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Effect of Leaf Extract of *Senna alata* on Biochemical Indices of Wistar Rats Infected with *Trypanosoma brucei brucei*

M. A. Kugama[1*], T. Tese[2], H. Sabo[1], T. Andrew[2], Y. A. Onaolapo[2], O. G. Allen[2], A. Agbo[2], D. W. Danjuma[2], A. M. Usman[3], L. M. Umar[2], and P. J. Baji[2]

[1]Department of Human African Trypanosomiasis Research, Nigerian Institute for Trypanosomiasis Research, Kaduna, Nigeria.
[2]Department of Vector and Parasitological Studies Research, Nigerian Institute for Trypanosomiasis Research, Kaduna, Nigeria.
[3]Kaduna State Ministry of Health, Nigeria.

Authors' contributions

This work was carried out in collaboration among all authors. Authors MAK and TT designed the study. Authors TA, MAK and HS wrote the protocol and wrote the first draft of the manuscript. Authors TT and MAK managed the analyses of the study. Authors AMU, PJB, LMU, DWD, YAO and MAK carried out the laboratory analysis. Authors OGA, AA, TT and MAK managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

This study was aimed at determining the effect of leaves of *Senna alata* extract on biochemical indices of Wistar rats infected with *Trypanosoma brucei brucei*. Phytochemical screening revealed the absence of steroids in all extracts, absence of saponins in chloroform extracts and the presence of free anthraquinones only in chloroform extract. Post-infection treatment of animals stirred the emergence of parasitaemia by Day 3. Only animals receiving 200 mg/kg b.wt. of chloroform extract

**Corresponding author: E-mail: meshackkugama@gmail.com;*

survived by day 16. A significant (P<0.05) decrease in ALT for groups receiving methanol (400 mg/kg b.wt.), chloroform and aqueous extracts and significant (P<0.05) increase in unconjugated bilirubin in the group receiving methanol extract (200 mg/kg b.wt.) compared to infected not treated rats. Significant (P<0.05) decrease in potassium concentration in groups receiving methanol and chloroform, and a significant (P<0.05) increase in sodium concentration in the group receiving 400 mg/kg b.wt. of aqueous extract compared to the infected not treated rats. These results thereby demonstrate the ameliorative potential of *Senna alata* leaves against *T. brucei brucei*.

Keywords: Trypanosomiasis; Senna alata; biochemical indices; parasitaemia; phytochemical.

1. INTRODUCTION

Plants are known to provide a rich source of raw materials in African traditional medicine and other parts of the developing world [1]. Reports on the ethnobotanical records indicate a consensus on the use of antimicrobial active medicinal plants in the development of cheaper drugs [2]. Medicinal plants have long drawn the history of use by human beings for the cure of various ailments. More than 70% of the third world population now depends on the traditional medicinal system, otherwise known as a complementary or alternative system of medicine [3]. Also, studies have shown that some medicinal plants have antitrypanosomal activity [4].

Trypanosomiasis is a vector-borne disease in vertebrates caused by a parasitic protozoan of the genus *Trypanosoma* [5]. The parasites are transmitted by the bite of Tse-tse fly of the genus *Glossina*. The flies are found in vegetation along with water courses and lakes, forest edges and gallery forests, extending to vast areas of guinea savannah. The parasite lives extracellularly in the blood and tissue fluids in the mammalian host. The most important species responsible for the disease complex known as African animal trypanosomiasis include *Trypanosoma brucei*, *T. congolense* and *T. vivax*. The Human African Trypanosomiasis is caused by *T. gambiense* and *T. rhodesiense* [5,6]. In Africa, the wide occurrence of African trypanosomiasis in people and their livestock is a great constraint to development. Currently, chemotherapy and vector control strategies are principally used in the management of African trypanosomiasis. However, these methods are faced with problems ranging from imperfect diagnostic techniques [7,8], inadequate drug treatment, resistance to current trypanocides [9], high prevalence of drug resistance even in animal trypanosomes without a history of drug exposure [10], and ineffective vector control strategies [11].

Senna alata which is commonly called candle bush plant is an important medicinal plant which contains antimicrobial activity [12,13]. The leaf extract has been reported to be useful in the treatment of heart failure, gonorrhea, abdominal pains, and oedema. Leaf extracts of *S. alata* contain glycosides, saponins, flavonoids, and tannins [13]. This study is aimed at investigating the effect of *S. alata* leaves extract on the biochemical indices of wistar rats infected with *Trypanosoma brucei brucei.*

2. MATERIALS AND METHODS

2.1 Collection and Authentication of Plant Materials

The plant *S. atata* was harvested in the morning from Sakaru village, Jos Road, Zaria Kaduna State. It was authenticated by a botanist in the herbarium, Department of Biological Sciences, Ahmadu Bello University, Zaria and given a voucher number 906. The leaves were dried at 28ºC (room temperature), pulverized in a mortar and sieved to obtain a fine powder and then stored in an airtight container with proper labelling for future use.

2.2 Plant Extraction

One thousand grams (1000 g of powdered) plant material was weighed and extraction was done in light petroleum ether (60 – 80ºC). The petroleum ether was concentrated *in vacuo*, to obtain a product referred to as petroleum ether extract (PEE). The marc (petroleum ether defatted residue) was extracted with ninety-five per cent (95%) methanol and the solvent recovered *in vacuo*, to obtain crude methanol extract (CME). One hundred gram (100 g) of the CME was dissolved in water and partitioned four times with an equal portion of chloroform in one thousand meals (1000 ml) separatory funnel. The chloroform portion was pooled and concentrated *in vacuo*, to obtain a product referred to as the chloroform extract (CE).

2.3 Experimental Animals

In this study, twenty-seven wistar rats (*Rattus rattus novergicus*) were purchased from the animal house of Nigerian Institute for Trypanosomiasis Research, Kaduna. The animals were housed in standard rat cages, maintained on standard pellet diet from Vital Plc, Jos, Nigeria and water *ad libitum*.

2.4 Infection of Animals

10^4 parasites per meal were introduced intraperitoneally into rats in 0.1-0.2 ml blood/PBS solution. The number of parasite/ml was estimated using the method of Herbert and Lumsden (1976).

2.5 Phytochemical Screening of Crude Extract of *S. alata*

Method of Sofowora [1], as described in the work by Ayoola [14] was used to detect the presence of anthraquinones, flavonoides, alkaloids, saponin, glycosides, tannins, and steroids.

2.6 Determination of Biochemical Indices

Alanine Aminotransferase (ALT) and Aspartate Aminotransferase (AST): Activities of Aspartate Aminotransferase (AST) and Alanine Aminotransferase (ALT) were assayed for by the method of Reitman and Frankel [15] using Randox assay kits.

Alanine Aminotransferase *(*ALT*)*: Procedure: Exactly, 0.5 ml of reagent 1 which is made up of phosphate buffer, L-alanine and α-Oxoglutarate was added into two clean test tubes, one containing 0.1 ml of serum and the other containing 0.1 ml distilled water (blank). The content in each test tube was mixed, incubated for exactly 30 minutes at 37ºC. 0.5 ml of reagent 2 which is made up of 2, 4-dinitrophenylhydrazine was added to each of the test tubes, mixed and allowed to stand for exactly twenty minutes at 20-25ºC. Then 0.5 ml of sodium hydroxide solution was added to each of the test tubes, the content in each of the test tubes was mixed and absorbance was read against the blank at 540 nm after 5 minutes. The ALT activity (U/l) was determined from the standard calibration table provided in the manual of Randox Lab. Ltd, UK Reagent Kit.

Aspartate Aminotransferase (AST): Procedure: 0.5 ml of reagent 1 which is made up of phosphate buffer, L-aspartate and α-oxoglutarate was added into two clean test tubes labelled as test sample and reagent blank, containing 0.1 ml of serum and 0.1 ml of distilled water, the content in each test tubes was mixed and incubated for exactly thirty minutes at 37ºC. 0.5 ml of reagent 2 which is made up of 2,4-dinitrophenylhydrazine was added to each of the test tubes, the content of each of the test tubes was mixed and allowed to stand for exactly twenty minutes at 20-25ºC. To each of the test tubes, 0.5 ml of sodium hydroxide solution was added, mixed and absorbance was read against the blank at 540 nm after five minutes. The AST activity (U/l) was determined from the standard calibration table provided in the manual of Randox Lab. Ltd, UK Reagent Kit.

Alkaline Phosphatase (ALP): The serum level of alkaline phosphatase was quantified by an optimized standard method described by Haussament, [16] using Randox assay kits.

Procedure: Exactly 1 ml of reagent 1 containing Diethanolamine buffer, magnesium chloride and substrate (p-nitrophenylphosphate) was added into a clean test tube containing 0.02 ml of serum. This was mixed, initial absorbance was read and the timer was set simultaneously, absorbance was read again after 1, 2, and 3 min at 405 nm.

Determination of serum Total Bilirubin (TB) concentration: The serum TB concentration was determined using the Randox Kit (Randox laboratories limited UK) based on the method described by Jendrassik and Grof, [17] and Sherlock [18].

Determination of serum unconjugated bilirubin concentration: The serum unconjugated bilirubin concentration was determined using the Randox Kit (Randox laboratories limited UK) based on the method described by Jendrassik and Grof [17] and Sherlock [18].

Procedure: Two hundred meals (200 ml) of reagent 1 (Sulphanillic acid) was dispensed each into two different test tubes labelled ‘sample blank' and ‘sample' followed by the addition of one drop (50 µl) of reagent 2 (nitrite) and two thousand µl of 0.9% NaCl. Two hundred microlitres (200 µl) of the test serum was dispensed into each of the test tubes and the mixtures incubated in a water bath for ten minutes at 25ºC. The absorbance of the sample (A_{TB}) was then read against the sample blank

using a colourimeter at 546 nm wavelength. The direct bilirubin concentration was then calculated using the formula.

Unconjugated bilirubin (mg/dl) = 14.4 × A_{UB} (546 nm)

Where A_{UB} = absorbance of unconjugated bilirubin

2.7 Determination of Serum Creatinine Concentration

The colourimetric method was used to determine serum creatinine concentration using Randox assay kits [19].

Procedure: One meal (1 ml) of working reagent containing picric acid and sodium hydroxide was added into two clean test tubes labelled sample test and standard, containing 0.1 ml of sample and 0.1 ml of the standard solution. The content in each test tube was mixed and after thirty seconds, the absorbance A_1 of the standard and sample were read. Two minutes later, absorbance A_2 of the standard and sample were read at 490 nm.

2.8 Determination of Electrolyte Concentration in Serum

Determination of sodium, potassium and chloride ions.

The concentrations of sodium, chloride, and potassium ions were determined using Elyte kits [20].

Sodium

Procedure

Precipitation

The following were Pipetted into a clean dry test tube labelled as Standard (S) and Test (T)

Addition Sequence	S (ml)	T (ml)
Precipitating Reagent (L1)	1.0	1.0
Na+/K+ Standard (S)	0.02	-
Sample	-	0.02

The solutions were mixed well and left to stand at room temperature for 5 minutes, shaking well intermittently. The solution was centrifuged at 2500 rpm to obtain a clear supernatant.

Colour development: The following were pipetted into clean dry test tubes labelled as Blank (B), Standard (S), and Test (T)

Addition Sequence	B (ml)	S (ml)	T (ml)
Acid Reagent (L2)	1.0	1.0	1.0
Supernatant from Step1.	-	0.02	0.02
Precipitating Reagent (L1)	0.02	-	-
Colour Reagent (L3)	0.1	0.1	0.1

The solution was mixed well and incubated at room temperature for 5 min. The absorbance of the Blank (Abs.B)

Standard (Abs.S) and Test Sample (Abs.T) were measured, against distilled water within 15 min at 530 nm.

Calculation

Sodium concentration (mmol/l) = $\frac{Abs.B-Abs.T}{Abs.B-Abs.S} X\ 150$

Potassium

Procedure

The following were pipetted into clean dry test tubes labelled as Blank (B), Standard (S), and Test (T) as follows

Addition Sequence	B (ml)	S (ml)	T (ml)
Potassium Reagent (L1)	1.0	1.0	1.0
Deionised water	0.02	-	-
Na+/K+ Standard (S)	-	0.02	-
Sample	-	-	0.02

The solutions were mixed well and incubated at room temperature for 5 min. The absorbance of the Standard (Abs.S), and Test Sample (Abs.T) were measured against Blank, at 630nm, within 15 min.

Calculation

Potassium concentration (mmol/l) = $\frac{Abs.T}{Abs.S}\ X\ 5$

Chloride

Procedure

The following were pipetted into clean dry test tubes labelled as Blank (B), Standard (S), and Test (T).

Addition Sequence	B (ml)	S (ml)	T (ml)
Chloride Reagent (L1)	1.0	1.0	1.0
Deionised water	0.01	-	-
Chloride Standard (S)	-	0.01	-
Sample	-	-	0.01

The solution was mixed well and incubated at R.T. for 2 minutes. The absorbance of the Standard (Abs.S) and Test Sample (Abs.T) were measured against Blank, within 60 min at 505 nm.

Calculations

Chloride concentration (mmol/l) = $\frac{Abs.T}{Abs.S}\ X\ 100$

Determination of Bicarbonate concentration

2.9 Therapeutic Monitoring

Parasitaemia of each rat was monitored every forty-eight hours during the administration of the extracts and reference drug (diaminazene Diaceturate) and the animals were anaesthetized and sacrificed to obtain blood by cardiac puncture at high parasitaemia.

2.10 Statistical Analysis

Values of the data obtained from the study were summarized and expressed as mean ± standard deviation. Data analysis was performed using Statistical Package for Social Science (SPSS), version 20.0. To compare the results obtained from different groups, one way ANOVA followed by Duncan multiple comparison tests were performed to determine statistical significance. P values less than 0.05 were considered significant.

3. RESULTS

3.1 Phytochemical Analysis of crude Extract of *S. alata* Leaves

The qualitative phytochemical constituents of extracts of *S. alata* leaves indicate that flavonoids were absent only in petroleum ether extract while saponins were absent in chloroform extracts. Carbohydrates, cardiac glycosides, triterpenes, and alkaloids were present in all the extracts, Anthraquinones were present only in chloroform extract while steroids were absent in all the extracts (Table 1).

3.2 Effects on Liver and Kidney Function Indices

The effects of extracts of *S. alata* in *T. brucei brucei* infected rats are presented in Tables 2 to 4. Administration of methanol extract recorded a significant increase ($p<0.05$) in AST, ALT, Unconjugated bilirubin levels and no significant difference ($p>0.05$) in ALP, Total bilirubin and creatinine compared to the normal control. Compared to the induced not treated group, a significant increase ($p<0.05$) was recorded in Unconjugated bilirubin for groups that received 200 mg/kg b.wgt, no significant different ($p>0.05$) in levels of AST, ALT for groups that received 200 mg/kg b.wgt, ALP, Total bilirubin, Creatinine, and a significant decrease ($p<0.05$) in ALT for group that received 400 mg/kg b.wgt was also recorded (Table 2). When chloroform extract was administered, in the infected groups treated with extract, a significant increase ($p<0.05$) was recorded in levels of ALT and no significant difference ($p>0.05$) was obtained for levels of AST, ALP, Total bilirubin, Unconjugated bilirubin, and Creatinine compared to the normal control. However, a significant ($p<0.05$) decrease in ALT and no significant change ($p>0.05$) in AST, ALP, Total bilirubin, unconjugated bilirubin, and creatinine compared to the infected not treated group (Table 3). Effects of aqueous extract did not generate significant difference ($p>0.05$) in AST, ALP, total bilirubin, and creatinine in groups that received 200 mg/kg b.wgt and a significant increase ($p<0.05$) in ALT, unconjugated bilirubin, and creatinine in groups that received 400 mg/kg b.wgt compared to the normal control group. Compared to the induced not treated group, a

Table 1. Phytochemical constituents of extracts of *Senna alata* leaves

Phytochemical constituent	Meth.	Chloro.	Aq.
Carbohydrates	+	+	+
Cardiac glycosides	+	+	+
Steroids	_	_	_
Triterpenes	+	+	+
Flavonoids	+	+	+
Saponins	+	_	+
Alkaloids	+	+	+
Anthraquinones	_	+	_

KEY: + = presence of component; - = absence of component

Table 2. Effects of methanol extract of *S. alata* leaves on some liver and kidney function markers in *T. brucei brucei* infected albino rats

Group/ Dose of extract	AST	ALT	ALP	Total bilirubin	Unconjugated bilirubin	Creatinine
NC	49.58 ± 5.66^{a}	26.07 ± 7.07^{a}	59.07 ± 7.07^{a}	42.21 ± 1.41^{a}	19.49 ± 9.90^{a}	0.54 ± 0.28^{a}
DC	98.78 ± 18.38^{c}	103.28 ± 8.49^{c}	96.46 ± 4.24^{b}	47.43 ± 2.83^{a}	35.85 ± 5.66^{ab}	1.44 ± 0.28^{a}
200 mg/kg	8128 ± 8.49^{bc}	91.53 ± 2.12^{bc}	78.63 ± 24.04^{ab}	47.49 ± 9.90^{a}	60.64 ± 4.24^{c}	0.70 ± 0.57^{a}
400 mg/kg	92.92 ± 12.73^{bc}	79.07 ± 7.07^{b}	91.58 ± 5.66^{ab}	47.70 ± 7.07^{a}	51.71 ± 11.31^{bc}	0.84 ± 0.28^{a}
SD	56.49 ± 9.90^{ab}	31.94 ± 9.90^{a}	72.94 ± 9.90^{ab}	47.55 ± 4.95^{a}	21.64 ± 4.24^{a}	0.66 ± 0.42^{a}

Values are mean ± SD, n=3. Values with different superscripts down the column are significantly (P<0.05) different. NC = Normal Control. DC: Disease control; SD: Standard drug (3.5 mg/kg of diaminazane aceturate); AST: Aspartate Aminotransferase; ALT: Alanine aminotransferase, ALP: Alkaline phosphatase; Total bilirubin: Total Bilirubin; Unconjugated bilirubin: Unconjugated bilirubin; Creatinine: Creatinine

Table 3. Effects of chloroform extract of *S. alata* leaves on some liver and kidney function markers in *T. brucei brucei* infected albino rats

Group/ Dose of extract	AST	ALT	ALP	Total bilirubin	Unconjugated bilirubin	Creatinine
NC	49.85 ± 5.66^{a}	26.07 ± 7.07^{a}	59.07 ± 7.07^{a}	42.21 ± 1.14^{a}	19.49 ± 9.90^{a}	0.54 ± 0.28^{a}
DC	98.78 ± 18.38^{b}	103.28 ± 8.49^{c}	96.64 ± 4.24^{a}	47.43 ± 2.83^{a}	35.85 ± 5.66^{a}	1.44 ± 0.28^{b}
200 mg/kg	76.29 ± 12.73^{ab}	71.71 ± 11.31^{b}	71.46 ± 4.24^{a}	47.28 ± 8.49^{a}	21.43 ± 2.83^{a}	0.86 ± 0.42^{ab}
400 mg/kg	62.92 ± 12.73^{a}	65.49 ± 9.90^{b}	74.27 ± 28.28^{a}	46.85 ± 5.66^{a}	32.35 ± 15.56^{a}	0.82 ± 0.14^{ab}
SD	56.49 ± 9.90^{a}	31.94 ± 9.90^{a}	72.49 ± 9.90^{a}	47.57 ± 4.95^{a}	21.64 ± 4.24^{a}	0.66 ± 0.42^{ab}

Values are mean ± SD, n=3. Values with different superscripts down the column are significantly (P<0.05) different. NC = Normal Control. DC: Disease control; SD: Standard drug (3.5 mg/kg of diaminazane aceturate); AST: Aspartate Aminotransferase; ALT: Alanine aminotransferase, ALP: Alkaline phosphatase; Total bilirubin: Total Bilirubin; Unconjugated bilirubin: Unconjugated bilirubin; Creatinine: Creatinine

Table 4. Effects of aqueous extract of *S. alata* leaves on some liver and kidney function markers in *T. brucei brucei* infected albino rats

Group/ Dose of extract	AST	ALT	ALP	Total bilirubin	Unconjugated bilirubin	Creatinine
NC	49.58 ± 5.66^{a}	26.07 ± 7.07^{a}	59.07 ± 7.07^{a}	42.21 ± 1.41^{a}	19.49 ± 9.89^{a}	0.54 ± 0.28^{a}
DC	98.078 ± 18.38^{b}	103.28 ± 8.48^{c}	96.64 ± 4.24^{b}	47.43 ± 2.83^{a}	35.085 ± 5.66^{ab}	1.44 ± 0.28^{b}
200 mg/kg	62.02 ± 21.21^{ab}	74.64 ± 4.24^{b}	79.84 ± 25.46^{ab}	49.35 ± 15.56^{a}	44.71 ± 11.31^{b}	0.55 ± 0.21^{a}
400 mg/kg	65.85 ± 5.66^{ab}	62.28 ± 8.48^{b}	84.28 ± 8.49^{ab}	45.58 ± 5.66^{a}	51.28 ± 8.49^{b}	0.75 ± 0.21^{b}
SD	56.49 ± 9.90^{a}	31.49 ± 9.90^{a}	72.49 ± 9.90^{ab}	47.54 ± 4.95^{a}	21.64 ± 4.24^{a}	0.60 ± 0.42^{a}

Values are mean ± SD, n=3. Values with different superscripts down the column are significantly (P<0.05) different. NC = Normal Control. DC: Disease control; SD: Standard drug (3.5 mg/kg of diaminazane aceturate); AST: Aspartate Aminotransferase; ALT: Alanine aminotransferase, ALP: Alkaline phosphatase; Total bilirubin: Total Bilirubin; Unconjugated bilirubin: Unconjugated bilirubin; Creatinine: Creatinine

significant decrease was recorded for ALT and creatinine receiving 200 mg/kg b.wgt while the other extract-treated groups recorded no significant difference (p>0.05) (Table 4).

3.3 Effects on Some Serum Electrolytes

The effects of extracts of *S. alata* in *T. brucei brucei* infected rats are presented in Tables 5 to 7. Administration of methanol extract, the results indicated no significant difference (p>0.05) in sodium, potassium, chloride, and bicarbonate ions compared to the normal control. When the infected rats treated with extracts were compared to the infected not treated rats, a significant decrease (p<0.05) in potassium ion concentration and no significant difference in sodium, chloride, and bicarbonate ions were recorded (Table 5). Administration of chloroform extract did not cause significant difference (p>0.05) in levels of sodium, potassium, chloride, and bicarbonates in infected rats treated with extract compared to the normal control. However, there was a significant decrease (p<0.05) in potassium compared to the infected not treated rats (Table 6). After the aqueous extract was given, there was no significant difference (p>0.05) in sodium, chloride, and bicarbonates compared to the normal control and the infected but not treated group. Though administration of extract generated a non-significant difference (p>0.05) in potassium concentration compared to the normal control, there was a significant decrease (p<0.05) in potassium ion concentration compared to the infected but not treated group (Table 7).

Table 5. Effects of Methanol Extract of *S. alata* leaves on Some Serum Ions in *T. brucei brucei* Infected Albino Rats

Group/ Dose of extract	Na	K	Cl	B.Car
NC	144.42 ± 22.63^{b}	6.10 ± 3.25^{a}	103.87 ± 18.38^{a}	22.70 ± .07^{a}
DC	100.92 ± 12.73^{a}	28.14 ± 6.22^{b}	92.94 ± 9.90^{a}	29.58 ± 5.66^{a}
200 mg/kg	129.07 ± 7.07^{ab}	13.97 ± 5.94^{a}	98.92 ± 12.72^{a}	22.21 ± 1.41^{a}
400 mg/kg	136.71 ± 11.31^{ab}	15.28 ± 3.96^{a}	94.49 ± 9.90^{a}	27.49 ± 9.90^{a}
SD	132.35 ± 15.56^{ab}	11.32 ± 2.12^{a}	105.14 ± 14.14^{a}	21.21 ± 1.41^{a}

Values are mean ± SD, n=3. Values with different superscripts down the column are significantly (P<0.05) different. NC = Normal Control. DC: Disease control; SD: Standard drug (3.5 mg/kg of diaminazane aceturate); Na: Sodium; K: Potassium; Cl: Chloride; B.Car: Bicarbonate

Table 6. Effects of Chloroform Extract of *S. alata* leaves on Some Serum Ions in *T. brucei brucei* Infected Albino Rats

Group/ Dose of extract	Na	K	Cl	B.Car
NC	144.42 ± 22.63^{a}	6.19 ± 3.28^{a}	103.78 ± 18.38^{a}	22.17 ± 7.07^{a}
DC	100.92 ± 12.73^{a}	28.14 ± 6.22^{b}	92.49 ± 9.90^{a}	29.85 ± 5.66^{a}
200 mg/kg	139.35 ± 15.56^{a}	6.08 ± 3.96^{a}	102.52 ± 15.85^{a}	26.07 ± 7.07^{a}
400 mg/kg	139.20 ± 21.21^{a}	4.93 ± 0.85^{a}	109.92 ± 12.73^{a}	30.14 ± 14.14^{a}
SD	132.35 ± 15.56^{a}	11.33 ± 2.12^{a}	105.14 ± 14.14^{a}	21.21 ± 1.41^{a}

Values are mean ± SD, n=3. Values with different superscripts down the column are significantly (P<0.05) different. NC = Normal Control. DC: Disease control; SD: Standard drug (3.5 mg/kg of diaminazane aceturate); Na: Sodium; K: Potassium; Cl: Chloride; B.Car: Bicarbonate

Table 7. Effects of Aqueous Ether Extract of *S. alata* leaves on Some Serum Ions in *T. brucei brucei* Infected Albino Rats

Group/ Dose of extract	Na	K	Cl	B.Car
NC	144.42 ± 22.63^{b}	6.10 ± 3.25^{a}	103.78 ± 18.38^{a}	22.07 ± 7.07^{a}
DC	100.92 ± 12.73^{a}	28.10 ± 6.33^{b}	92.49 ± 9.90^{a}	29.85 ± 5.66^{a}
200 mg/kg	140.35 ± 15.56^{ab}	7.90 ± 0.85^{a}	101.14 ± 14.14^{a}	24.85 ± 5.66^{a}
400 mg/kg	145.92 ± 12.73^{b}	6.60 ± 1.70^{a}	100.92 ± 12.73^{a}	24.07 ± 7.07^{a}
SD	132.35 ± 15.56^{ab}	11.30 ± 2.12^{a}	105.14 ± 14.14^{a}	21.21 ± 1.41^{a}

Values are mean ± SD, n=3. Values with different superscripts down the column are significantly (P<0.05) different. NC = Normal Control. DC: Disease control; SD: Standard drug (3.5 mg/kg of diaminazane aceturate); Na: Sodium; K: Potassium; Cl: Chloride; B.Car: Bicarbonate

4. DISCUSSION

The variety of phytochemicals present in the methanol, chloroform and aqueous extracts of *S. alata* could account for its medicinal values. This is in line with the findings of Akinyemi [21] who revealed that there are many different anti-nutrient elements like alkaloids, anthraquinones, saponins, tannins, terpenes, steroids, flavonoids, and carbohydrates found in different parts of *S. alata*. Different solvents have different solubility capacities for different phytoconstituents, hence the differences in the activities of the various extracts [22]. This accounts for the absence of some phytochemicals in some extracts and presence in others. Previous phytochemical studies carried out by Sule [23] indicated the presence of alkaloids, anthraquinones, carbohydrates, flavonoids, saponins, tannins, terpenes, and steroids in the crude extract of *S. alata* L. stem bark. It has been observed that antimicrobial activity of the plants is associated with the presence of some chemical components such as phenols, tannins, saponins, alkaloids, steroids, flavonoids, and carbohydrates. The phytochemical screening revealed the presence of anthraquinones in chloroform extract only.

The liver and kidney are two important organs that perform vital function for the healthy survival of the body. The liver detoxifies harmful substances, secretes bile into the intestine, synthesizes and store important molecules. The kidney helps in maintaining homeostasis of the body by reabsorbing important material and excreting waste products [24]. The measurement of these enzymes and other biochemical markers such as total bilirubin in serum constitutes the liver function tests [25]. Enzymes appear frequently in blood as a result of cellular injury. The most widely used enzymes to assess hepatocellular injury are the aminotransferase (ALT and AST) whereas alkaline phosphatases are useful in the diagnosis of hepatobiliary or cholestatic obstruction [26]. AST is not specific for the liver only but is also located in other organs like the heart, brain, kidney and skeletal muscle. ALT is a liver-specific enzyme for diagnostic use; when the integrity of the hepatocellular membrane is compromised, there is extrusion of the enzyme into the plasma [27]. Because of their presence in the blood, these invading parasites produce numerous changes in the cellular and biochemical constituents of blood [28,29].

A significant ($P<0.05$) increase in ALT, AST, and ALP in the infected not treated rats compared to the normal control is an indication of hepatocellular injury. It has been revealed that hepatic abnormalities such as elevations of transaminases and alkaline phosphatase (ALP) are common in diabetes mellitus [30,31,32]. The rise in levels of ALT is always accompanied by an elevation in the level of AST, which play a role in the conversion of amino acid to keto acid. Both AST and ALT are an excellent marker of liver damage caused by exposure to toxic substances [33]. Administration of extract caused a significant reduction in ALT, AST, and ALP of all induced treated groups compared to the normal control.

Creatinine is synthesized in the liver, passes into the circulation and is taken up almost entirely by skeletal muscle. Bilirubin is transported to the liver bound to albumin. High plasma conjugated bilirubin concentration indicates impaired hepatic excretory function [34].

T. brucei brucei infection has been shown to cause a perturbation in the electrolyte homeostasis [35]. The elevated serum potassium level after infection may be due to the damaging effect of *T. brucei* on the kidney and the heart myocardium. Our results indicated that *T. brucei* infection generated increased serum potassium which has been reported in myocardial ischemia [36] and renal damage [37] could cause a reduction in the resting membrane potential and quicken depolarization [38].

5. CONCLUSION

The result indicates that crude extracts of *S. alata* contain a variety of phytochemicals which have documented antimicrobial effects. The extract has demonstrated ameliorative in vivo effects on some biochemical indices in wistar rats with chloroform extract being the most potent ahead of aqueous and methanol extracts.

ETHICAL APPROVAL

The ethical clearance for the use of laboratory animals was obtained from the Animal Committee of Kaduna State Ministry of Agriculture and Forestry.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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