oral Toxicity Study**

The method of the Organization for Economic Cooperation and Development (OECD 425, 2008) was used to determine the acute toxicity of extract using limit test dose of 2 g/kg. Five apparently healthy mice were which had been denied food for 4 h were dosed and subsequently weighed to determine the dose. No mortality was recorded on the first animal administered a limit dose of 2000 mg/kg. The remaining four male mice were dosed and observed for signs of toxicity suchas absence of tremor, diarrhea, lethargy, and paralysis periodically for the first four and were later followed for 14 days for any lethality [9].

**Parasite passaging**

The approach of by Peter and Anatoli (1998) was relied upon to inoculate*Plasmodium berghei*(NK 65) into the mice by intraperitoneal route (Anatoli, 1998). Red blood cells infected with *Plasmodium berghei*was obtained from the tail vein of the infected mice and was subsequently diluted in 5 mL of phosphate buffered saline (PBS), so that 1 mL of parasitized blood contains 5 x 109 RBC m-1 infected erythrocytes, each 0.2 mL of the blood that was subsequently injected into an animal contained 1 x 106 *Plasmodium berghei*parasitized red cells (Huang et al., 2015). Administration of extract began three (3) days after inoculation.

**Biochemical analysis**

**Sample preparation:**Kidneyfunction test wasperformed by introducing exactly 2 mL of blood into the EDTA tube prior to centrifugation at 4,000 rpm for 15 min and the plasma obtained was stored for biochemical analysis.

**Serum Urea Determination**

Exactly 10 µL of sample was placed into a tube containing 1000 µL of the working reagent. The contents of the tube were thoroughly mixed, incubated for 5 minutes at 37$^{o}$C (Kaplan, 1982). Blood urea concentration was determined using the formular below:

$$Urea\ conc\ (mg/dl) = (A\ Sample)/(Acal/STD) \times conc.cal/STD(mg/dl)$$

**HaematologicalEvaluation**

Hematological parameters (Red Blood Cells, Haemoglobin concentration and packed cell volume and white blood cell) were determined with the aid of an automatic hematological analyzer (Coulter STKS, Beckman) (Yang et al., 2019).

**Data Analysis**

Data obtained were expressed as Mean ± Standard Deviation using SPSS (Ver. 23). Data were analysed using one way Analysis of Variance (ANOVA). Differences in mean were compared using Turkey Test. *p-values* less than 0.05 was considered statistically significant.



Table 1: Effect of Oral administration of Aqueous Extract of *Sidacordifolia*on the Haematological Indices of *Plasmodium berghei* Infected Rats

| TREATMENT | PCV | Hb (g/dl) | WBC ($\times10^9$/l) | RBC($\times10^6$/l) |
|---|---|---|---|---|
| Group I (Normal Ctrl) | $48.05\pm3.73^{d}$ | $18.09\pm2.42^{e}$ | $6.24\pm0.24^{b}$ | $7.20\pm2.59^{d}$ |
| Group II (Negative Ctrl) | $28.09\pm1.05^{a}$ | $8.40\pm1.98^{a}$ | $9.82\pm0.82^{e}$ | $3.70\pm3.05^{a}$ |
| Group III: INF+200 mg/kg Extract | $35.02\pm4.54^{b}$ | $12.09\pm2.33^{b}$ | $8.21\pm0.20^{d}$ | $4.20\pm3.22^{ab}$ |
| Group IV: INF+400 mg/kg Extract | $39.09\pm5.08^{c}$ | $13.07\pm1.82^{bc}$ | $7.08\pm2.31^{c}$ | $5.20\pm2.30^{c}$ |
| Group V: INF+STD | $47.08\pm3.42^{d}$ | $15.07\pm0.92^{d}$ | $4.70\pm5.30^{a}$ | $7.30\pm3.52^{d}$ |

Values are expressed as mean ± standard deviation of three determinations. Values with different superscript in a column are significantly ($p<0.05$) different

Table 2: Effect of Oral Administration of Aqueous Extract of *Sidacordifolia* on the Renal Health

| Treatment | Urea (mg/dl) | Creatinine (mg/dl) |
|---|---|---|
| Group I (Normal CTRL) | $14.40\pm5.87^{a}$ | $0.81\pm0.98^{a}$ |
| Group II (Negative CTRL) | $31.50\pm4.32^{c}$ | $3.10\pm0.19^{c}$ |
| Group III: INF+200 mg/kg Extract | $20.12\pm2.02^{b}$ | $1.02\pm0.32^{b}$ |
| Group IV: INF+400 mg/kg Extract | $19.14\pm0.34^{b}$ | $0.96\pm0.23^{c}$ |
| Group V: INF+STD (chloroquine) | $14.98\pm0.98^{a}$ | $0.89\pm0.23^{a}$ |

Values are expressed as mean ± standard deviation of three determinations. Values with different

superscript in a column are significantly ($p<0.05$) different

## RESULT AND DISCUSSION

Assessment of haematological parameters can be diagnostic of adverse effects of foreign compounds on blood constituents of animals[6]. Table 1 shows the effect of oral administration of aqueous extract of *Sidacordifolia* on the haematological indices of *Plasmodium berghei* infected rats indicating that the packed cell volume (PCV) reported for the negative control was significantly ($p<0.05$) was lower than that reported for the normal control. However, oral administration of aqueous extract of*S.cordifolia* significantly ($p<0.05$) increased it in a dose dependent manner. It was also observed that there was no there was no significant ($P>0.05$) difference in the PCV reported for the normal control and the standard control.The haemoglobin concentration (Hb) reported for the negative control was significantly ($p<0.05$) lower than that reported for other groups. However, following oral administration of extract, there was a significant increase in the Hb reported for groups II and III which though were not significantly ($p>0.05$) different from each other but lower than that reported for the normal control. The red blood cell (RBC) reported for the negative control was significantly ($p<0.05$) lower than those reported for other groups. The RBC reported for group III was not significantly ($p>0.05$) different from that reported for the negative control. Similar observation was made between the normal control and the standard control. The anti-anemic effect of *S.cordifolia* extract could be attributed to antioxidant activity of the plant extract which had been reported in previous study.This is consistent with the finding of Ukpanukpong et al. [12] which showed that ethanolic leaf extract of S.*rhobofolia*, a member of the *malvaceae* family to which *S. cordifolia* belongs orally administered on rats induced with artificial infertility significantly ($P<0.05$) increased the

red blood cell. The White Blood Cell (WBC) reported for the negative control was significantly ($p<0.05$) higher than that reported for the normal control which in turn was significantly ($p<0.05$) lower than those reported for groups III and IV which were infected and treated with extract. This is contrary to the finding made byUkpanukpong et al. [12] which showed that oral administration of 200 and 400 mg/kg of*S. rhombifolia* extract significantly increased the white blood cell in rats induced with artificial infertility. Biochemical markers play an important role in accurate diagnosis and also for assessing risk and adopting therapy that improve clinical outcome [8]. Furthermore, creatinine is commonly used as a measure of kidney function [13]. In addition, the most frequently determined clinical indices for estimating renal function depends upon the concentration of urea in the serum [14].The kidney is one the organs that receive the bulk of the toxic impacts of toxic substances. Table 2 shows the effect of oral administration of aqueous extract of *Sidacordifolia* on the renal health of rats infected with *Plasmodium berghei* indicating that serum creatinine and urea were significantly ($p<0.05$) higher in the negative control group which were infected without treatment than those reported for the normal control group. These were significantly ($p<0.05$) reduced in a dose dependent manner.This is consistent with the finding of Ukpanukpong et al. (2019) which showed that ethanolic leaf extract of *Sidacordifolia* was not toxic.

## Conclusions

It has been reveled through this study that aqueous leaf extract of *Sidacordifolia*wield the potential to reverse distorted haematological and biochemical status of rats previously infected with *P. berghei*.

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