COMPARATIVE ANTI-MALARIA POTENTIALS OF LEAF AND ROOT EXTRACTS OF *ALSTONIA BOONEI* ON THE HAEMATOLOGICAL INDICES OF MICE INFECTED WITH *PLASMODIUM BERGHEI*

ONYISHI, Grace Chinenye, UGWU, Onyinye Costecia and NWOSU, Chigozie Godwin
Department of Zoology and Environmental Biology, University of Nigeria, Nsukka, Enugu State, Nigeria.

Corresponding Author: Nwosu, C. G. Department of Zoology and Environmental Biology, University of Nigeria, Nsukka, Enugu State, Nigeria. **Email:** gozzynwosu@gmail.com **Phone:** +234 8034431548

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ABSTRACT

The present study assessed the anti-malaria potentials and effects of aqueous and ethanolic leaf and root extracts of Alstonia boonei on the haematological indices of mice infected with Plasmodium berghei. The aim was to comparatively evaluate the anti-Plasmodium effects of the plant extracts in the infected mice against the standard drug. A total of 216 male albino mice were randomly assigned to six treatment groups each containing six mice for both aqueous and ethanolic extracts experimentation. The artesunate-sensitive strain of the rodent parasite P. berghei NK-65 was inoculated into the mice intraperitoneally and infection was established by both clinical and microscopic examination. Administration of the aqueous and ethanolic leaf and root extracts of A. boonei was done after phytochemical and acute toxicity tests at varying concentrations, for both suppressive and curative tests. Blood samples were collected by ocular puncturing and were examined for the haematological indices of PCV, Hb, RBC, MCHC, MCV, WBC, MHC, lymphocyte, neutrophil, platelets, eosinophil, and monocyte using the standard procedures for estimation. Results showed a comparable suppressive ability of A. boonei extracts to the standard drug, as well as a significantly ($p<0.05$) higher after treatment recovery of mice than the untreated infected mice. The haematological indices examined showed significantly ($p<0.05$) normalized and increased concentration after 7 days post-infection. In conclusion, the extracts of A. boonei leaf and root anti-Plasmodium activity was dependent on both dosage and duration as observed from the study, and have demonstrated satisfactory normalization efficacy to haematological indices in malaria treatment.

Keywords: Comparative anti-malaria, *Alstonia boonei*, Haematological, *Plasmodium berghei*, Mice

INTRODUCTION

Malaria is a disease caused by a haemoparasitic protozoon of the genus *Plasmodium*. It infects the red blood cells and considered the most significant public health problem in Nigeria (Gbadamosi *et al*., 2011). In 2012, there were 627, 000 malaria deaths worldwide, 90 % of which was in the African region followed by Southeast Africa (7 %) and 3 % in the Eastern Mediterranean (Sinha *et al*., 2014).

The disease results in loss of life, loss of productivity due to illness and premature deaths, and hinders children in their academic and social development through absence from school and permanent neurological or other damage associated with severe episodes of malaria (Kazembe *et al*., 2012). Haematological changes are measured as variations in the levels of blood cells and are used to access the pathological and physiological state of animals (Saxena *et al*., 2011).

Malaria infections have been established to cause alterations in haematological parameters (Dondorp *et al*., 2008; Momoh and Longe, 2014). The diversity of resistance types will require that public health measures to control malaria should be region-specific (Willcox and Bodeker, 2004). One of the most promising prospects in the search for new antimalarial drugs is the large repository of medicinal plants used in the treatment of malaria in traditional societies. *Alstonia boonei* have been reported to have effects on the haematological and biochemical characteristics of rats (Ebiloma *et al*., 2012; Onyishi *et al*., 2020). This study, therefore, evaluated the anti-malaria potentials of extracts of *A. boonei* and its effects on the haematological indices of albino mice infected with *P. berghei*.

MATERIALS AND METHODS

Procurement and Extraction of *Alstonia boonei*: The plant leaves and roots were collected from Inyi in Enugu-Ezike (Igbo-Eze North Local Government Area) of Enugu State by a traditional herbalist. The plant parts were identified and authenticated by a plant taxonomist in the Department of Plant Science and Biotechnology, University of Nigeria, Nsukka, where the voucher specimens (PSBH 2017-141) were kept in the departmental herbarium. Fresh leaves and roots of *A. boonei* were washed, sliced and air-dried separately at room temperature. Each of them was pulverized and 800 g of the leaves and roots fine powder obtained were divided into two. Half of the separate fine powders were percolated in 1400 ml of water and the other half in 70 % ethanol, for aqueous and ethanolic extracts, respectively. They were filtered after 72 hours and filtrates evaporated to dryness using a temperature-regulated water bath preset at 40°C to yield the extracts concentrates. These were labelled accordingly and stored in a refrigerator at 4°C until use.

Procurement and Maintenance of Albino Mice: Two hundred and sixteen male albino mice used in the study were procured from the Genetic and Breeding Laboratory in the Department of Zoology and Environmental Biology. The mice were maintained according to National Research Council Guidelines on laboratory animal use (NRC, 2011). Food and water were available *ad libitum*.

Procurement and Inoculation of *Plasmodium berghei*: The artesunate-sensitive strain of the rodent parasite *Plasmodium berghei* NK-65 was obtained from our institution Veterinary Teaching Hospital. The strain was maintained in the laboratory for the period of the study by *in vivo* serial blood passage from mouse to mouse (Ebiloma *et al*., 2012). A set of mice parasitized with *P. berghei* NK-65 were anaesthetized after 6 days having shown clinical symptoms of malaria and confirmed microscopically. Blood samples were collected by cardiac puncture using a sterile syringe and needle. The samples were diluted in normal saline (1 ml of blood in 10 ml of normal saline), and 0.3 ml of the parasitized erythrocyte used to infect each of the experimental mice intraperitoneally.

Study Design and Anti-*Plasmodium* Effects of Plant Extracts: This study adopted a completely randomized design. Six treatment groups containing six mice each in three replicates for each extract was used in the present study. Group I: normal control, mice not infected and not treated; Group II: negative control, infected and not treated; Group III: standard control, infected and treated with the standard drug (artesunate); Groups IV, V and VI: 400, 600 and 800 mg/kg body wt/day, infected and treated with 400, 600 and 800 mg/kg/day of extract. Three mice from each group were used for the suppressive test (treated 4 hours after parasite inoculation for 4 days), and the remaining for the curative test (treated 72 hours after parasite inoculation for 4 days) as well as for biochemical examination, after which their mean survival time was estimated (Ryley and Peters, 1970).

Collection and Examination of Blood Samples: Blood samples collection were made at both tail and ocular regions at three periods (24 hours before parasite inoculation; 72 hours after parasite inoculation; and 24 hours after

overall treatment for 4 days). Blood samples were collected from the tail of each mouse to make a thin blood smear following standard procedure, and the parasitaemia level was determined microscopically (CDC, 2015). From the parasitaemia level, percentage parasitaemia and suppression were then deduced (Peters, 1967; CDC, 2015). Also, blood samples collection by ocular puncturing were dispensed into EDTA bottles for haematological examinations of packed cell volume (PCV), haemoglobin (Hb) concentration, red blood cell (RBC) counts, mean corpuscular haemoglobin concentration (MCHC), mean corpuscular volume (MCV), white blood cell (WBC) counts, mean haemoglobin concentration (MHC), lymphocyte, neutrophil, platelets, eosinophil, and monocyte using the methods of Adewuyi and Olatunji (1995).

Statistical Analysis: Data were entered into Microsoft Excel and analyzed using SPSS version 20. Analysis of Variance (ANOVA) and Duncan tests was used to compare the mean differences of the concentrations of the extracts. The mean difference of $p<0.05$ was regarded as significant.

RESULTS

The Anti-*Plasmodium* Effects: The anti-malaria activities of the aqueous and ethanolic leaf and root extracts of *A. boonei* in *Plasmodium berghei* infected mice are summarized in Tables 1 – 4. Tables 1 and 2 show the percentage parasitaemia and average percentage suppression of parasitaemia when treatment was commenced 4 hours post-infection. The aqueous leaf, ethanolic leaf and ethanolic root extracts at concentration 400, 600 and 800 mg/kg body wt/day had similar parasitaemia suppressive ability comparable to the standard drug (Table 2). This suppressive ability was concentration-dependent, increasing as concentration increases. Tables 3 and 4 represent the percentage parasitaemia and percentage suppression of *A. boonei* extracts on mice infected with *P. berghei* for the curative test.

Table 3 show the percentage of parasitaemia in mice infected by *P. berghei* at the end of day 4 treatment with either standard drug or root and leaf extracts of *A. boonei*. The parasitaemia suppressive effect of the extracts or standard drug at the end of day 4 treatment is represented as percentage suppression (Table 4). The percentage parasitaemia was significantly less ($p<0.05$) in the groups administered 400, 600 and 800 mg/kg body wt/day concentrations of *A. boonei* extracts compared to the negative control group. The percentage suppression of parasitaemia in the mice administered 400, 600 or 800 mg/kg body wt/day of the aqueous leaf, ethanolic leaf and root extracts of *A. boonei* was high and comparable to 5 mg/kg body wt/day of the standard drug after day 4 treatment.

The Effects on Haematological Indices: The effects of the aqueous and ethanolic leaf and root extracts of *A. boonei* in *Plasmodium berghei* infected mice haematological indices are summarized in Tables 5 – 16. The haematological indices of packed cell volume, haemoglobin level, red blood cell, mean corpuscular haemoglobin concentration and mean cell volume of mice infected with *P. berghei* between 4 and 7 days treatment with extracts demonstrated significantly ($p<0.05$) increased and normalized concentrations comparable to the standard control, as against decrease in concentrations seen in the negative control (Tables 5 – 9). Also, as observed from the results, the parameters of white blood cell, lymphocyte, and neutrophil were significantly ($p<0.05$) reduced and their levels returned to normalcy. The extract effects compared satisfactorily to the standard control (Tables 10 – 12). The mean haemoglobin concentration demonstrated an insignificantly ($p\geq0.05$) inconsistent increase comparable to the negative control (Table 13). The platelet count decreased after inoculation but normalized after treatment, but the eosinophil and monocyte concentrations of the mice were observed to show indifferent to treatments and/or infection. There were no significant ($p\geq0.05$) changes and the concentrations remained more or less unaltered (Tables 14 – 16).

Table 1: Percentage parasitaemia of *Plasmodium berghei* in infected mice post treated with aqueous and ethanolic leaf and root extracts of *Alstonia boonei* (suppressive test)

Treatments (mg/Kg/b.wt/day)	Percentage (%) parasitaemia of *Plasmodium berghei*			
	Aqueous Leaf Extract	Aqueous Root Extract	Ethanolic Leaf Extract	Ethanolic Root Extract
Normal Control	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}
Negative Control	21.23 ± 6.41^{b2}	51.27 ± 46.87^{bc2}	55.77 ± 48.29^{b2}	65.17 ± 17.97^{c2}
Standard Control	6.87 ± 1.53^{b1}	6.70 ± 1.93^{b1}	6.23 ± 1.14^{b1}	5.47 ± 0.67^{b1}
400	9.57 ± 2.03^{b1}	31.17 ± 5.73^{c2}	7.53 ± 2.41^{b1}	12.60 ± 3.81^{b1}
600	9.10 ± 2.00^{b1}	22.57 ± 7.95^{c2}	6.67 ± 1.76^{b1}	12.23 ± 1.19^{b1}
800	7.00 ± 1.75^{b1}	15.00 ± 3.05^{b12}	6.60 ± 2.44^{b1}	7.87 ± 2.67^{b1}

All values expressed as mean ± standard error mean. Different superscript letters on the same row indicates significance difference ($p<0.05$) in mean values among different extracts. Different superscript numerals in the same column indicates significance difference ($p<0.05$) in mean values within same extract

Table 2: Percentage suppression of *Plasmodium berghei* in infected mice post treated with aqueous and ethanolic leaf and root extracts of *Alstonia boonei* (suppressive test)

Treatments (mg/Kg/b.wt/day)	Percentage (%) suppression of *Plasmodium berghei*			
	Aqueous Leaf Extract	Aqueous Root Extract	Ethanolic Leaf Extract	Ethanolic Root Extract
Negative Control	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}
Standard Control	67.67 ± 7.42^{b1}	87.00 ± 3.79^{d2}	89.00 ± 2.00^{b2}	90.00 ± 1.16^{c2}
400	54.66 ± 9.62^{b2}	39.33 ± 11.14^{b1}	86.33 ± 4.49^{b3}	77.33 ± 7.77^{b3}
600	57.00 ± 9.50^{b1}	56.00 ± 15.31^{bc1}	88.00 ± 3.22^{b2}	78.00 ± 2.31^{b2}
800	66.33 ± 7.69^{b1}	71.00 ± 5.51^{c1}	88.33 ± 4.26^{b2}	86.00 ± 4.58^{bc2}

All values expressed as mean ± standard error mean. Different superscript letters on the same row indicates significance difference ($p<0.05$) in mean values among different extracts. Different superscript numerals in the same column indicates significance difference ($p<0.05$) in mean values within same extract

Table 3: Percentage parasitaemia of *Plasmodium berghei* in infected mice post treated at day four with aqueous and ethanolic leaf and root extracts of *Alstonia boonei* (curative test)

Treatments (mg/Kg/b.wt/day)	Percentage (%) parasitaemia of *Plasmodium berghei*			
	Aqueous Leaf Extract	Aqueous Root Extract	Ethanolic Leaf Extract	Ethanolic Root Extract
Normal Control	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}
Negative Control	65.27 ± 8.43^{c1}	67.66 ± 10.11^{c1}	67.00 ± 11.50^{c1}	82.60 ± 9.27^{c2}
Standard Control	7.00 ± 0.58^{b1}	16.00 ± 3.06^{b1}	6.47 ± 1.53^{b1}	10.63 ± 2.20^{b1}
400	12.00 ± 1.73^{b1}	24.00 ± 2.08^{b2}	12.93 ± 2.30^{b1}	19.60 ± 1.93^{b1}
600	9.33 ± 1.86^{b1}	21.33 ± 4.18^{b2}	11.50 ± 3.33^{b1}	18.43 ± 0.81^{b1}
800	8.17 ± 1.01^{b1}	14.03 ± 2.31^{b1}	7.67 ± 1.96^{b1}	15.33 ± 1.20^{b1}

All values expressed as mean ± standard error mean. Different superscript letters on the same row indicates significance difference ($p<0.05$) in mean values among different extracts. Different superscript numerals in the same column indicates significance difference ($p<0.05$) in mean values within same extract

Table 4: Percentage suppression of *Plasmodium berghei* in infected mice post treated at day four with aqueous and ethanolic leaf and root extracts of *Alstonia boonei* (curative test)

Treatments (mg/Kg/b.wt/day)	Percentage (%) suppression of *Plasmodium berghei*			
	Aqueous Leaf Extract	Aqueous Root Extract	Ethanolic Leaf Extract	Ethanolic Root Extract
Negative Control	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}
Standard Control	88.33 ± 1.76^{b2}	76.73 ± 4.32^{c1}	89.67 ± 2.33^{c2}	87.00 ± 2.52^{c2}
400	81.67 ± 2.60^{b2}	64.73 ± 2.99^{b1}	79.67 ± 3.53^{b2}	76.33 ± 2.33^{b2}
600	85.66 ± 2.85^{b2}	66.40 ± 3.82^{b1}	87.33 ± 1.20^{c2}	77.66 ± 0.88^{b2}
800	87.66 ± 1.45^{b2}	73.00 ± 2.52^{bc1}	88.00 ± 3.06^{c2}	81.67 ± 1.20^{b2}

All values expressed as mean ± standard error mean. Different superscript letters on the same row indicates significance difference ($p<0.05$) in mean values among different extracts. Different superscript numerals in the same column indicates significance difference ($p<0.05$) in mean values within same extract

Table 5: Effects of aqueous and ethanolic leaf and root extracts of *Alstonia boonei* treatments on packed cell volume of *Plasmodium berghei* infected mice

Treatments (mg/Kg/b.wt/day)	PCV (%)											
	Aqueous Leaf Extract			Aqueous Root Extract			Ethanolic Leaf Extract			Ethanolic Root Extract		
	BI	AI	AT	BI	AI	AT	BI	AI	AT	BI	AI	AT
Normal control	45.00 ± 6.56^{a1}	45.00 ± 6.56^{b1}	44.00 ± 3.06^{b1}	51.00 ± 0.58^{a1}	51.00 ± 0.58^{b1}	50.0 0 ± 0.58^{b1}	43.33 ± 3.84^{a1}	41.00 ± 4.04^{b1}	41.66 ± 3.67^{b1}	46.00 ± 4.16^{a1}	50.00 ± 3.51^{b1}	46.67 ± 4.37^{b1}
Negative control	45.67 ± 3.18^{a3}	36.66 ± 6.89^{ab2}	23.67 ± 13.67^{a1}	50.33 ± 2.91^{a3}	39.00 ± 3.21^{a2}	15.00 ± 3.50^{a1}	42.33 ± 2.96^{a3}	35.68 ± 2.60^{ab2}	11.33 ± 0.88^{a1}	49.33 ± 2.03^{a3}	42.33 ± 6.23^{b2}	24.00 ± 14.01^{a1}
Standard control	46.33 ± 4.81^{a2}	36.66 ± 2.03^{ab1}	41.33 ± 10.17^{b12}	49.00 ± 4.51^{a2}	39.00 ± 2.40^{a1}	46.00 ± 3.51^{b2}	44.33 ± 2.96^{a2}	32.00 ± 2.50^{a1}	41.00 ± 4.04^{b2}	49.00 ± 2.65^{a2}	38.00 ± 8.67^{ab1}	46.66 ± 3.84^{b2}
400	43.00 ± 4.04^{a2}	30.33 ± 5.37^{a1}	35.67 ± 4.91^{ab12}	49.00 ± 1.53^{a2}	41.00 ± 5.57^{a1}	44.66 ± 2.40^{b12}	39.00 ± 1.53^{a2}	33.33 ± 1.45^{a1}	35.67 ± 2.33^{b12}	47.00 ± 7.77^{a2}	32.67 ± 5.37^{a1}	38.67 ± 7.06^{ab12}
600	41.33 ± 2.03^{a2}	31.33 ± 4.06^{a1}	36.33 ± 4.81^{ab12}	50.00 ± 3.61^{a2}	39.00 ± 2.65^{a1}	45.67 ± 3.84^{b12}	41.00 ± 4.93^{a1}	35.00 ± 2.52^{ab1}	36.33 ± 3.48^{b1}	47.00 ± 7.77^{a2}	33.67 ± 5.78^{a1}	38.67 ± 6.33^{ab12}
800	42.33 ± 16.74^{a2}	29.33 ± 2.33^{a1}	37.66 ± 4.10^{ab2}	51.33 ± 2.91^{a2}	39.67 ± 5.04^{a1}	45.67 ± 5.36^{b12}	45.33 ± 3.84^{a3}	28.66 ± 1.76^{a1}	39.00 ± 0.58^{b2}	46.00 ± 6.03^{a2}	34.33 ± 7.22^{a1}	40.67 ± 9.84^{ab2}

All values expressed as mean ± standard error mean. Different superscript letters on the same row indicates significance difference ($p<0.05$) in mean values among different extracts. Different superscript numerals in the same column indicates significance difference ($p<0.05$) in mean values within same extract. BI = Before Inoculation, AI = After Inoculation and AT = After Treatment

Table 6: Effects of aqueous and ethanolic leaf and root extracts of *Alstonia boonei* treatments on haemoglobin level of *P. berghei* infected mice

Treatments (mg/Kg/b.wt/day)	Hb (g/dL)											
	Aqueous Leaf			Aqueous Root			Ethanolic Leaf			Ethanolic Root		
	BI	AI	AT	BI	AI	AT	BI	AI	AT	BI	AI	AT
Normal control	14.23 ± 3.12^{a1}	14.23 ± 3.12^{b1}	14.67 ± 1.04^{ab1}	17.01 ± 0.18^{a1}	16.33 ± 0.33^{a1}	16.66 ± 0.20^{b1}	14.43 ± 2.96^{a1}	14.66 ± 1.35^{b1}	13.80 ± 1.25^{b1}	15.67 ± 1.17^{a1}	15.34 ± 1.38^{a1}	15.56 ± 1.44^{b1}
Negative control	15.03 ± 2.19^{a2}	11.11 ± 3.18^{a12}	7.90 ± 4.56^{a1}	17.28 ± 1.47^{a2}	13.00 ± 1.07^{a2}	5.00 ± 1.15^{a1}	15.80 ± 1.16^{a2}	11.89 ± 0.87^{ab2}	3.77 ± 0.29^{a1}	16.45 ± 0.69^{a2}	14.12 ± 2.08^{a2}	8.03 ± 4.68^{a1}
Standard control	15.20 ± 1.05^{a1}	12.39 ± 0.53^{a1}	13.77 ± 3.38^{a1}	16.28 ± 1.48 ^{a1}	13.55 ± 0.80^{a1}	15.33 ± 1.16^{b1}	14.78 ± 0.99 ^{a1}	10.68 ± 0.83^{a1}	13.70 ± 1.37^{b1}	16.34 ± 0.89^{a2}	12.69 ± 2.88^{a1}	15.00 ± 1.00^{b2}
400	12.13 ± 1.62^{a1}	10.11 ± 1.79^{a1}	11.90 ± 1.63^{ab1}	16.34 ± 0.53^{a1}	13.67 ± 1.86^{a1}	14.86 ± 0.81^{b1}	13.01 ± 0.51^{a1}	11.11 ± 0.49^{ab1}	11.90 ± 0.78^{b1}	13.33 ± 2.14^{a1}	10.89 ± 1.79^{a1}	12.90 ± 2.11^{ab1}
600	11.03 ± 1.35^{a1}	10.46 ± 1.36^{a1}	12.13 ± 1.62^{ab1}	16.65 ± 1.21^{a1}	12.99 ± 0.88^{a1}	15.10 1.72^{b1}	13.68 ± 1.66 ^{a1}	11.71 ± 0.82^{ab1}	12.10 ± 1.16^{b1}	15.66 ± 2.59^{a1}	11.22 ± 1.93^{a1}	13.10 ± 2.57^{ab1}
800	13.77 ± 0.67^{a12}	9.78 ± 0.78^{a1}	12.57 ± 1.37^{ab12}	17.11 ± 0.98 ^{a1}	13.22 ± 1.86^{a1}	15.26 ± 1.29^{b1}	15.12 ± 1.28^{a2}	9.57 ± 0.58^{a1}	13.00 ± 0.23^{b12}	14.22 ± 3.12^{a1}	11.45 ± 2.41^{a1}	13.70 ± 3.35^{ab1}

All values expressed as mean ± standard error mean. Different superscript letters on the same row indicates significance difference ($p<0.05$) in mean values among different extracts. Different superscript numerals in the same column indicates significance difference ($p<0.05$) in mean values within same extract. BI = Before Inoculation, AI = After Inoculation and AT = After Treatment

Table 7: Effects of aqueous and ethanolic leaf and root extracts of *Alstonia boonei* treatments on red blood cell count of *Plasmodium berghei* infected mice

Treatments (mg/Kg/b.wt/day)	RBC (10^{12}/L)											
	Aqueous Leaf Extract			Aqueous Root Extract			Ethanolic Leaf Extract			Ethanolic Root Extract		
	BI	AI	AT	BI	AI	AT	BI	AI	AT	BI	AI	AT
Normal control	108.00 ± 9.54^{a1}	108.00 ± 9.54^{c1}	105.33 ± 5.04^{b1}	128.67 ± 7.69^{ab1}	136.00 ± 7.55^{b1}	135.33 ± 7.42^{b1}	120.00 ± 2.89^{a1}	135.33 ± 2.72^{b2}	132.33 ± 5.37^{c2}	141.00 ± 5.51^{a1}	150.00 ± 8.51^{b2}	139.66 ± 0.88^{b1}
Negative control	106.00 ± 7.02^{a3}	88.33 ± 9.28^{ab2}	55.00 ± 6.03^{a1}	128.33 ± 4.41^{ab3}	110.33 ± 7.33^{a2}	88.66 ± 5.93^{a1}	122.33 ± 2.33^{ab3}	102.00 ± 7.57^{ab2}	58.33 ± 9.21^{a1}	142.00 ± 6.08^{a3}	112.33 ± 24.04^{ab2}	93.66 ± 6.36^{a1}
Standard control	113.00 ± 3.22^{a2}	81.00 ± 9.50^{a1}	105.33 ± 4.38^{b2}	129.66 ± 10.41^{ab2}	121.33 ± 4.81^{ab2}	131.67 ± 5.24^{b2}	124.33 ± 3.38^{a2}	99.67 ± 11.32^{ab1}	123.33 ± 3.71^{bc3}	151.33 ± 2.40^{a2}	102.33 ± 24.36^{ab1}	149.66 ± 1.73^{b2}
400	112.67 ± 1.45^{a3}	88.66 ± 4.9^{ab1}	101.67 ± 5.88^{b2}	133.67 ± 5.81^{ab2}	112.67 ± 11.10^{a1}	116.66 ± 10.17^{b1}	135.66 ± 5.90^{b3}	100.33 ± 8.37^{ab1}	110.67 ± 4.18^{b2}	150.00 ± 4.16^{a3}	115.33 ± 23.13^{ab1}	140.00 ± 2.31^{b2}
600	107.33 ± 8.74^{a2}	89.67 ± 9.87^{ab1}	102.67 ± 3.76^{b2}	126.00 ± 7.02^{a2}	117.33 ± 6.44^{ab1}	123.66 ± 7.45^{b2}	127.67 ± 4.49^{ab3}	89.33 ± 9.68^{a1}	116.00 ± 3.06^{bc2}	142.66 ± 1.45^{a2}	108.67 ± 6.33^{ab1}	140.56 ± 3.49^{b2}
800	113.67 ± 2.33^{a3}	84.00 ± 6.25^{a1}	104.33 ± 4.37^{b2}	131.67 ± 5.24^{ab2}	122.33 ± 3.93^{ab1}	128.33 ± 4.41^{b2}	122.33 ± 5.48^{ab2}	101.33 ± 9.96^{ab1}	118.67 ± 10.50^{bc2}	142.66 ± 4.91^{a2}	86.00 ± 12.22^{a1}	148.33 ± 4.41^{b3}

All values expressed as mean ± standard error mean. Different superscript letters on the same row indicates significance difference ($p<0.05$) in mean values among different extracts. Different superscript numerals in the same column indicates significance difference ($p<0.05$) in mean values within same extract. BI = Before Inoculation, AI = After Inoculation and AT = After Treatment

Table 8: Effects of aqueous and ethanolic leaf and root extracts of *Alstonia boonei* treatments on mean corpuscular haemoglobin concentration of *Plasmodium berghei* infected mice

Treatments (mg/Kg/b.wt/day)	MCHC (pg)											
	Aqueous Leaf Extract			Aqueous Root Extract			Ethanolic Leaf Extract			Ethanolic Root Extract		
	BI	AI	AT	BI	AI	AT	BI	AI	AT	BI	AI	AT
Normal control	0.28 ± 0.06^{a1}	0.33 ± 0.02^{a12}	0.39 ± 0.08^{b2}	0.33 ± 0.01^{a1}	0.33 ± 0.00^{a1}	0.33 ± 0.00^{b1}	0.33 ± 0.02^{a1}	0.33 ± 0.01^{a1}	0.34 ± 0.01^{a1}	0.34 ± 0.01^{a1}	0.33 ± 0.01^{a1}	0.33 ± 0.01^{a1}
Negative control	0.34 ± 0.01^{a2}	0.33 ± 0.04^{a12}	0.22 ± 0.02^{a1}	0.33 ± 0.01^{a2}	0.33 ± 0.00^{a2}	0.06 ± 0.22^{a1}	0.38 ± 0.05^{a1}	0.32 ± 0.02^{a1}	0.32 ± 0.03^{a1}	0.33 ± 0.01^{a1}	0.32 ± 0.01^{a1}	0.32 ± 0.00^{a1}
Standard control	0.34 ± 0.01^{a1}	0.34 ± 0.01^{a1}	0.33 ± 0.00^{ab1}	0.35 ± 0.00^{a1}	0.33 ± 0.01^{a1}	0.34 ± 0.01^{b1}	0.35 ± 0.00^{a1}	0.33 ± 0.01^{a1}	0.33 ± 0.01^{a1}	0.34 ± 0.01^{a1}	0.33 ± 0.01^{a1}	0.33 ± 0.08^{a1}
400	0.34 ± 0.01^{a1}	0.34 ± 0.00^{a1}	0.32 ± 0.01^{ab1}	0.34 ± 0.01^{a1}	0.32 ± 0.01^{a1}	0.34 ± 0.01^{b1}	0.35 ± 0.01^{a1}	0.32 ± 0.03^{a1}	0.33 ± 0.01^{a1}	0.32 ± 0.01^{a1}	0.31 ± 0.01^{a1}	0.32 ± 0.00^{a1}
600	0.34 ± 0.01^{a1}	0.33 ± 0.01^{a1}	0.33 ± 0.01^{ab1}	0.36 ± 0.03^{a1}	0.34 ± 0.01^{a1}	0.34 ± 0.02^{b1}	0.33 ± 0.00^{a1}	0.32 ± 0.02^{a1}	0.32 ± 0.01^{a1}	0.34 ± 0.00^{a1}	0.33 ± 0.01^{a1}	0.32 ± 0.01^{a1}
800	0.33 ± 0.01^{a1}	0.32 ± 0.01^{a1}	0.35 ± 0.02^{ab1}	0.34 ± 0.00^{a1}	0.33 ± 0.01^{a1}	0.33 ± 0.01^{b1}	0.34 ± 0.01^{a1}	0.32 ± 0.01^{a1}	0.33 ± 0.02^{a1}	0.33 ± 0.01^{a1}	0.34 ± 0.01^{a1}	0.33 ± 0.00^{a1}

All values expressed as mean ± standard error mean. Different superscript letters on the same row indicates significance difference ($p<0.05$) in mean values among different extracts. Different superscript numerals in the same column indicates significance difference ($p<0.05$) in mean values within same extract. BI = Before Inoculation, AI = After Inoculation and AT = After Treatment

Table 9: Effects of aqueous and ethanolic leaf and root extracts of *Alstonia boonei* treatments on mean cell volume of *Plasmodium berghei* infected mice

Treatments (mg/Kg/b.wt/day)	MCV (g/dl)											
	Aqueous Leaf Extract			Aqueous Root Extract			Ethanolic Leaf Extract			Ethanolic Root Extract		
	BI	AI	AT	BI	AI	AT	BI	AI	AT	BI	AI	AT
Normal control	0.42 ± 0.12^{a12}	0.34 ± 0.09^{a1}	0.49 ± 0.08^{b2}	0.37 ± 0.02^{a1}	0.37 ± 0.02^{a1}	0.40 ± 0.07^{b1}	0.36 ± 0.04^{a1}	0.36 ± 0.05^{a1}	0.37 ± 0.05^{b}	0.36 ± 0.04^{a1}	0.34 ± 0.01^{a1}	0.35 ± 0.07^{a1}
Negative control	0.42 ± 0.06^{a2}	0.42 ± 0.05^{a2}	0.20 ± 0.13^{a1}	0.33 ± 0.02^{a2}	0.36 ± 0.04^{a2}	0.14 ± 0.07^{a1}	0.36 ± 0.03^{a1}	0.33 ± 0.03^{a1}	0.20 ± 0.04^{a1}	0.35 ± 0.03^{a12}	0.34 ± 0.17^{a2}	0.24 ± 0.06^{a1}
Standard control	0.41 ± 0.04^{a1}	0.37 ± 0.07^{a1}	0.41 ± 0.13^{b1}	0.41 ± 0.07^{a2}	0.32 ± 0.03^{a1}	0.37 ± 0.04^{b12}	0.39 ± 0.04^{a1}	0.33 ± 0.03^{a1}	0.36 ± 0.04^{b1}	0.34 ± 0.02^{a1}	0.33 ± 0.05^{a1}	0.33 ± 0.04^{a1}
400	0.32 ± 0.04^{a1}	0.30 ± 0.06^{a1}	0.33 ± 0.06^{ab1}	0.38 ± 0.03^{a1}	0.37 ± 0.08^{a1}	0.34 ± 0.32^{b1}	0.39 ± 0.03^{a1}	0.33 ± 0.03^{a1}	0.27 ± 0.03^{ab1}	0.31 ± 0.05^{a1}	0.30 ± 0.05^{a1}	0.26 ± 0.06^{a1}
600	0.37 ± 0.04^{a1}	0.31 ± 0.09^{a1}	0.36 ± 0.06^{ab1}	0.40 ± 0.04^{a1}	0.33 ± 0.03^{a1}	0.35 ± 0.06^{b1}	0.42 ± 0.03^{b1}	0.31 ± 0.08^{a2}	0.30 ± 0.04^{ab1}	0.33 ± 0.05^{a1}	0.32 ± 0.07^{a1}	0.27 ± 0.04^{a1}
800	0.37 ± 0.03^{a1}	0.36 ± 0.04^{a1}	0.40 ± 0.06^{b1}	0.39 ± 0.04^{a1}	0.33 ± 0.05^{a1}	0.37 ± 0.02^{b1}	0.37 ± 0.03^{a1}	0.30 ± 0.04^{a1}	0.31 ± 0.03^{ab1}	0.37 ± 0.07^{a12}	0.31 ± 0.09^{a2}	0.34 ± 0.03^{a1}

All values expressed as mean ± standard error mean. Different superscript letters on the same row indicates significance difference (p<0.05) in mean values among different extracts. Different superscript numerals in the same column indicates significance difference (p<0.05) in mean values within same extract. BI = Before Inoculation, AI = After Inoculation and AT = After Treatment

Table 10: Effects of aqueous and ethanolic leaf and root extracts of *Alstonia boonei* treatments on white blood cell count of *Plasmodium berghei* infected mice

Treatments (mg/Kg/b.wt/day)	WBC (10^9L)											
	Aqueous Leaf Extract			Aqueous Root Extract			Ethanolic Leaf Extract			Ethanolic Root Extract		
	BI	AI	AT	BI	AI	AT	BI	AI	AT	BI	AI	AT
Normal control	21666.67± 18666.67^{a2}	20666.67 ± 10728.98^{a2}	19366.67 ± 1770.44^{a1}	11766.67± 392.99^{a1}	11746.66± 127.19^{a1}	12816.67± 101.37^{a1}	13033.33± 1010.50^{a1}	14166.67± 783.87^{a1}	12000.00± 577.35^{a1}	12500.00± 1167.62^{a1}	13066.66± 829.32^{a1}	13066.66± 970.11^{a1}
Negative control	24550.00± 12402.05^{a1}	28666.67 ± 10728.98^{ab12}	30466.67 ± 10848.25^{b12}	11600.00± 208.17^{a1}	17666.66± 881.91^{a12}	68900.00± 1021.44^{b3}	12800.00± 901.85^{a1}	14533.33± 866.60^{a1}	28333.33 ± 1201.85^{c2}	12766.66± 693.62^{a1}	13400.00± 802.08^{a1}	46933.33± 17076.43^{b2}
Standard control	19366.67± 1770.44^{a1}	21166.67 ± 18025.01^{a1}	16833.33 ± 1319.51^{a1}	14533.33± 2051.2^{a1}	16166.67± 664.161^{a1}	14266.67± 589.73^{a1}	11666.67± 664.16^{a1}	13333.67± 5364.49^{a1}	12666.33 ± 1452.97^{ab1}	12200.00± 757.19^{a1}	12700.00± 665.83^{a1}	12300.00± 650.64^{a1}
400	24550.00± 12402.05^{a1}	30466.67 ± 10848.25^{b12}	28666.67 ± 10728.98^{ab12}	13500.00± 550.78^{a1}	16333.33± 2603.42^{a1}	17966.67± 1017.08^{a1}	12500.00± 929.16^{a1}	16166.33± 433.33^{a1}	15233.67 ± 1301.71^{b1}	13700.00± 971.25^{a1}	15966.33± 1301.71^{a1}	14833.66± 4524.13^{a1}
600	20700.00± 10051.04^{a1}	30333.33 ± 11921.04^{b1}	28700.00 ± 11735.56^{ab1}	13233.33± 4394.82^{a1}	15100.00± 1069.27^{a1}	15983.33± 2658.37^{a1}	13566.66± 433.33^{a1}	14300.00± 378.59^{a1}	14000.00 ± 351.19^{b1}	12766.67± 926.16^{a1}	14533.33± 866.67^{a1}	14233.33± 1277.15^{a1}
800	24666.66± 44409.59^{a1}	25000.00 ± 4409.59^{a1}	21333.33 ± 4419.03^{a1}	15133.33± 1179.45^{a1}	18033.33± 983.76^{a1}	14866.67± 1125.95^{a1}	13066.67± 1449.52^{a1}	14500.00± 1322.88^{a1}	13000..00± 748.56^{ab1}	14500.00± 3752.78^{a1}	17666.66± 5174.73^{a1}	13833.33± 693.62^{a1}

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Table 11: Effects of aqueous and ethanolic leaf and root extracts of *Alstonia boonei* treatments on lymphocyte count of *Plasmodium berghei* infected mice

Treatments (mg/Kg/b.wt/day)	Lymphocytes (%)											
	Aqueous Leaf Extract			Aqueous Root Extract			Ethanolic Leaf Extract			Ethanolic Root Extract		
	BI	AI	AT	BI	AI	AT	BI	AI	AT	BI	AI	AT
Normal control	60.33 ± 8.88^{a1}	60.00 ± 7.55^{a1}	60.67 ± 10.34^{a1}	76.66 ± 1.77^{a1}	70.33 ± 1.45^{a1}	70.33 ± 1.45^{a1}	76.66 ± 1.20^{a1}	70.66 ± 1.33^{a1}	70.00 ± 4.58^{a1}	70.00 ± 6.51^{ab2}	70.00 ± 6.51^{a1}	73.33 ± 4.41^{a1}
Negative control	60.00 ± 5.29^{a1}	78.66 ± 9.94^{ab12}	80.33 ± 5.84^{b2}	71.33 ± 5.21^{a1}	79.67 ± 0.88^{a1}	89.67 ± 1.20^{b2}	70.33 ± 0.88^{a1}	74.66 ± 1.20^{a1}	88.68 ± 0.88^{b2}	67.33 ± 7.77^{a1}	69.67 ± 0.88^{a1}	80.00 ± 0.88^{a2}
Standard control	62.00 ± 9.07^{a1}	88.33 ± 4.91^{b1}	62.67 ± 6.84^{a1}	74.33 ± 4.41^{a1}	79.66 ± 2.60^{a1}	74.33 ± 2.96^{a1}	74.00 ± 3.22^{a1}	78.66 ± 1.33^{ab1}	79.33 ± 0.33^{a1}	68.00 ± 1.53^{ab1}	70.00 ± 5.77^{a1}	70.00 ± 2.89^{a1}
400	64.00 ± 7.94^{a1}	80.33 ± 5.90^{ab1}	68.00 ± 1.53^{a1}	78.00 ± 1.53^{a1}	80.00 ± 4.06^{a1}	77.33 ± 4.41^{a1}	73.67 ± 0.87^{a1}	81.66 ± 0.88^{b1}	81.67 ± 0.88^{b1}	70.67 ± 1.45^{ab1}	72.00 ± 3.22^{a1}	73.33 ± 2.60^{a1}
600	65.00 ± 2.08^{a1}	76.67 ± 2.03^{ab2}	66.67 ± 6.36^{a2}	70.33 ± 2.03^{a1}	80.33 ± 3.18^{a1}	74.33 ± 0.88^{a1}	70.66 ± 0.88^{a1}	70.66 ± 1.20^{a1}	80.67 ± 0.33^{b1}	71.33 ± 1.86^{ab1}	72.00 ± 2.01^{a1}	73.00 ± 6.51^{a1}
800	60.00 ± 7.02^{a1}	80.33 ± 8.51^{ab1}	60.33 ± 6.01^{a1}	71.00 ± 4.73^{a1}	74.00 ± 6.08^{a1}	72.00 ± 5.57^{a1}	77.67 ± 1.45^{a1}	82.33 ± 0.88^{b1}	80.00 ± 1.00^{b1}	69.00 ± 2.65^{ab1}	70.67 ± 0.88^{a1}	70.33 ± 0.88^{a1}

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Table 12: Effects of aqueous and ethanolic leaf and root extracts of *Alstonia boonei* treatments on neutrophil count of *Plasmodium berghei* infected mice

Treatments (mg/Kg/b.wt/day)	Neutrophils (%)											
	Aqueous Leaf Extract			Aqueous Root Extract			Ethanolic Leaf Extract			Ethanolic Root Extract		
	BI	AI	AT	BI	AI	AT	BI	AI	AT	BI	AI	AT
Normal control	18.33 ± 4.84^{a1}	18.33 ± 2.03^{a1}	18.33 ± 2.33^{a1}	15.00 ± 1.73^{a1}	16.67 ± 2.03^{a1}	16.66 ± 2.03^{a1}	14.66 ± 3.38^{a1}	16.66 ± 3.33^{a1}	16.67 ± 3.33^{a1}	24.00 ± 3.06^{a1}	22.67 ± 3.33^{a1}	20.00 ± 5.78^{a1}
Negative control	18.00 ± 1.53^{a1}	20.67 ± 0.33^{ab2}	28.33 ± 4.84^{b2}	15.33 ± 1.86^{a1}	30.00 ± 2.08^{b2}	42.66 ± 3.71^{b2}	12.00 ± 1.53^{a1}	18.33 ± 4.41^{a12}	23.33 ± 6.66^{b2}	21.00 ± 3.46^{a1}	26.33 ± 1.45^{b1}	30.66 ± 0.88^{b2}
Standard control	13.33 ± 1.76 ^{a1}	25.67 ± 1.86^{b2}	16.33 ± 2.33^{a1}	16.00 ± 1.53^{a1}	29.66 ± 2.03^{b2}	18.00 ± 1.15^{a1}	12.00 ± 1.73^{a1}	18.67 ± 4.67^{a1}	15.00 ± 2.87^{a1}	20.33 ± 4.41^{a1}	27.00 ± 2.08^{b1}	21.66 ± 6.01^{a1}
400	19.33 ± 1.76^{a1}	24.66 ± 4.06^{b2}	22.67 ± 0.88^{ab12}	21.66 ± 2.40^{a1}	30.00 ± 1.15^{b2}	24.33 ± 2.19^{ab12}	13.67 ± 2.60^{a1}	20.66 ± 2.96^{a2}	19.67 ± 6.01^{ab12}	20.33 ± 5.37^{a1}	24.33 ± 2.33^{ab1}	23.00 ± 1.15^{a1}
600	17.00 ± 4.04^{a1}	25.67 ± 4.06^{b2}	22.00 ± 1.73^{ab12}	15.67 ± 2.33^{a1}	29.33 ± 0.67^{b2}	19.00 ± 3.05^{a1}	16.33 ± 3.28^{a1}	24.33 ± 4.70^{b2}	18.67 ± 6.01^{a12}	23.33 ± 1.45^{a1}	29.33 ± 1.20^{b1}	22.67 ± 1.20^{a1}
800	13.33 ± 1.46^{a1}	21.33 ± 1.20^{ab2}	19.33 ± 4.10^{a12}	17.33 ± 2.33^{a1}	23.00 ± 3.61^{b1}	19.00 ± 2.65^{a1}	20.33 ± 4.70^{a1}	25.66 ± 5.81^{b1}	15.00 ± 3.01^{a1}	24.33 ± 2.03^{a1}	28.66 ± 0.88^{b1}	21.67 ± 3.33^{a1}

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Table 13: Effects of aqueous and ethanolic leaf and root extracts of *Alstonia boonei* treatments on mean haemoglobin concentration of *Plasmodium berghei* infected mice

Treatments (mg/Kg/b.wt/day)	MHC (fl)											
	Aqueous Leaf Extract			Aqueous Root Extract			Ethanolic Leaf Extract			Ethanolic Root Extract		
	BI	AI	AT	BI	AI	AT	BI	AI	AT	BI	AI	AT
Normal control	0.11 ± 0.02^{a1}	0.11 ± 0.03^{a1}	0.20 ± 0.07^{b2}	0.12 ± 0.01^{a1}	0.12 ± 0.01^{a1}	0.23 ± 0.01^{b2}	0.12 ± 0.02^{a1}	0.12 ± 0.02^{a1}	0.12 ± 0.10^{b1}	0.12 ± 0.02^{a1}	0.11 ± 0.02^{a1}	0.11 ± 0.02^{b1}
Negative control	0.14 ± 0.02^{a2}	0.12 ± 0.03^{a2}	0.07 ± 0.04^{a1}	0.12 ± 0.01^{a2}	0.11 ± 0.02^{a2}	0.06 ± 0.02^{a1}	0.13 ± 0.01^{a2}	0.12 ± 0.01^{a2}	0.06 ± 0.01^{a1}	0.12 ± 0.01^{a1}	0.11 ± 0.06^{a1}	0.08 ± 0.02^{a1}
Standard control	0.16 ± 0.01^{a1}	0.13 ± 0.02^{a1}	0.14 ± 0.04^{ab1}	0.14 ± 0.02^{a1}	0.11 ± 0.01^{a1}	0.13 ± 0.02^{ab1}	0.13 ± 0.01^{a1}	0.11 ± 0.01^{a1}	0.12 ± 0.03^{b1}	0.11 ± 0.01^{a1}	0.11 ± 0.01^{a1}	0.12 ± 0.03^{b1}
400	0.11 ± 0.02^{a1}	0.10 ± 0.02^{a1}	0.11 ± 0.02^{ab1}	0.13 ± 0.02^{a1}	0.12 ± 0.01^{a1}	0.12 ± 0.01^{ab1}	0.12 ± 0.02^{a2}	0.10 ± 0.00^{a2}	0.11 ± 0.01^{ab1}	0.13 ± 0.02^{a1}	0.10 ± 0.02^{a1}	0.11 ± 0.01^{b1}
600	0.13 ± 0.01^{a1}	0.10 ± 0.03^{a1}	0.12 ± 0.02^{ab1}	0.14 ± 0.01^{a1}	0.11 ± .02^{a1}	0.12 ± 0.02^{ab1}	0.14 ± 0.01^{a1}	0.10 ± 0.3 ^{a1}	0.11 ± 0.01^{ab1}	0.11 ± 0.02^{a2}	0.11 ± 0.02^{a2}	0.11 ± 0.02^{b1}
800	0.12 ± 0.01^{a1}	0.12 ± 0.02^{a1}	0.14 ± 0.03^{ab1}	0.13 ± 0.01^{a1}	0.11 ± 0.02^{a1}	0.12 ± 0.02^{ab1}	0.13 ± 0.01^{a1}	0.10 ± 0.02^{a1}	0.10 ± 0.02^{ab1}	0.14 ± 0.02^{a1}	0.10 ± 0.03^{a1}	0.12 ± 0.01^{b1}

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Table 14: Effects of aqueous and ethanolic leaf and root extracts of *Alstonia boonei* treatments on platelet count of *Plasmodium berghei* infected mice

Treatments (mg/Kg/b.wt/day)	Platelets											
	Aqueous Leaf Extract			Aqueous Root Extract			Ethanolic Leaf Extract			Ethanolic Root Extract		
	BI	AI	AT	BI	AI	AT	BI	AI	AT	BI	AI	AT
Normal control	113.67 ± 0.89^{a2}	105.00 ± 2.52^{a1}	111.66 ± 7.62^{ab12}	141.33 ± 4.91^{a1}	141.67 ± 4.06^{b1}	143.00 ± 11.59^{b1}	130.33 ± 20.84^{a1}	127.00 ± 11.59^{a1}	135.33 ± 2.91^{b1}	132.67 ± 9.33^{a1}	133.33 ± 9.39^{a1}	130.66 ± 9.74^{b1}
Negative control	118.00 ± 2.65^{a1}	110.66 ± 4.98^{a1}	108.00 ± 7.60^{a1}	141.00 ± 5.03^{a2}	117.66 ± 14.24^{a1}	101.00 ± 14.98^{a1}	127.67 ± 6.36^{a1}	122.67 ± 8.74^{a1}	116.00 ± 1.53^{a1}	138.00 ± 9.01^{a1}	134.00 ± 9.45^{a1}	118.33 ± 26.82^{b1}
Standard control	117.33 ± 6.74^{a1}	107.00 ± 6.51^{a1}	118.67 ± 10.73^{a1}	148.67 ± 10.41^{a2}	118.00 ± 7.57^{a1}	128.33 ± 2.73^{ab12}	129.66 ± 5.24^{a1}	126.66 ± 11.92^{a1}	132.33 ± 2.91^{b1}	131.66 ± 7.69^{a1}	127.00 ± 15.63^{a1}	134.00 ± 6.11^{b1}
400	119.00 ± 18.77^{a1}	118.67 ± 6.10^{a1}	125.00 ± 19.92^{b1}	145.66 ± 15.72^{a12}	120.00 ± 6.21^{a1}	145.00 ± 3.61^{b2}	123.66 ± 6.94^{a1}	122.33 ± 2.84^{a1}	124.00 ± 3.06^{ab1}	136.66 ± 7.22^{a1}	124.66 ± 5.70^{a1}	138.66 ± 7.22^{b1}
600	119.67 ± 14.97^{a1}	116.33 ± 6.84^{a1}	128.00 ± 14.11^{b1}	144.00 ± 3.06^{a2}	119.66 ± 6.96^{a1}	142.67 ± 6.57^{b2}	122.67 ± 10.40^{a1}	120.67 ± 2.91^{a1}	129.67 ± 11.92^{ab1}	135.00 ± 5.13^{a1}	123.67 ± 8.29^{a1}	137.33 ± 5.46^{b1}
800	117.00 ± 6.66^{a1}	109.67 ± 4.10^{a1}	111.00 ± 5.85^{ab1}	147.67 ± 2.96^{a2}	115.66 ± 7.42^{a1}	140.67 ± 2.91^{b2}	129.33 ± 6.69^{a1}	124.66 ± 5.84^{a1}	131.33 ± 20.84^{ab1}	142.33 ± 6.44^{a1}	131.00 ± 4.16^{a1}	134.00 ± 8.71^{b1}

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Table 15: Effects of aqueous and ethanolic leaf and root extracts of *Alstonia boonei* treatments on eosinophil count of *Plasmodium berghei* infected mice

Treatments (mg/Kg/b.wt/day)	Eosinophils (%)											
	Aqueous Leaf Extract			Aqueous Root Extract			Ethanolic Leaf Extract			Ethanolic Root Extract		
	BI	AI	AT	BI	AI	AT	BI	AI	AT	BI	AI	AT
Normal control	1.00 ± 0.58^{a1}	2.00 ± 1.00^{a1}	1.00 ± 1.00^{a1}	1.67 ± 0.67^{a1}	1.67 ± 0.88^{a1}	1.66 ± 0.88^{a1}	1.33 ± 0.67^{a1}	1.67 ± 0.33^{a1}	0.66 ± 0.66^{a1}	0.33 ± 0.33^{a1}	0.66 ± 0.33^{ab1}	0.33 ± 0.33^{a1}
Negative control	0.00 ± 0.00^{a1}	0.67 ± 0.33^{a1}	0.00 ± 0.00^{a1}	2.00 ± 0.00^{a1}	1.00 ± 0.58^{a1}	2.00 ± 1.00^{a1}	1.00 ± 1.00^{a1}	1.67 ± 0.33^{a1}	2.33 ± 0.33^{a1}	0.67 ± O.33^{a1}	1.00 ± 0.58^{ab1}	1.33 ± 0.68^{a1}
Standard control	1.00 ± 1.00^{a1}	1.00 ± 0.58^{a1}	1.00 ± 0.58^{a1}	0.66 ± 0.66^{a1}	1.33 ± 0.67^{a1}	0.67 ± 0.67^{a1}	0.33 ± 0.33^{a1}	1.67 ± 0.33^{a1}	0.33 ± 0.33^{a1}	0.66 ± 0.33^{a1}	0.00 ± 0.00^{a1}	0.33 ± 0.33^{a1}
400	1.00 ± 0.57^{a1}	1.00 ± 0.58^{a1}	0.67 ± 0.67^{a1}	0.33 ± 0.33^{a1}	1.33 ± 0.33^{a1}	1.00 ± 1.00^{a1}	1.00 ± 0.58^{a1}	1.00 ± 0.58^{a1}	1.66 ± 0.88^{a1}	1.00 ± 0.58^{a1}	1.00 ± 0.05^{ab1}	1.00 ± 0.58^{a1}
600	0.67 ± 0.33^{a1}	1.00 ± 0.58^{a1}	0.67 ± 0.67^{a1}	1.00 ± 0.100^{a1}	1.00 ± 1.00^{a1}	0.67 ± 0.67^{a1}	1.00 ± 0.58^{a1}	1.33 ± 0.33^{a1}	1.33 ± 0.66^{a1}	0.33 ± 0.33^{a1}	0.33 ± 0.33^{a1}	0.67 ± 0.67^{a1}
800	1.00 ± 0.58^{a1}	1.00 ± 0.58^{a1}	1.00 ± 1.00^{a1}	0.33 ± 0.33^{a1}	1.00 ± 0.58^{a1}	0.67 ± 0.67^{a1}	0.66 ± 0.33^{a1}	1.66 ± 0.33^{a1}	1.00 ± 0.58^{a1}	1.00 ± 0.58^{a1}	0.33 ± 0.33^{a}	0.67 ± 0.67^{a1}

All values expressed as mean ± standard error mean. Different superscript letters on the same row indicates significance difference (p<0.05) in mean values among different extracts. Different superscript numerals in the same column indicates significance difference (p<0.05) in mean values within same extract. BI = Before Inoculation, AI = After Inoculation and AT = After Treatment

Table 16: Effects of aqueous and ethanolic leaf and root extracts of *Alstonia boonei* treatments on monocyte count of *Plasmodium berghei* infected mice

Treatments (mg/Kg/b.wt/day)	Monocytes (%)											
	Aqueous Leaf Extract			Aqueous Root Extract			Ethanolic Leaf Extract			Ethanolic Root Extract		
	PI	AI	AT	PI	AI	AT	PI	AI	AT	PI	AI	AT
Normal control	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a}	0.67 ± 0.33^{a1}	0.66 ± 0.66^{a1}	0.67 ± 0.67^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.33 ± 0.33^{a1}	0.67 ± 0.67^{a1}
Negative control	0.00 ± 0.00^{a1}	0.66 ± 0.66^{a1}	0.67 ± 0.67^{a1}	0.67 ± 0.33^{a1}	0.67 ± 0.33^{a1}	1.33 ± 0.67^{a1}	0.66 ± 0.66^{a1}	0.33 ± 0.33^{a1}	2.00 ± 0.00^{b2}	0.66 ± 0.66^{a1}	1.33 ± 0.88^{a1}	2.33 ± 2.33^{a1}
Standard control	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.63 ± 0.33^{a1}	0.00 ± 0.00^{a1}	0.33 ± 0.33^{a1}	0.00 ± 0.00^{a1}	0.34 ± 0.30^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.33 ± 0.33^{a1}	0.67 ± 0.67^{a1}
400	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.65 ± 0.00^{a1}	0.33 ± 0.33^{a1}	1.00 ± 1.00^{a1}	0.00 ± 0.00^{a1}	0.33 ± 0.33^{a1}	0.33 ± 0.33^{a1}	0.00 ± 0.00^{a1}	2.00 ± 0.00^{a2}	2.00 ± 0.58^{a2}
600	0.00 ± 0.00^{a1}	0.33 ± 0.33^{a1}	0.00 ± 0.00^{a1}	0.66 ± 0.33^{a1}	1.00 ± 0.58^{a1}	0.67 ± 0.67^{a1}	0.33 ± 0.33^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.33 ± 0.33^{a1}	0.67 ± 0.67^{a12}	1.33 ± 0.67^{a2}
800	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	0.63 ± 0.33^{a1}	1.00 ± 0.58^{a1}	0.33 ± 0.33^{a1}	0.00 ± 0.00^{a1}	0.33 ± 0.33^{a1}	0.00 ± 0.00^{a1}	0.00 ± 0.00^{a1}	1.33 ± 0.67^{a2}	1.00 ± 0.58^{a2}

All values expressed as mean ± standard error mean. Different superscript letters on the same row indicates significance difference (p<0.05) in mean values among different extracts. Different superscript numerals in the same column indicates significance difference (p<0.05) in mean values within same extract. BI = Before Inoculation, AI = After Inoculation and AT = After Treatment

DISCUSSION

Anti-*Plasmodium* Effects: Phytochemical compounds such as steroids, saponins, tannins, protein, flavonoids, are present in the extracts especially in the ethanolic leaf extract. In other studies, anti-*Plasmodium* effects of plant substances have been caused by alkaloids, terpenoids and flavonoids (Milliken, 1997; Tringali, 2000). These chemical compounds may be acting singly or in synergy with one another to exert the observed anti-malaria activity of the aqueous and ethanolic leaf and root extracts of *A. boonei* which were identified in this study. The dose-dependent anti-malaria activity is associated with high drug concentration in the blood due to repeated dosing with the drug and the extracts which have gained access into the parasites and exhibiting their effects. This was in agreement with the finding of Jeruto *et al*. (2015) where parasite clearance was much more pronounced on the fourth day. Furthermore, an earlier study of Onwusonye and Uwakwe (2012), who worked on stem bark of *A. boonei* observed better performance of the extracts when compared with chloroquine. Momoh and Longe (2014) also observed that the ethanolic leaf extract of *A. boonei* showed a marked anti-malaria effect in a dose-dependent manner; with parasitaemia being significantly reduced at higher concentrations compared to lower concentrations. One of the mechanisms behind the anti-malaria activity displayed by different plant extracts is the eliciting of anti-*Plasmodium* effects either by inducing an elevation of erythrocyte oxidation or by inhibiting the synthesis of proteins (Kirby *et al*., 1989). *Alstonia boonei* may owe its anti-*Plasmodium* potency to any of the above-mentioned mechanisms.

Effects on the Haematological Indices: Malaria anaemia is reported to be complex as it involves red blood cell destruction either by the parasites or as a result of the immune response or both (Mulenga *et al*., 2006). After inoculation of *P. berghei*, there was a decrease in the negative control group, standard control group and groups treated with extracts of different concentration when compared to the normal control groups. This was as a result of anaemia caused by the excessive destruction of red blood cells by the malaria parasite. Meraiyebu *et al*. (2012) had earlier reported a significant decrease in PCV of infected humans as a result of the destruction of red blood cells. After treatment, the normal control group, the standard control group and the groups treated with different concentration of extracts were significantly ($p<0.05$) higher when compared with the negative control group. This continuous decrease in PCV value in the negative control group as a result of excessive destruction of red blood cells which was normalized during the treatment in the standard control group and the groups treated with different concentration of the extracts. This is consistent with Momoh and Longe (2014) who observed that there was increased PCV in the animals treated with standard control and the groups treated with different concentration of the extract when compared to the negative control. After inoculation with *P. berghei*, there was a reduction in haemoglobin level and red blood cell of the infected group, but after treatment, the haemoglobin level and red blood cell in the treated groups of standard control and groups treated with extract of different concentration increased compared to the negative control group in all the extracts. This result agreed with Momoh and Longe (2014) and Ifeanyichukwu and Esan (2014) who observed that there was a normalization of the red blood cell by a herbal extract, respectively. This may be as a result of anaemia caused by the parasite infection which the extracts and standard drug try to normalize. This is most probable as implicated from earlier findings of Gavigan *et al*. (2001) who reported that growing parasites consume and degrade the intracellular proteins which are mainly haemoglobin and thus causes the reduction in haemoglobin. From the present study, it was observed that after treatment, there was a significant decrease in mean corpuscular haemoglobin concentration and mean cell volume in the negative control group when compared to the normal control group, standard control group and the groups treated with different concentration of the extract. This may be as a result of low-grade production of

tumour necrosis factor (TNF) in response to malaria parasitaemia which contributes to the pathogenesis of malaria anaemia. This was similar to the findings of Tchinda *et al*. (2007) who reported a decrease in MCHC due to low production of TNF as a result of anaemia caused by continuous multiplication of the malaria parasite in the negative control. However, this was contrary to the findings of Adesina *et al*. (2009) who reported an increase in MCHC and MCV of negative control as a result of the presence of the parasite at the cellular level and destruction of red blood cells. From the present study, it was observed that after treatment, there was a significant increase in the white blood cell and lymphocyte counts of the negative control group when compared to the standard control group and the groups treated with different concentration of the extracts as a result of leukaemia or tissue damage of the mice caused by continuous multiplication of the *P. berghei* in the untreated group. This implies that the extracts can normalize the white blood cell and lymphocytes population and suggests the ability of the extract to enhance blood component to phagocytose. This was similar to the findings of Odeghe-Othuke *et al.* (2012) that phagocytosis of the blood component can be enhanced by some extract and that extracts of *Anthocleista grandiflora* normalized the lymphocyte of infected mice. In the present study, the decrease in mean cell haemoglobin values in the negative control group was as a result of anaemia induced by malaria parasite infection and was similar to the observation made by Dondorp *et al*. (2008), that the severity and type of anaemia can be determined by the levels of MCV and MHC. This was against the observation made by Sowunmi *et al.* (2009), who stated that after the recovery period of malaria infection, MCV and MHC values were expected to decrease. In this study, platelet counts were significantly reduced in malaria-infected mice when compared to the normal control in all the extracts. This implies that thrombocytopenia may be a marker of *Plasmodium* infection. This supports the findings of Manas *et al.* (2014) that a decrease in platelet count of the malaria-infected patient is a result of thrombocytopenia in malaria infection. After treatment in this study, there was a continuous dropping in the level of platelets in the infected not treated group. This was as a result of continued infection when compared to the standard control group and those treated with different concentration of the extract thus suggesting a stimulatory effect of the extract on the platelet of the mice. In eosinophil and monocyte, no significant change was observed concerning the parasite and the extracts. The significant difference in negative control after treatment might be as a result of bacterial infection. This is most probable as Warimwe *et al.* (2013) who reported that they are not specific in malaria diagnosis.

Conclusion: The extracts of *A. boonei* leaf and root anti-*Plasmodium* activity was dependent on both dosage and duration as observed from the study, and have demonstrated satisfactory normalization efficacy to haematological indices in malaria treatment.

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