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Alchornea laxiflora [Benth.] Pax and K.Hoffm stem bark extract and fractions attenuated oxidative stress and liver injury in *Plasmodium berghei*-infected mice

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Abstract

Alchornea laxiflora [Benth.] Pax and K.Hoffm (Euphorbiaceae) is used in Ibibio ethnomedicine for the treatment of various diseases such as malaria. The stem bark extract and fractions of *Alchornea laxiflora* were investigated for effect on parasitaemia, hematological parameters, oxidative stress markers, lipid profile, liver function indices and histopathology of the liver in *Plasmodium berghei*-infected mice using curative test model. Acute toxicity test and phytochemical screening of the extract were also carried out. Phytochemical screening revealed the presence of active compounds. The stem bark extract (141 - 424 mg/kg, p.o.) with LD_{50} of 1414.21 mg/kg exerted significant ($P < 0.05–0.001$) antimalarial activity against *P. berghei* infection in curative tests with methanol and dichloromethane fractions having the highest activity. Significantly ($P < 0.05$-0.001) elevated PCV, eosinophils, WBC and platelets count as well as significantly ($P < 0.05$-0.001) reduced neutrophils, lymphocytes and monocytes percentages were observed with various treatments, while RBC counts, Hb concentration and basophils percentages were not altered relative to the control group. The extract/fractions treatment did not alter significantly ($P > 0.05$) liver function indices (AST, ALT, ALP, total protein, albumin, total cholesterol and total bilirubin) except significantly ($P < 0.05$) elevated conjugated bilirubin level in the group treated with methanol fraction. The levels of GSH, SOD, GPX and CAT were not affected significantly ($P > 0.05$), while significant ($P < 0.05$-0.001) reduction of MDA level in all extract/fractions -treated groups relative were evident. The stem bark extract and fractions did not cause any significant ($P > 0.05$) change in the lipid profile parameters (total cholesterol, triglycerides, HDL, LDL, and VLDL) when compared to control. Histology of liver sections revealed significant reductions in pathological features in infected mice treated with the extract and fractions. Phytochemical screening revealed the presence of alkaloids, flavonoids, phenol, terpenes, saponins and reducing sugars. These results suggest that the stembark extract/fractions of *Alchornea laxiflora* possess antimalarial, antiplasmodial, antioxidative stress and hepatoprotective activities due to its phytochemical constituents.

Keywords: Malaria, *Alchornea laxiflora,* Plasmodium *berghei,* antiplasmodial, antioxidative stress

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Introduction

Alchornea laxiflora [Benth.] Pax and K.Hoffm (Euphorbiaceae) is a deciduous shrub, ~6–10 m tall, that grows in most areas of Africa including Nigeria, Congo, Ethiopia, and throughout East Africa to Zimbabwe (Burkill, 1994). *A. laxiflora* is called "Opoto" and "Nwariwa," respectively, among the Yoruba and Ibibio tribes of Nigeria. The stem bark and branches have been used in traditional medicine for various purposes, notably for malaria, anemia, ringworm, venereal disease, typhoid fever, infertility in females, infectious diseases, tumor, inflammation, teething problems, and toothache in South Africa, Ghana, and Nigeria (Jain *et al.*, 2022).

Biological activities reported of the stem bark include; antioxidant (Farombi *et al.*, 2003; Oloyede *et al.,* 2010), anti-HIV, antibacterial and cytotoxic activities (Siwe-Noundou *et al.,* 2019), larvicidal effect (Oyedeji *et al.,* 2024), anticholinesterase activity (Elufioye, 2017), antibacterial activity against multi-drug resistant (MDR) bacteria, (Mbaveng *et al.,* 2015) and analgesic (Okokon *et al.,* 2025) activities. The phytochemical investigation of methanol extract from the stem bark resulted in the isolation of some compounds, including ellagic acid, 3-O-methylellagic acid, and 3-O-methylellagic acid-3-O-α-rhamnopyranoside (Sandjo *et al.*, 2011). A novel ellagic acid derivative, namely, 3,4,3′-tri-O-methylellagic acid, was isolated from the methanol extract obtained of the stem bark (Mbaveng *et al.,* 2015). Sandjo *et al.* (2011) and Tapondjou *et al.* (2016) isolated and established the structure of a known steroidal glycoside, β-sitosterol-3-O-β-D-glucopyranoside from the methanol extract of stem bark, ellagic acid; 3-O-methyl-ellagic acid, 3-O-β-D-glucopyranosyl-β-sitosterol, 3-O-acetyloleanolic acid and 3-O-acetyl-ursolic acid. Several pentacyclic triterpenoids, such as 3-acetyloleanolic acid, 3-acetoxyursolic acid, adipedatol, and betulin, have been found in the stem bark. Squalene and 2,2,4-trimethyl-3-(3,8,12,16-tetramethyl heptadeca-3,7,11,15-tetraenyl) -cyclohexanol have also been identified as triterpenoids in the stem bark extract (Sanjo *et al*., 2011). This study investigated the effect of stem bark extract and fractions of *A. laxiflora* on biochemical, haematological and lipid parameters of mice infected with *Plasmodium berghei*.

Materials and Methods

Plants collection

The stem bark of *Alchornea laxiflora* were collected in bushes in Uyo area, Akwa Ibom State, Nigeria in April 2024. The collection was identified and authenticated by a taxonomist in Department of Botany and Ecological Studies, University of Uyo, Uyo, Nigeria.

Extraction

The leaves were washed and shade dried for two weeks. The dried leaves were cut into smaller pieces and pulverized to powder. The powdered leaves were divided into two parts. One part was macerated in ethanol for 72 hours, while the remaining part was successively and gradiently macerated for 72 hours in each of, n-hexane, dichloromethane, ethyl acetate and methanol respectively. The liquid filtrate of each extract and fraction was concentrated and evaporated to dryness in *vacuo* 40^0C using a rotary evaporator. The various yields were calculated and the extract and fractions were stored in a refrigerator at -4^0C, until used

Phytochemical Screening

Phytochemical screening of the crude extract was carried out employing standard procedures (Trease and Evans, 1996, Sofowora, 1993),

Parasite inoculation

Chloroquine-sensitive strain of *Plasmodium berghei ANKA* strain was obtained from the National Institute of Medical Research (NIMER), Yaba Lagos, Nigeria and maintained by sub-passage of blood from infected to healthy mouse once every 7-8 days.

Each mouse used in the experiment was inoculated intraperitoneally with 0.2 mL of infected blood containing about 1 x 10^7 *P. berghei* parasitized erythrocytes collected from an infected mouse with 20-30% parasitaemia. The inoculum consisted of 5 x 10^7 *P. berghei* infected erythrocytes per milliliter prepared by determining both the percentage parasitemia and the erythrocytes count of the donor mouse and diluting the blood with isotonic saline in proportions indicated by both determinations (Atanu *et al.,* 2021; Enyiekere *et al.*, 2024). Parasitemia was monitored by standard methods. Thin blood smears were made on glass slides, fixed using methanol, and stained using Giemsa stain, and parasitemia was counted using a microscope and was calculated as a percentage of infected red blood cells (RBCs) relative to the total number of cells in a microscopic field at ×100 magnification according to the formula of Okokon and Nwafor (2010) as given below:

$$\text{Parasitemia (\%)} = \frac{\text{Total number of parasitised RBCs}}{\text{Total number of RBCs}} \times 100.$$

Experimental animals

Swiss albino mice (18-25 g), male and female, used in the study were obtained from the University of Uyo Animal House. They were kept in standard plastic cages in a well-ventilated room and left to acclimatized for a period of 10 days before the experiments. The mice were fed on standard pelleted diet and water *ad libitum*. The care and use of animals was in accordance with the National Institute of Health Guide for the Care and Use of laboratory Animals

(NIH Publication, 1996). Approval for the study was obtained from the University of Uyo Animal Ethics Committee.

Drug administration

The extract, fractions, chloroquine that were used in the study were administered orally with the aid of a stainless metallic feeding cannula.

Determination of median lethal dose (LD_{50})

The median lethal dose (LD_{50}) of the extract was estimated using albino mice by oral route using a modified method of Lorke (1983) as earlier described by Okokon *et al.* (2019). This was carried in two phases. The first phases involved oral administration of 3 different doses of the stem bark extract (10,100 and1000 mg/kg) to groups of three mice each. The second phase was done using four doses (2000, 3000, 4000 and 5000 mg/kg) and 1 mouse per dose. The animals were observed for manifestation of physical signs of toxicity and death. The number of deaths in each group within 24 hours was recorded. The LD_{50} was calculated as geometrical means of the maximum dose producing 0 % (a) and the minimum dose producing 100 % mortality.

Evaluation of Curative Activities of the extract and fractions .

This test was used to evaluate the schizontocidal activity of the extract, fractions and chloroquine in established plasmodial infection. This was conducted according to the methods described by Okokon *et al*. (2016). *P. berghei berghei* was injected intraperitoneally into ninety (90) mice on the first day *(D_0)*. Seventy two hours later (D_3), the mice were divided into nine groups of ten mice per group. Groups 1-3 were given different doses of extract, 141, 282 and 424 mg/kg, respectively, groups 4-7 were given 282 mg/kg of *n*-hexane, ethyl acetate, dichloromethane, and n-butanol fractions respectively, group 8 was given 5 mg/kg/ chloroquine (positive control) and group 9 was given 10 mL/kg distilled water (negative control). The crude extract, fractions and chloroquine were administered once daily for 5 days. Giemsa stained thin smears were prepared from tail blood samples collected on each day of treatment to monitor the parasitemia level. The mean survival time (MST) of the mice in each group was determined over a period of 29 days (D_0-D_{28}). Rectal temperatures of the mice were taken on days 0, 3, 5, and 7. On the sixth day, five mice from each group were sacrificed under diethyl ether vapour. Blood samples were collected into EDTA bottles and plain centrifuge tubes. The blood samples in the centrifuge tubes were allowed to stand for 2 hours before centrifuged at 2500 rpm for 15 min to separate the serum at room temperature and used for biochemical assays and oxidative stress markers assay.

Liver Function Test

The following liver function parameters were determined; Aspartate transaminase (AST), Alanine aminotransferase (ALT), Total Cholesterol, Alkaline phosphatase (ALP), Total plasma protein, Total and direct bilirubin. The determinations were done spectrophotometrically using Randox analytical kits according to standard procedures of manufacturer's protocols (Tietz, 1976) at the Chemical Pathology Department of University of Uyo Teaching Hospital.

Hematological study

Under diethyl ether anesthesia, blood samples were collected from each mouse by cardiac puncture using needles mounted on a 2 mL syringe into ethylene diamine tetra-acetic acid (EDTA) – coated sample bottles for hematological analyzes. Hematological parameters such as Red blood cell count (RBC), hemoglobin, (Hb), packed cell volume (PCV), platelet concentration (PLC) and total and differential white blood cell count (WBC) were determined. These parameters were analyzed using automatic hematological system (Sysmex Hematology – Coagulation system, Model MO-1000 I, Trans Asia, Japan).

Oxidative stress markers Assay

The liver parts of the animals were removed, weighed and washed with ice cold 0.9% NaCl and reduced to homogenates in a ratio of 1 g of wet tissue to 9 mL of 1.25% KCl by using motor driven Teflon-pestle. The homogenates were centrifuged at 7000 rpm for 10 min at 4˚C and the supernatants were used for the assays of superoxide dismutase (SOD) (Marklund and Marklund, 1974), catalase (CAT) (Sinha,1972), glutathione peroxidase (GPx) (Lawrence and Burk,1976), malondialdehyde (MDA) (Ester and Cheeseman, 1990) and reduced glutathione (GSH) (Ellman,1959).

Lipid Profile

Serum cholesterol, triglyceride and high density lipoprotein (HDL) levels of the treated animals were measured using standard colorimetric methods (Tietz, 1976). Low and very low-density lipoprotein (LDL and VLDL) were estimated from the formula of Friedwald *et al.* (1972). The determinations were done spectrophotometrically using Randox Analytical Kits® according to standard procedures of manufacturer's protocols (Tietz, 1976) at the Chemical Pathology Department of University of Uyo Teaching Hospital.

Histopathological Examination

The liver sections fixed in buffered formalin were processed and stained with haematotoxylin and eosin (H&E) for histological examination

Statistical Analysis

Data obtained from this study were statistically analysed using SPSS version 17. Statistical significance between the groups were analysed using one-way analysis of variance (ANOVA) and followed by Tukey-Kramer multiple comparison tests as post hoc test. Results were presented as Mean ± S.E.M. and values less than ($P < 0.05$ and 0.001) were considered significant.

Results

Yields of extract and fractions

The percentage yields of the extract and fractions were; crude-20.01%, n-hexane- 0.22%, DCM -0.82%, ethyl acetate -0.32%, n-butanol -2.1%.

Phytochemical constituents of the extract

The results of the phytochemical screening of the stem bark extract revealed the presence of alkaloids, phenol, terpenes, saponins, flavonoids and reducing sugars

Median Lethal Dose (LD_{50})

The oral median lethal dose of the extract was determined as 1414.21mg/kg.

Antiplasmodial effect of the extract and fractions on established infection

There was a dose-dependent reductions of parasitaemia in the extract/fraction-treated groups relative to control. These reductions were statistically significant ($P < 0.05$-0.001) especially in the groups treated with 424 mg/kg, DCM, ethyl acetate and methanol fractions. The methanol fraction had the highest activity with chemo suppression of 65.15 % on day 7, this was lower compared with that of the standard, chloroquine, 92.23% (Table 1). The extract and fractions demonstrated significant ($P < 0.05$-0.001) protective potentials as evidenced by the mean survival time of the animals. The groups treated with methanol fraction had a longer mean survival time, 17.46 ± 1.72 days followed by dichloromethane fraction treated mice, 16.54 ± 0.33 days (Table 1).

Table 1: Antiplasmodial activity of the extract and fractions on established *Plasmodium berghei* infection

Treatment	*Dose (mg/kg)*	Parasitaemia Day 7	% Chemossupression	MST (days)
Control	-	28.30±1.24	-	11.30 ±0.18
Extract	141	25.23±0.16	10.84	12.50±0.44
	282	23.44±0.24	17.17	13.88±0.28[a]
	424	12.22±1.34[c]	56.81	16.63±1.35[c]
n- HF	282	23.68±0.67	16.32	14.55±1.26[a]
DCMF	282	13.28±0.44[b]	53.07	16.54±0.33[c]
EAF	282	16.15±0.24[a]	42.93	15.33±1.34[b]
MF	282	9.86±0.14[c]	65.15	17.46±1.72[c]
Chloroquine	5	2.20±0.18[c]	92.23	29.17±0.13[c]

Values are expressed as mean ± SEM. Significant relative to control. [a]$P < 0.05$; [b]$P < 0.01$; [c]$P < 0.001$. n = 6.; HF-*n*-hexane fraction, DCMF-dichloromethane fraction, EAF-ethyl acetate fraction, MF-methanol fraction, MST-mean survival time.

Effect of the extract and fractions on haematological indices.

Administration of the extract and fractions did not cause any significant ($P > 0.05$) change on RBC, Hb concentration and basophils when compared to the control group (Table 2). The WBC count was significantly ($P < 0.05$) elevated in groups treated with low and middle doses of the extract (141 and 282 mg/kg) while neutrophils were significantly ($P < 0.05$-0.001) reduced in the groups treated with middle dose of the extract (282 mg/kg), DCM, ethyl acetate and methanol fractions as well as chloroquine when compared to control untreated infected mice. Percentage of lymphocytes was significantly ($P < 0.05$-0.001) reduced in groups treated with low and middle doses of the extract (141 and 282 mg/kg), methanol fraction and chloroquine when compared to control untreated infected mice. Monocytes were reduced ($P < 0.05$-0.01) in groups treated with low and middle doses of the extract (141 and 282 mg/kg) and chloroquine when compared to control untreated infected mice. Eosinophils were significantly ($P < 0.05$-0.01) increased in groups treated with middle and high doses of the extract, and chloroquine when compared to control untreated infected mice. PCV of the treated infected mice were significantly increased in groups treated with middle and high doses of the extract and *n*-hexane fraction. Platelets counts were slightly increased in high dose (424 mg/kg) treated group, but were reduced significantly ($p<0.05$-0.001) in groups treated with low and middle doses of the extract (141 and 282 mg/kg), n-hexane, DCM, ethyl acetate and methanol fractions as well as chloroquine (Table 2).

Effect of the extract and fractions on liver function parameters.

Treatment of *P. berghei* infected mice with the extract and fractions did not cause any significant ($P > 0.05$) alteration in the f liver function indices (AST, ALT, ALP, total protein, albumin, total cholesterol and total bilirubin. The level of conjugated bilirubin was however ($P < 0.05$) elevated in the group treated with methanol fraction relative to the untreated infected group (Table 3).

Effect of the extract and fraction on liver oxidative stress markers.

The extract and fractions did not cause significant ($P > 0.05$) change in the levels of GSH, SOD, GPX and CAT when compared to control. However, there was significant ($P < 0.05$-0.001) reduction in MDA level in the extract/fractions -treated groups relative to control (Table 4).

Effect of the extract and fraction on lipid profile

Administration of the extract and fractions did not produce significant ($P > 0.05$) alteration in the lipid profile parameters (total cholesterol, triglycerides, HDL, LDL, and VLDL) when compared to control. (Table 5).

Effect of the extract and fractions on the histology of liver.

The control animals had liver tissue demonstrating a severely altered hepato-architecture with multiple areas of haemorrhagic necrosis bordered by microvesicular steatosis with nuclear pyknosis and vacuolation. The presence of mononuclear inflammatory infiltrate was also observed (Figure 1A). Infected mice treated with the extract (141 mg/kg) had liver tissue showing moderately altered hepato-architecture with normal hepatocytes and multiple areas of haemorrhagic necrosis bordered by microvesicular steatosis with nuclear pyknosis and vacuolation (Figure 1B). The group with the extract (282 mg/kg) had liver tissue demonstrating a moderately altered hepato-architecture with microvesicular steatosis, nuclear pyknosis and vacuolation (Figure 1C), while the group treated with the extract (424 mg/kg) had liver tissue demonstrating a moderately altered hepato-architecture with normal hepatocytes and multiple areas of haemorrhagic necrosis with necrotic cells. There are presences of mononuclear inflammatory infiltrate (Figure 1D). Liver tissues of the group treated with n-hexane fraction showed moderately altered hepato-architecture with areas of hydrophic hepatocytes and focal area of microvesicular steatosis with nuclear pyknosis and vacuolation (Figure 1E), while the treated with DCM fraction had liver tissue showing a moderately altered hepato-architecture with areas of hepatocytes both normal and hydropic hepatocytes and interstitial inflammation (Figure 1F). There was also focal area of interstitial degeneration. Ethyl acetate fraction treated group had liver tissue demonstrating a moderately altered hepato-architecture with areas of normal hepatocytes and interstitial inflammation, with some showing ballooning degeneration (Figure 1G), while the methanol fraction treated mice had liver tissue demonstrating a moderately altered hepato-architecture with areas of hydrophic hepatocytes, focal area of interstitial degeneration and vacuolation (Figure 1H). Chloroquine treated mice had liver tissue demonstrating a moderately altered hepato-architecture with areas of normal hepatocytes and areas of haemorrhagic necrosis with necrotic cells. Mononuclear inflammatory infiltrate was also present (Figure 1I).

Table 2: Effect of the extract/fractions on haematological parameters

Treatment	Dose	WBC ($x10^9$/L)	NEUT. (%)	LYM (%)	MONO (%)	ESINO (%)	BASO (%)	RBC ($x10^{12}$/L)	HGB (g/dL)	PCV (%)	PLATELETS. ($x10^9$/L)
Control normal saline	10 mg/ ml	9.27± 1.95	59.00±1.60	35.66±1.65	4.66± 0.28	0.26± 0.21	1.00± 0.10	6.00± 0.28	12.13± 0.78	43.76±3.22	940.0± 35.76
Ethanol extract	141	11.60±1.56 [a]	51.86±3.16	45.86±2.06[a]	2.96±0.24 [a]	0.16 ±0.06 [c]	0.90±0.05	6.72±0.53	12.63± 0.56	43.20±2.36	709.3± 54.14[c]
	282	12.15±3.34 [b]	50.40±0.64[a]	45.30±0.88[a]	2.63±0.29[a]	0.76 ± 0.12[a]	1.10±0.20	7.83±0.78	14.10±1.25	53.76±7.61[a]	684.6± 92.13[c]
	424	10.86±1.66	57.93±0.60	36.53±4.27	3.76±0.38	0.66± 0.12[a]	0.50±0.00	7.17±0.18	12.10± 0.68	50.80±1.55[a]	1025.6± 61.50[c]
HF	282	9.90±1.51	59.50±0.60	34.90±1.44	4.96±0.03	0.43 ± 0.17	0.60±0.26	6.08±0.69	12.46± 0.08	49.50±0.40[a]	768.0± 82.92[c]
DCMF	282	9.32±2.31	48.10±14.13[b]	65.30±5.71[c]	3.40± 0.58	0.36±0.51	0.90 ± 0.05	6.65 ± 0.90	12.56±1.07	45.90±7.50	723.0± 35.80[c]
EAF	282	10.31±1.64	50.66±0.82[a]	43.46±0.73	3.60 ±1.01	0.30±0.51	0.50±0.17	7.28± 0.17	12.13±1.21	43.90±4.43	775.0±59.53[c]
MF	282	14.55±0.98[c]	47.60±6.45[b]	46.93±6.59[c]	4.16±0.72	0.40±0.15	0.53±0.03	6.70±0.63	13.33 ± 0.74	48.90±2.82	867.3±74.71[c]
Chloroquine	5	12.03±3.76[c]	33.16±0.79[c]	61.56±1.66[c]	2.23±0.28[a]	0.06±0.03[c]	0.55±0.03	7.56±0.31	12.66±1.18	47.90±2.82	888.0±47.05[c]

Data are expressed as MEAN ± SEM, Significant at [a]$P < 0.05$, [b]$P < 0.01$, [c]$P < 0.001$, when compared to control. (n=6). WBC-white blood cell, NEUT-Neutrophils-Lymphocytes, MONO-Monocytes, ESINO-Esinophils, BASO- Basophils, RBC- Red blood cell, HGB-Hemoglobin, PCV- Packed cell volume, HF-*n*-hexane fraction, DCMF-dichloromethane fraction, EAF-ethyl acetate fraction, MF-methanol fraction.

Table 3: Effect of the extract and fractions on liver function parameters.

Treatment	Dose (mg/kg)	Liver Function Parameters						
		AST (IU/L)	ALT (IU/L)	ALP (IU/L)	Total protein (g/L)	Total cholesterol (g/L)	Total bilirubin (µmol/mL)	Conjugated bilirubin (µmol/mL)
Control	-	24.66±3.28	16.06 ±1.08	24.80±4.01	61.66 ±2.02	3.10 ±0.55	5.36±0.42	3.13±0.23
Extract	141	28.0±2.88	14.70±0.23	23.40±1.62	55.0±1.73	2.96±0.26	5.83±0.61	3.73±0.47
	282	26.66±2.33	19.80±2.29	31.90±5.08	68.33±2.02	3.26±0.79	5.66±0.49	3.26±0.14
	424	17.33±1.45	15.90±1.49	28.56±3.73	70.0±1.73	2.86±0.40	4.03±0.14	2.40±0.20
NF	282	23.33±0.88	15.93 ±1.10	30.30±1.33	58.33±2.40	2.86 ±0.28	4.90±0.15	3.23±0.24
DCMF	282	30.33±2.40	17.50± 2.40	34.26±5.41	61.0 ±1.52	2.83 ±0.33	6.33±0.43	4.33±0.24
EAF	282	24.66±0.88	14.50± 0.75	30.0± 0.57	67.33 ±1.45	2.20 ±0.05	5.16±0.20	3.46±0.20
MF	282	33.66±1.45	16.43± 0.63	30.63± 1.17	71.0 ±2.08	3.66 ±0.23	6.83±0.37	4.73±0.26[a]
Chloroquine	5	37.00±2.08[a]	19.13 ±2.72	31.40±6.51	67.33±1.45	3.50±0.17	7.50±0.41	4.33±0.32

Values are expressed as mean ± SEM. Significant relative to control. [a]$P < 0.05$ n = 6.ALT- Alanine aminotransferase, Aspartate aminotransferase, ALP- Alkaline phosphatase, HF-*n*-hexane fraction, DCMF-dichloromethane fraction, EAF-ethyl acetate fraction, MF-methanol fraction.

Table 4: Effect of the extract and fractions on liver antioxidant enzymes

Treatment	Dose (mg/kg)	Antioxidant Parameters					Liver weight (g)
		GSH (µg/mL)	SOD (µg/mL)	CAT (µg/mL)	GPx (µm/mL)	MDA (µmol/mL)	
Control	-	1.24±0.31	0.45±0.04	6.20 ±0.56	0.055 ±0.014	0.42±0.02	2.65±0.10
Extract	141	1.06±0.22	0.61±0.01	5.86±0.53	0.047±0.008	0.17±0.0[b]	2.25±0.15
	282	0.64±0.13	0.47±0.04	5.44±0.22	0.029±0.004	0.38±0.06	2.28±0.16
	424	1.54±0.28	0.59±0.03	4.72±0.97	0.068±0.012	0.18±0.03[a]	2.26±0.12
HF	282	0.74±0.15	0.53 ±0.02	5.63±0.27	0.035±0.005	0.24±0.03[a]	2.21±0.18
DCMF	282	0.84±0.10	0.54± 0.02	5.30±0.37	0.038 ±0.004	0.21±0.01[a]	2.24±0.16
EAF	282	0.81±0.11	0.57± 0.04	5.23± 0.51	0.039 ±0.006	0.20±0.04[a]	2.39±0.13
MF	282	0.99±0.10	0.56± 0.02	5.31± 0.51	0.046 ±0.004	0.21±0.02[a]	2.16±0.10
CQ	5	1.06 ±0.22	0.37±0.06	4.67 ±0.74	0.047±0.010	0.46±0.06	2.22±0.18

Values are expressed as mean ± SEM. Significant relative to control. [a]P < 0.05; [b]P < 0.01; [c]P < 0.001. n = 6. SOD-superoxide dismutase, CAT- catalase, GPx- glutathione peroxidase, MDA- malondialdehyde and GSH-reduced glutathione. HF-*n*-hexane fraction, DCMF-dichloromethane fraction, EAF-ethyl acetate fraction, MF-methanol fraction, CQ-Chloroquine.

Table 5: Effect of the extract and fractions lipid profile

Treatment	Dose mg/kg	Total cholesterol (mMol/L)	Triglyceride (mMol/L)	HDL-C (mMol/L)	LDL-C (mMol/L)	VLDL (mMol/L)
Control	10 mL/kg	3.10 ±0.55	1.40±0.74	2.56±0.04	0.75±0.01	0.28±0.02
Chloroquine	5	2.96±0.26	1.28±0.16	2.26±0.03	0.71±0.01	0.22±0.01
Crude extract	141	3.26±0.79	2.15±0.64	2.14±0.01	0.72±0.01	0.28±0.01
	282	2.86±0.40	2.04±0.16	2.19±0.02	0.64±0.02	0.22±0.01
	424	2.86 ±0.28	1.86±0.11	2.35±0.0[c]	0.70±0.01	0.30±0.01
n-hexane fraction	282	2.83 ±0.33	1.05±0.06	2.30±0.12	0.68±0.05	0.20±0.01
DCM fraction	282	2.20 ±0.05	1.12±0.08	2.68±0.20	0.69±0.02	0.25±0.02
Ethyl acetate fraction	282	3.66 ±0.23	1.04±0.10	2.78±0.12	0.64±0.02	0.21±0.01
Methanol Fraction	282	3.50±0.17	1.28±0.16	2.88±0.14	0.79±0.01	0.23±0.01

Data are expressed as mean ± SEM, (n=6).

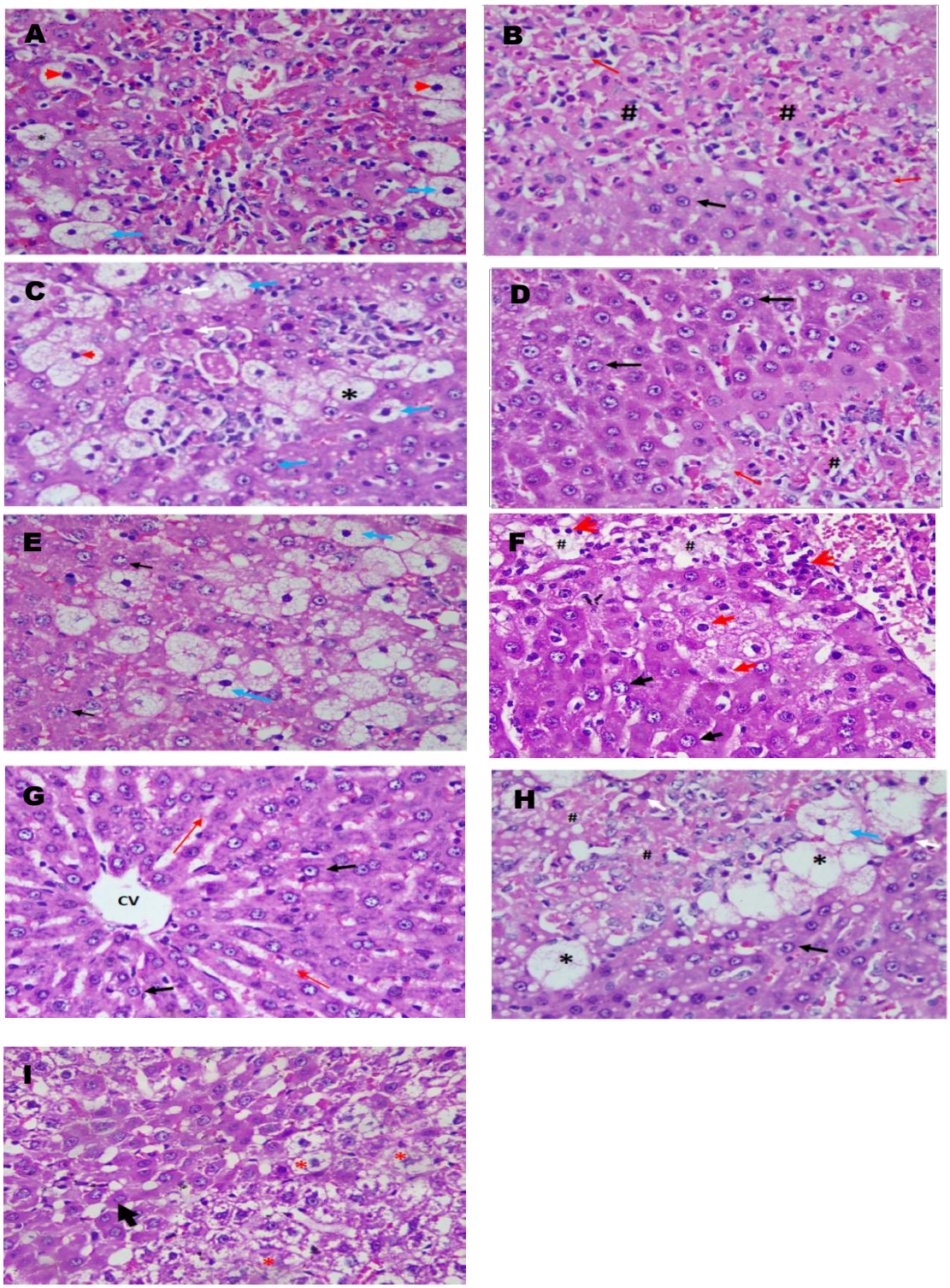

Figure 1: Liver sections of *Plasmodium berghei*-infected mice untreated with normal saline (A), stembark extract of A. *laxiflora*, 141 mg/kg (B), 282 mg/kg (C), 424 mg/kg (D), n-hexane fraction(E), DCM fraction (F), ethyl acetate fraction (G), methanol fraction (H) and chloroquine, 5 mg/kg (I) at magnification X400. Keys: normal hepatocytes (black arrow), multiple areas of haemorrhagic necrosis (#) bordered by microvesicular steatosis (blue arrow) with nuclear pyknosis (red arrowhead) and vacuolation (*), ballooning degeneration (Red asterisks), hydropic hepatocytes (red closed arrows) and interstitial inflammation (red open arrow), focal area of interstitial degeneration (#).

Discussion

The stem bark extract and fractions of *A. laxiflora* were investigated for antimalarial activity against rodent malaria parasite, *P. berghei* infection in mice using standard *in vivo* curative model. It was found that the extract and fractions significantly reduced the parasitaemia and prolonged the mean survival time of the mice, this activity could have resulted from plasmodicidal or plasmodistatic activity of the extract and fractions. These results corroborate previous reports on antimalarial activities of the leaf and root extracts of *A. laxiflora* (Okokon *et al.*, 2017a; Okokon *et al.,* 2017b). The inability of the extract/fractions to offer complete protection to the could have resulted from the doses used, short half-life or duration of action of the extract and fractions due to rapid biotransformation processes and subsequent elimination (Waako *et al.,* 2005).

The phytochemical screening of the extract revealed the presence of some pharmacologically active compounds such as tannins, flavonoids, alkaloids, terpenes, phenols among others. The previously reported phytoconstituents of the stem bark (Sanjo *et al*., 2011; Tapondjou *et al*.,2016) could be responsible for the activities of the extract and fractions. Some secondary metabolites of plants such as alkaloids, flavonoids and triterpenoids have been reported to have antiplasmodial properties (Kirby *et al.*, 1989; Philipson and Wright 1991; Christensen and Kharazmi 2001). Polyunsaturated fatty acids reported in the stembark extract have been implicated in the antiplasmodial activity (Suksamrarn *et al.,* 2005; Attioua *et al.*, 2007, Kumaratilake *et al.,* 1992; Krugliak *et al.,* 1995; Melariri *et al.,* 2011, 2012). Flavonoids have been shown to possess significant antiplasmodial activity against chloroquine sensitive and resistant strains of *P. falciparum* (Attioua *et al.,* 2011; Ganesh *et al.,* 2012; Ezenyi *et al.*, 2014). Antioxidant property of flavonoids has been suggested to be responsible for its antiplasmodial activity (Cimanga *et al.,* 2009; Ganesh *et al.,* 2012), as elevated free radical levels are common features of malaria disease and are implicated in severe malaria complications. Scavenging of these free radicals could be one of the mechanisms of action of this extract as the stem bark extract and fractions have been reported to exert strong antioxidant activity (Farombi *et al*., 2003; Oloyede *et al.,* 2010). The findings of this study suggest that stem bark of *A.laxiflora* possess antimalarial activity which is due to the activities of its phytochemical constituents.

The stembark extract and fractions did not significantly affect the liver function parameters. Temporary hepatic dysfunction is associated with malaria infection characterized by the increase of relative liver weight and liver enzymes activities. The distortions in liver may result from alteration in blood flow through the organ as parasitized RBC adhere to endothelial cells, blocking the sinusoids and obstructing the intrahepatic blood flow. However, administration of the extract and fractions did not affect total protein, albumin, AST, ALT, ALP, total and conjugated bilirubin. This suggests the inability of the extract and fractions to offer significant protection to the liver perhaps due to the low doses used. Increase in serum total protein, albumin, ALT, AST, ALP, direct and total bilirubin usually recorded in the parasitized non-treated mice are suggested to be due to response to hyper-parasitemia (Orhue *et al*., 2005). Malaria parasite infections are often accompanied by cellular mobilization of T-cells and its complements with a resultant synthesis and secretion of antibody molecules leading to elevated protein levels in parasitized non-treated mice (Orhue *et al*., 2005). The increased activities of serum AST, ALT and ALP in the serum of *P. berghei* infected mice may be due to hepatic dysfunction (Guthrow *et al*., 2007; George *et al.,* 2011) or hepatic damage as could be confirmed in the liver histology, which indicates lack of hepatoprotective activity. In this study, the infected mice showed a significant general architectural disorganization of liver that attenuated following extract and fractions treatments which confirm the hepatoprotective activity of the stembark extract.

Obstruction of hepatic blood and blockade of sinosoids by parasitized erythrocytes in addition to destruction of the liver cells and membranes integrity by the activities of generated free radicals during malaria infection are known to cause injuries to the liver, thereby leading to leakages of cellular enzymes such as transaminases. Reticuloendothelial blockage and disturbance of hepatocyte microvilli compromised the secretory capacity in the liver thereby resulting in hyperbilirubinemia (Onyesom and Onyemakonor, 2011). This could have occurred in this study, and the extract and fractions were unable to ameliorate it.

Oxidative stress contributes to complications associated with malaria infection such as anemia, jaundice and pre-eclampsia (Fabbri *et al.,* 2013; Sarr *et al.,* 2017). Large number of free radicals are generated due to the hypoxic condition caused by malaria infection which triggers body immune responses (Becker *et al.,* 2004; Percario *et al.,* 2012), leading to pathogenesis and development of systemic complications caused by malaria (Guha *et al.,* 2006; Ojezele *et al.,* 2017). Malarial infection has been observed to reduce the levels of antioxidant enzymes and other non-enzymatic anti-oxidants such as catalase (CAT), glutathione (GSH) peroxidase, superoxide dismutase (SOD), albumin, glutathione, ascorbate and plasma tocopherol (Asagba *et al*., 2010). Malaria severity has been correlated with increased lipid peroxidation and malondialdehyde levels (Asagba *et al*., 2010; Adil *et al*., 2013). During malaria infection, reduced activities of antioxidant enzymes (catalase and SOD), and increased lipid peroxidation (MDA) are observed (Luersen *et al*., 2000). These indices have been used to measure the severity

of malaria infection. In this study, the extract and fractions did not significantly affect some oxidative stress markers except GST activity which was significantly increased by both extract and fractions. This could have been due to the doses administered in this study. The extract and fractions were able to scavenged free radical generated by enhancing GST activity thereby reducing the level of MDA thus lipid peroxidation.

The alteration in lipid metabolism has been attributed to acute phase response to the infection (Memon *et al*., 2000). However, treatment with the extract and fractions did not significantly ($P > 0.05$) on serum lipid profile. This s suggest that the plant may not possess hypolipidemic potential at the doses employed in this study.
Haematological alterations resulting in the decrease of RBC count, Hb level, PCV, and mean haemoglobin concentration levels are often observed in infected animals as conventional signs of anaemia (Surve *et al.,* 2017). The extract and fractions did not alter significantly the haematological parameters of the treated mice., except significant elevation of WBC count and reduction of platelet count, neutrophil, lymphocytes, monocytes and eosinophil However, the PCV of the treated infected mice were significantly elevated. This implies that the extract has erythropoietic potentials to alleviate anemia. The significant increase in WBC as reported previously in malaria infection (Guyton, 2007), is attributable to immunogenic response to the parasite and malaria pigment (hemozoin) (Malaguarnera *et al.*, 2002).

Conclusion

The results of this study show that the stembark of *Alchornea laxiflora* possesses anti-malarial and, anti-plasmodial activities and a weak anti-oxidative stress and hepato protective activities.

Conflict of Interest

Authors declare no conflicts of interest

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References

Adil R, Sumit K, Varshney S, Shahid MK *et al* (2013). Lipid peroxidation in cerebral malaria and role of antioxidants. *International Organization of Scientific Research Journal of Pharmacy* 3(1): 15-18.

Agrawal,H. S. Tsay, L. F. Shyur, Y. C. Wu, and S. Y. Wang (Singapore: Springer),487–510.

Asagba SO, Eriyamremu GE, George BO. Okoro I (2010). Biochemical indices of severity in human malaria. *Journal of Medical Sciences* 10(4): 87-92. *https://doi.org/* 10.3923/jms.2010.87.92

Atanu O, Idih FM, Nwonuma CO, Hetta, HF *et al* (2021). Evaluation of antimalarial potential of extracts from *Alstonia boonei* and *Carica papaya* in *Plasmodium berghei*-infected mice. *Evidence-Based Complementary and Alternative Medicine,* Article ID 2599191, 2021;11. *https://doi.org/* 10.1155/2021/2599191.

Attioua B, Lagnikab L, Yeoc D, Antheaumed, C *et al* (2011). In vitro antiplasmodial and antileishmanial activities of flavonoids from *Anogeissus leiocarpus* (Combretaceae). *International Journal of Pharmaceutical Sciences Review and Research* 11:1–6.

Attioua B, Weniger B, Chabert P (2007). Antiplasmodial activity of constituents isolated from *Croton lobatus*. *Pharmaceutical Biology* 45:263–266. https://doi.org/10.1080/13880200701214607

Becker K, Tilley L, Vennerstrom JL, Roberts D *et al* (2004). Oxidative stress in malaria parasite-infected erythrocytes: Host-parasite interactions. *International Journal of Parasitology* 34(2): 163-189. *https://doi.org/* 10.1016/j.ijpara.2003.09.011.

Bero J, Quetin-Leclercq, J. (2011). Natural products published in 2009 from plants traditionally used to treat malaria. *Planta Medica*, 2011; 77(06): 631- 640. *https://doi.org/* 10.1055/s-0030-1250405.

Buffet PA, Safeukui I, Deplaine G, Brousse V *et al* (2010). The pathogenesis of *Plasmodium falciparum* malaria in humans: Insights from splenic physiology. *Blood,* 2010; 4: 202911.*https://doi.org/* 10.1182/blood-2010-04-202911.

Burkill HM. (1985). The useful plants of west tropical Africa 2nd Ed. Vol. 1, *Royal Botanic Garden* K. E.W. 960.

Christensen SB, Kharazmi A. (2001). Antimalarial natural products. Isolation, characterization and biological properties. In: Tringali C, editor. *Bioactive compounds from natural sources: isolation, characterization and biological properties*. London: Taylor & Francis; p. 379–432.

Cimanga RK, Tona GL, Kambu OK, Mesia GK *et al* (2009). Antimalarial, antiamoebic and cytotoxic activities of some extracts and isolated constituents from the leaves of Morinda morindoides (Baker) Milne- Redh. (Rubiaceae). *Rec Progr Med Plants* 25:225–242.

Ellman GL. (1959). Tissue sulphydryl groups. *Archives of Biochemistry and Biophysics* 82(1): 72-77. *https://doi.org/10.1016/0003-9861(59)90090-6.*

Elufioye TO. (2017). Memory-enhancing and memory-related beneficial effects of selected medicinal plants from the Nigerian flora, in Medicinal plants and fungi: Recent advances in research and development. Editors D.

EnyiekereVJ, Anagboso MO, Ise UP, Okokon JE, *et al*. (2024). Effect of *Justicia insularis* leaf extract and fractions on oxidative stress markers, Liver function parameters and Liver histology of *Plasmodium berghei*-infected mice. *Biology, Medicine, & Natural Product Chemistry* 13 (2), 351-359.

Esterbauer H, Cheeseman KH. (1990). Determination of aldehydic lipid peroxidation products: malonaldehyde and 4-hydroxynonenal. *Methods Enzymology* 186: 407–421.*http://dx.doi.org/ 10.1016/0076-6879(90)86134-h.*

Ezenyi IC, Salawu OA, Kulkarni R, Emeje M. (2014). Antiplasmodial activity aided isolation and identification of quercetin-40-methyl ether in *Chromolaena odorata* leaf fraction with high activity against chloroquine-resistant *Plasmodium falciparum. Parasitology Research* 113:4415–4422.*https://doi.org/*10.1007/s00436-014-4119-y.

Fabbri C, de Cássia Mascarenhas-Netto R, Lalwani P, Melo GC *et al* (2013). Lipid peroxidation and antioxidant enzymes activity in *Plasmodium vivax* malaria patients evolving with cholestatic jaundice. *Malaria Journal* 12: 315. *https://doi.org*/10.1186/1475-2875-12-315.

Farombi EO, Ogundipe OO, Uhunwangho ES, Adeyanju MA *et al*. (2003). Antioxidant properties of extracts from *Alchornea laxiflora* (Benth) Pax and Hoffman. *Phytotherapy Research* 17, 713–716. *https://doi.org/* 10.1002/ptr.1050

Ferreira JF, Luthria DL, Sasaki T, Heyerick A (2010). Flavonoids from *Artemisia annua* L. as antioxidants and their potential synergism with artemisinin against malaria and cancer. *Molecules* 15:3135–3170.*https://doi.org/*10.3390/molecules15053135.

Friedewald WT, Levy RT, Frederickson DS (1972). Estimation of the concentration of low lipoprotein cholesterol in plasma without use of preparative ultracentrifuge. *Clinical Chemistry* 18: 499 – 502.*PMID: **4337382***

Ganesh D, Fuehrer H, Starzengruber P, Swoboda P *et al*. (2012). Antiplasmodial activity of flavonol quercetin and its analogues in *Plasmodium falciparum*: evidence from clinical isolates in Bangladesh and standardized parasite clones. *Parasitology Research* 110:2289–2295. *https://doi.org/* 10.1007/s00436-011-2763-z.

George BO, Osioma E, Okpoghono J1, Aina OO (2011). Changes in liver and serum transaminases and alkaline phosphatase enzyme activities in *Plasmodium berghei* infected mice treated with aqueous extract of *Aframomum sceptrum*. *African Journal of Biochemistry Research* 5(9): 277-281.

Gora D, Sandhya M, Shiv G, Praveen S (2006). Oxidative stress, α-tocopherol, ascorbic acid and reduced glutathione status in schizophrenics. *Indian Journal of Clinical Biochemistry* 21: 34–38. *https://doi.org/* 10.1007/BF02912908

Guha M, Kumar S, Choubey V, Maity P, Bandyopadhya U (2006). Apoptosis in liver during malaria: role of oxidative stress and implication of mitochondrial pathway. *Federation of American Societies for Experimental Biology Journal* 20, 439-449. *https://doi.org/* 10.1096/fj.05-5338fje.

Guthrow CE, Morris JF, Day JW (2007). Enhanced non-enzymatic glycosylation of human serum albumin. *Quart. T. Med.,* pp. 30-38.

Guthrow CE, Morris MA, Day JF, Thorpe SR *et al* (1979). Enhanced nonenzymatic glucosylation of human serum albumin in diabetes mellitus. *Procedings of the National Academy of Sciences United States of America* 76(9): 4258-4261.*https://doi.org/* 10.1073/pnas.76.9.4258.

Guyton A, Hall J (2007). Textbook of medical physiology. 12th ed. Philadelphia; 2007.

Jain NK, Tailang M, Kumar S, Chandrasekaran B, *et al* (2022). Appraising the therapeutical potentials of *Alchornea laxiflora* (Benth.) Pax & K.Hoffm., an underexplored medicinal herb: A systematic review. *Frontier in Pharmacology* 13:958453. *https://doi.org/* 10.3389/fphar.2022.958453

Khan SL, Siddiqui FA (2020). Beta-Sitosterol: As Immunostimulant, Antioxidant and Inhibitor of SARS-CoV-2 Spike Glycoprotein. *Archives of Pharmacology and Therapeutics 2(1):12-16.* https:// doi.org/ 10.33696/ Pharmacol 2.014

Kirby GC, O'Neill MJ, Phillipson JD, Warhurst DC (1989). *In vitro* studies on the mode of action of quassinoids with against chloroquine resistant *Plasmodium falciparum*. *Biochemistry and Pharmacology* 38: 4367–4374.*https://doi.org/* 10.1016/0006-2952(89)90644-8.

Kirmizibekmez H, Calis I, Perozzo R, Brun R, *et al* (2004). Inhibiting activities of the secondary metabolites of *Phlomis brunneogaleata* against parasitic protozoa and plasmodial enoyl-ACP reductase, a crucial enzyme in fatty acid biosynthesis. *Planta Medica* 70: 711. *https://doi.org/* 10.1055/s-2004-827200.

Kohno Y, Egawa Y, Itoh S, Nagaoka S*, et al* (1995). Kinetic study of quenching reaction of singlet oxygen and scavenging reaction of free radical by squalene in n-butanol. *Biochimica et Biophysica Acta* 1256:52–56. *https://doi.org/* 10.1016/0005-2760(95)00005-w

Krishna AP, Chandrika Suchetha K, Manasa A*, et al.* (2009). Variation in common lipid parameters in malaria infected patients. *Indian Journal of Physiology and Pharmacol*ogy 53(3): 271-274.PMID: **20329375.**

Krugliak M, Deharo E, Shalmiev G, Sauvain M, *et al* (1995). Antimalarial effects of C18 fatty acids on *Plasmodium falciparum* in culture and on *Plasmodium vinckei petteri* and *Plasmodium yoelii nigeriensis* in vivo. *Experimental Parasitology* 81: 97-105. https: //doi. org/10. 1006/ expr. 1995. 1097

Kumaratilake LM, Robinson BS, Ferrante A, Poulos A (1992). Antimalarial properties of n-3 and n-6 polyunsaturated fatty acids: in vitro effects on *Plasmodium falciparum* and in vivo effects on *P. berghei. Journal of Clinical Investigation* 89: 961–967. *https://doi.org/*10.1172/JCI115678.

Lawrence RA, Burk RF (1976). Glutathione peroxidase activity in selenium- deficient rat liver. *Biochemical and Biophysical Research Communications* 71: 952-958. *http://dx.doi.org/10.1016/0006-291x(76)90747-6*

Lorke D (1983). A new approach to practical acute toxicity testing. *Archieves of Toxicology* 54: 275-286. *https://doi.org/* 10.1007/BF01234480

Lüersen K, Walter RD, Müller, S (2000). *Plasmodium falciparum*-infected red blood cells depend on a functional glutathione de novo synthesis attributable to an enhanced loss of glutathione. *Biochemistry Journal 346*(2): 545-552.PMID: 10677377

Lui KC, Yang SC, Roberts MF (1992). Antimalarial activity of *Artemisia annua* flavonoids from whole plants and cell cultures. *Plants Cell II*: 637–*https://doi.org/* 40.10.1007/BF00236389.

Malaguarnera L, Musumeci S (2002). The immune response to *Plasmodium falciparum* malaria*. Lancet Infectious Diseases* 2(8): 472478.*https://doi.org/* 10.1016/s1473-3099(02)00344-4.

Marklund S, Marklund G (1974). Involvement of superoxide anion radical in the autooxidation of pyrogallol and a convenient assay for superoxide dismutase. *Euroupean Journal of Biochemistry* 47: 469 - 474. https://doi.org/ 10.1111/j.1432-1033. 1974.tb03714.x

Mbaveng AT, Sandjo LP, Tankeo SB, Ndifor AR *et al* (2015). Antibacterial activity of nineteen selected natural productsagainst multi-drug resistant Gram-negative phenotypes. *Springerplus* 4: 823. *https://doi.org/* 10.1186/s40064-015-1645-8

Melariri P, Campbell W, Etusim P, Smith P (2011). Antiplasmodial properties and bioassay-guided fractionation of ethyl acetate extracts from *Carica papaya* leaves. *Journal of Parasitology Research* Article ID 104954, 7pages. *https://doi.org/* 10.1155/2011/104954.

Melariri P, Campbell W, Etusim P, Smith P (2012). In vitro and in vivo antimalarial activity of linolenic and linoleic acids and their methyl esters. *Advanced Studies in Biology* 4:333–349.

Memon RA., Staprans I, Noor M, Holleran WM *et al.* (2000). Infection and inflammation induce LDL oxidation *invivo*. *Arteriosclerosis, Thrombosis and Vascular Biol*ogy 20: 1536-1542. *https://doi.org/* 10.1161/01.atv.20.6.1536.

Nathawut S, Paveena Y, Srivicha K, Wattana L *et al* (2004). Increased fluidity and oxidation of malarial lipoproteins: Relation with severity and induction of endothelial expression of adhesion molecules. *Lipids in Health and Diseases* 3: 15. *https://doi.org/* 10.1186/1476-511X-3-15.

Odetola A, Basir O (1980). Evaluation of antimalarial properties of some Nigerian medicinal plants, pp275–283. *In:* A. Sofowora (Editor). *Proceeding of African Bioscience Network, Federal Ministry of Science and Technology, Nigerian Society of Pharmacology and Drug Research and Production Unit*. University of Ife, Ile-Ife.

Ojezele MO, Moke EG, Onyesom, I (2017). Impact of generic antimalarial or *Phyllanthus amarus* and vitamin co-administration on antioxidant status of experimental mice infested with *Plasmodium berghei*. *Beni-Suef University Journal of Basic and Applied Science* 6(3): 260-265.

Oloyede GK. Onocha PA, Soyinka J, Oguntokun OW *et al* (2010). Phytochemical screening, antimicrobial and antioxidant activities of four Nigerian medicinal plants. *Annals of Biolology Research* 1; 114–120.

Okokon JE, Nkemnele BA, Mohanakrishnan (2017a). Antimalarial, antiplasmodial and analgesic activities of root extract of *Alchornea laxiflora. Pharmaceutical Biology* 55: 10221031.*https://doi.org/*10.1080/13880209.2017.128594 7.

Okokon JE, Obot AU, Mittal G, Mohanakrishnan D. *et al* (2017b). Antimalarial and antiplasmodial activity of leaf extract of *Alchornea laxiflora. Journal of Herbs, Spices and Medicinal Plants* 23(2): 128 -141. *https://doi.org/*10.1080/10496475.2016.1275072

Okokon JE, Udoh AE, Osigwe CC, Uwaeme UF *et al* (2025). Analgesic activity of ethanol stembark extract of *Alchornea laxiflora. Asian Journal of Biochemistry, Genetics and Molecular Biology* 17(6): 22-29. https://doi.org/10.9734/ajbgmb/2025/v17i6470.

Okokon, JE, Nwafor PA. (2010). Antiplasmodial activity of root extract and fractions of *Croton zambesicus*. *Journal of Ethnopharmacology.* 121:74-78.

Okokon JE, Opara KN, Udobang JA, Bankhede H. (2019). In vivo antimalarial and antipyretic activities of leaf extract of *Ananas comosus. Tropical Journal of Natural Product Research.* 3(7): 240 -245.

Okokon JE, Davis K, Azare B, Okokon P. (2016). Analgesic and antimalarial activities of ethanol root extract of *Panicum maximum. African Journal of Pharmacology and Therapeutics.* 5(3): 128 -135.

Onyesom I, Onyemakonor N (2011). Levels of parasitaemia and changes in some liver enzymes among malarial infected patients in Edo-Delta Region of Nigeria. *Current Research Journal of Biological Science* 3(2): 78-81.

Orhue NEJ, Nwanze EAC, Okafor A (2005). Serum total protein, albumin and globulin levels in Trypanosoma *brucei*-infected rabbits: Effect of orally administered *Scoparia dulcis*. *African Journal of Biotechnology* 4(10): 1152-1155. *https://doi.org/* 10.4314/ajb. v4i10.71355

Oyedeji AA, Angaye TCN,Abowei JFN (2024). Evaluation of *Alchornea cordifolia* extracts for larvicidal activity against Anopheles. Implications for Malaria Control. *UMYU Scientifica* 3(1), 235 – 240.

Percário S, Moreira DR, Gomes BAQ, Ferreira MES et al (2012). Oxidative stress in malaria. *International Journal of Molecular Sciences* 13(12): 16346-16372. *https://doi.org/* 10.3390/ijms131216346

Peters W, Robinson BL (1992). The chemotherapy of rodent malaria. Studies on pyronaridine and other Mannich base antimalarials. *Annals of Tropical Medicine and Parasitology* 86, 455–465. *https://doi.org*/10.1080/ 00034983.1992.11812694.

Philipson JD, Wright CW (1991). Antiprotozoal compounds from plants sources. *Planta Medica* 57: 553–559. *https://doi.org*/10.1055/s-2006-960230.

Ponnamma SU, Manjunath K (2012). GC-MS analysis of phytocomponents in the methanolic extract of *Justicia wyaadensis* (NEES) T. Anders. *International Journal of Pharmaceutical and Biological Sciences* 3: 570-576.

Ryley JF, Peters W (1970). The antimalarial activity of some quinolone esters. *Annals of Tropical Medicine and Parasitology* 64(2): 209–222. *https://doi.org/* 10.1080/00034983.1970.11686683.

Saganuwan S, Onyeyili P, Ameh E, Etuk U (2011). In vivo antiplasmodial activity by aqueous extract of Abrus. *Revista Latinoamericana de Química*, 39(1-2): 32-44.

Sandjo LP, Poumale HMP, Siwe XN, Ntede HN *et al.* (2011). Two new fatty acid derivatives from the stem bark of *Alchornea laxiflora* (*E*uphorbiaceae). *Journal of American Oil and Chemical Society* 88: 1153–1159. https://doi.org/10.1007/s11746-011-1770-7

Sarr D, Cooper CA, Bracken TC, Martinez-Uribe O *et al* (2017). Oxidative stress: A potential therapeutic target in placental malaria. *Immuno Horizon* 1(4): 29-41. *https://doi.org/* 10.4049/immunohorizons.1700002
.

Sinha AK (1972). Colorimetric assay of catalase. *Analytical Biochemistry* 47: 389 - 94. https://doi.org/ 10.1016/0003-2697(72)90132-7

Sofowora A (1993). *Medicinal Plants and Traditional Medicine in Africa.* 2nd edn, Spectrum Book Ltd, Ibadan, Nigeria.

Suksamrarn A, Buaprom M, Udtip S, Nuntawong N, *et al,* (2005). Antimycobacterial and antiplasmodial unsaturated carboxylic acid from the twigs of *Scleropyrum wallichianum*. *Chemical and Pharmaceutical Bulletin* 53: 1327–1329. *https://doi.org/* 10.1248/cpb.53.1327.

Surve K, Kulkarni A, Rathod S, Bindu R (2017). Study of haematological parameters in malaria. *International Journal of Research in Medical Science* 5(6): 2552-2557. *https://doi.org/* 10.18203/2320-6012.ijrms20172446

Tapondjou LA, Jeuett-Siems K, Siems K, Melzig MF (2016). Alchornealaxine, an unusual prenylguanidinyl-epicatechin derivative from *Alchornea laxiflor*a (benth) Pax and hoffman. *Records of Natural Products* 10: 508–512.

Teffo LS, Aderogba MA, Eloff JN (2010). Antibacterial and antioxidant activities of four kaempferol methyl ethers isolated from *Dodonaea viscose* Jacq. Var. *angustifolia* leaf extracts. *South Africa Journal of Botany* 76: 25–29. https://doi.org/10.1016/j.sajb.2009.06.010

Trease GE. Evans WC (2002). *Pharmacognognosy.* 15th Edition, Bailierr Tindall, pp. 683-684.

Tietz NW (1995). *Clinical Guide to Laboratory Tests*. 3rd Edition. W. B. Saunders, Philadelphia, 76p.

Waako PJ, Gumede B, Smith P Folb PI (2005). The *in vitro* and *in vivo* Antimalarial activity of *Cardiospermum halicacabum* L. and *Momordica foetida* Schumch. et Thonn. *Journal of Ethnopharmacology* 99(1): 137-143. *https://doi.org/* 10.1016/j.jep.2005.02.017.

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