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Effect of *Telfairia occidentalis* Stem Extract and Fractions on Parasitaemia, Oxidative Stress Markers, Lipid Profile, Hematological Parameters, Liver Function Indices and Liver Histology in *Plasmodium berghei* Infected Mice

Jude Efiom Okokon [a*], Chinyelu C. Osigwe [b], Ukeme Essien Andrew [a], Uwaeme Ugonma Florence [b], John A Udobang [c] and Uduak P. Ise [d]

[a] Department of Pharmacology and Toxicology, Faculty of Pharmacy, University of Uyo, Uyo, Nigeria.
[b] Department of Pharmacology and Toxicology, Faculty of Pharmacy, Madonna University, Elele, Rivers State, Nigeria.
[c] Department of Clinical Pharmacology and Therapeutics, Faculty of Basic Clinical Sciences, University of Uyo, Uyo, Nigeria.
[d] Department of Pharmacology and Toxicology, Faculty of Pharmacy, Bingham University, Karu, Nigeria.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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**Corresponding author: E-mail: judeefiom@yahoo.com;*

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Original Research Article

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ABSTRACT

Telfairia occidentalis Hook (*Cucurbitaceae* family) is a pumpkin widely used for dietary purposes and in traditional healthcare by Ibibios of Nigeria, as remedy for various diseases such as malaria, diabetes, pains among others. *Telfairia occidentalis* stem extract and fractions activities were assessed in mice for effect on parasitaemia, hematological parameters, oxidative stress, lipid profile, liver function parameters and liver histopathology in *Plasmodium berghei*-infected mice employing curative test method. The stem extract (200- 600 mg/kg, p.o.) demonstrated significant ($p<0.05$) dose-related antiplasmodial activity against *P. berghei* infection and n-hexane fraction was the most potent fraction. The extract/fractions administration showed no effect on any blood parameters except elevation of WBC counts and neutrophil percentages in groups administered with 400 and 600 mg/kg of the extract. The extract/fractions administration had no significant ($p<0.05$) effect on indices of lipid profile, oxidative stress and liver function of the infected mice relative to control, although there were observable changes. Histology of liver sections revealed moderate decreases in the number of pathological signs observed in the treated infected mice especially in mice groups administered with extract (200,400 and 600 mg/kg) and hexane fraction respectively relative to control infected mice. These findings demonstrate that the stem extract/fractions of *Telfairia occidentalis* can exert moderate antioxidative stress and liver protective activities due to the low doses used and activities of its phytochemical constituents which could be beneficial in malaria therapy. Further study with higher doses of the extract is recommended.

Keywords: Malaria; Telfairia occidentalis; Plasmodium berghei; oxidative stress; liver protective.

1. INTRODUCTION

Malaria has continued to ravage human population especially the developing countries despite the various control and treatment modalities available (WHO, 2022; WHO, 2023). A significant number of countries (84) still bear serious burden of malaria regardless of the prominent fights against the disease (WHO, 2022; Center for Disease Control and Prevention, 2023). The recent reports documented that menace from malaria are increasing over the years; from 227 million cases, 568, 000 deaths in 2019 (WHO, 2020), to 249 million cases, 608,000 deaths in 2022 (WHO, 2023), accounting for significant number of deaths over the years. Worst still, human population in sub-Saharan Africa bear the greater burden of malaria than the rest of the world as a child, mostly under 5 years, dies of malaria every minute (WHO, 2022; WHO, 2023; UNICEF, 2024). Pregnant women in sub-Saharan Africa are equally highly affected (WHO, 2023). Oxidative stress associated with malaria infection contributes to development of systemic complications which leads to organ dysfunctions (Ojezele et al*.,* 2017). Plants, therefore, provide an alternative store for antimalarial compounds noted for being beneficial in providing many therapeutic effects and are safe.

Telfairia occidentalis Hook is a fluted pumpkin of the *Cucurbitaceae* family widely consumed as food in Nigeria (Okokon et al., 2009). It is a popular vegetable all over Nigeria, especially in the Niger-Delta region and the Eastern part of the country; varieties of meals are prepared from the leaves, stem and seeds of the plant (Usunomena and Okpiabhele, 2023). The various parts of the plant (seeds, leaves and stem) are used as remedies for various ailments and diseases. Antimalarial potentials of the seed, leaves and roots of the plant have been documented (Okokon et al*.,* 2007; Okokon et al*.,* 2009). Enin et al*.* (2023) had reported on antioxidant activity and in vivo inhibitory effect on alpha amylase and alpha glucosidase of the stem extract. Some polyunsaturated fatty acids have been identified in the various fractions as well as alkaloid, terpenes, saponin, flavonoid and tannin in the crude extract (Enin et al*.*, 2023). This study investigated the activities of stem extract and fractions of *T. occidentalis* on

oxidative stress, liver function and liver histology in *Plasmodium berghei* infected mice.

2. MATERIALS AND METHODS

2.1 Plant Materials

Fresh stems of *Telfairia occidentalis* were collected from farms in Uyo metroplis in Uyo LGA, Akwa Ibom State, Nigeria. Identification and authentication of the leaves as *Telfairia occidentalis* were carried out by a taxonomist in the Department of Botany and Ecological studies, University of Uyo, Uyo, Nigeria and a voucher specimen was preserved at the herbarium of the Department of Pharmacognosy and Natural Medicine, Faculty of Pharmacy, University of Uyo.

2.2 Extraction

Freshly collected stems of *Telfairia occidentalis* were washed, chopped to small pieces and shade-dried for 14 days. The dried stems were further powdered and the powder (2 kg) was soaked in ethanol (50%) at room temperature for 3 days. Thereafter, filtered and concentrated to dryness in *vacuo* 40 ˚C using a rotary evaporator (BuchiLab Switzerland). The crude extract (50.0 g) was partitioned in n-hexane, dichloromethane, ethyl acetate and methanol after dissolving in water to give the corresponding fractions of these solvents. The weighed extract and fractions were preserved in a refrigerator at -4 ˚C, and used for the proposed experiments.

2.3 Experimental Animals

Animals (Swiss albino mice of both sexes) used for these experiments were gotten from Animal house of University of Uyo. The mice were accommodated in standard plastic cages and fed on standard pelleted diet and water *ad libitum*. The care and use of animals was conducted in accordance with the National Institute of Health Guide for the Care and Use of laboratory Animals (NIH Publication, 1996). University of Uyo's Animal Ethics Committee approved the study.

2.4 Drug Administration

All the drugs; extract, fractions and chloroquine used in these experiments were given orally using a stainless metallic feeding cannula.

2.5 Determination of Median Lethal Dose (LD_{50})

Lorke (1983) method was used to estimate the median lethal dose (LD_{50}) of the extract orally using different extract doses (100 -1000 mg/kg) administered to groups of three mice each. Toxicity manifestation in mice were observed. These included writhing, decreased motor activity, decreased body/limb tone, decreased respiration and death. Any death observed 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 (b).

$LD_{50} = \sqrt{ab}$.

2.6 Micro-organism (parasite)

The strain of parasite used in this study was chloroquine-sensitive *Plasmodium berghei* strain gotten from National Institute of Medical Research (NIMER), Yaba Lagos, Nigeria and maintained by subpassage in mice.

2.7 Parasite Inoculation

A portion (0.2 mL) of infected blood containing about 1 x 10^7 *P. berghei* parasitized erythrocytes gotten from an infected mouse with 20-30% parasitaemia was inoculated into all the mice used in this experiment. The inoculum was made up of blood containing 5 x 10^7 *P. berghei* infected erythrocytes per milliliter prepared as earlier reported in previous works (Okokon et al*.,* 2019; Atanu et al*.,* 2021; Okokon et al*.,* 2022a). Monitoring of parasitemia using standard methods of preparation of thin blood smears on glass slides, fixed and stained using Giemsa stain was carried out, and counting of parasitemia was done under the microscope. Parasitaemia was calculated according to the formula of Enyiekere et al (2024a) as given below:

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

2.8 Assessment of Antimalarial Activities of Stem Extract and Fractions of *Telfairia occidentalis* on Established Infection

This investigation was done by employing curative test method previously reported by

Okokon et al. (2017) and Okokon et al. (2019), to assess the antimalarial activity of the extract, fractions and chloroquine in confirmed plasmodiasis. The experiment was carried out by intraperitoneal injection of *P. berghei* parasites on the first day (D_0) into ninety (90) mice, which were shared into nine groups of ten mice each after 3 days (D_2). The extract was orally given to mice in groups 1, 2 and 3 at doses of 200, 400, and 600 mg/kg respectively, while mice in groups 4 -7 were orally treated respectively with *n*-hexane, ethyl acetate, dichloromethane, and methanol fractions at a dose of 400 mg/kg. Chloroquine (5 mg/kg) was orally administered to mice in group 8 (positive control) and mice in negative control group 9 was given 10 mL/kg distilled water. The various daily treatments lasted for 5 days. Prepared tail blood thin smears, stained with Giemsa stain for the 5 days of treatments were used to monitor the parasitemia level. Five mice from each experimental group were sacrificed after being anaesthesized with light diethyl ether vapour on day 8. Collection of blood samples was done through cardiac puncture into EDTA bottles and plain centrifuge tubes. Blood samples collected into the EDTA bottles were used in hematological analyses, while biochemical analyses like liver function test and lipid profile were determined in sera samples separated from blood samples collected into plain tubes after they were centrifuged at 2500 rpm for 15 mins, which were stored at -20^{o}C. Livers were harvested from mice in their respective groups and weighed before being divided into two parts. The parts used for histological study were fixed in 10% formaldehyde and the other parts used in assay of oxidative stress markers were stored in ice cold normal saline. The average suppression of parasitemia was calculated using the formula earlier reported by Enyiekere et al. (2024a). The survival time of each mouse used in the experimental groups was monitored over a period of 29 days (D_0-D_{28}) and the mean survival time (MST) of each group calculated.

2.9 Effect of the Stem Extract and Fractions of *T. occidentalis* on Haematological Parameters of *P. berghei* Infected Mice

The hematological parameters analysed at the University of Uyo Teaching Hospital using automated hematological system (Sysmex Hematology – Coagulation system, Model MO-1000 I, Trans Asia, Japan) included Red blood cell count (RBC), hemoglobin, (Hb), packed cell volume (PCV), platelet concentration (PLC) and total and differential white blood cell count (WBC). These parameters were used to investigate the effect of the extract/fractions in the infected mice.

2.10 Effect of the Stem Extract and Fractions of *T. occidentalis* on Liver Function Parameters of *P. berghei* Infected Mice

The parameters used to assess the effect of the extract/fractions on liver functions of the infected mice included; total protein, albumin, aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), conjugated and total bilirubin. These were estimated spectrophotometrically in the stored sera samples using Randox analytical kits according to standard methods as instructed by the manufacturer (Tietz, 1990).

2.11 Effect of the Stem Extract and Fractions of *T. occidentalis* on Liver Oxidative Stress Markers of *P. berghei* Infected Mice

The homogenates of stored liver samples were prepared after washing with ice cold 0.9% NaCl in a ratio of 1 g of wet tissue to 9 ml of 1.25% KCl using motor driven Teflon-pestle. Supernatants separated after centrifugation of the homogenates at 7000 rpm for 10 min at 4˚C were used for the assays of glutathione peroxidase (GPx) (Lawrence and Burk,1976), superoxide dismutase (SOD) (Marklund and Marklund, 1974), catalase (CAT) (Sinha,1972), reduced glutathione (GSH) (Ellman,1959) and malondialdehyde (MDA) (Esterbauer and Cheeseman,1990).

2.12 Effect of the Stem Extract and Fractions of *T. occidentalis* on Lipid Profile of *P. berghei* Infected Mice

Standard colorimetric methods were used to determine the various lipid profile indices of the mice such as cholesterol, triglyceride and high-density lipoprotein (HDL) levels. Fortress Diagnostic Kits® were used for these assays according to the instructions of the manufacturer spectrophotometrically. Friedwald et al. (1972) formula was employed in the estimation of Low and very low-density lipoprotein (LDL and VLDL).

2.13 Effect of the Stem Extract and Fractions of *T. occidentalis* on Liver Histology of *P. berghei* Infected Mice

The formalin-fixed harvested liver parts from mice were processed and stained with haematotoxylin and eosin (H&E) according to standard protocols (Drury and Wallington, 1980) at Department of Chemical Pathology, University of Uyo Teaching Hospital, Uyo. Alterations in morphology were assessed and recorded in each liver sample from the sacrificed mice. Photomicrographs were taken using microscope.

2.14 Statistical Analysis

One way analysis of variance (ANOVA) followed by Tukey's multiple comparison post-test were employed in the analysis using Graph pad prism software Inc. La Jolla, CA, USA. Data are expressed as mean ± SEM and significant values were considered at $p<0.05$ relative to control.

3. RESULTS

3.1 Determination of Median Lethal Dose (LD_{50})

Administration of stem extract of *T. occidentalis* (100 - 5000 mg/kg) orally had no lethal effect to the animals groups administered. Moreover, no physical toxic signs of the extract was observed. The LD_{50} of stem extract of *T. occidentalis* was therefore estimated to be =5000 mg/kg.

3.2 Antiplasmodial Activities of *Telfairia occidentalis* Stem Extract and Fractions on Established Infection

Administration of extract/fraction to *P. berghei* infected mice resulted in a significant ($p<0.001$), progressive and dose-dependent parasitaemia reductions in all the extract/fraction-treated groups relative to control. Statistically significant ($p<0.001$) parasitaemia reductions relative to the control was recorded on day 7 with the highest effect of 69.74% and m.s.t value of 16.10±1.76 d recorded in the group treated with 600 mg/kg of the stem extract. The n-hexane fraction was the most potent fraction having chemosuppressive activity of 69.68 % and m.s.t value of 15.60±2.06 d) on day 7, which was lower relative to that of chloroquine, 92.77 %. Prominent protective activities of the stem extract and fractions were observed in the treated infected mice as recorded in the mean survival time of the animals, which was significant ($p<0.05$-0.001) only in groups treated with the highest dose (600 mg/kg) and n-hexane fraction relative to the control. The n-hexane fraction provided a much longer mean survival time, 15.60 ± 2.06 d followed by that of dichloromethane fraction-treated mice, 11.40 ± 0.50 d. These survival times were shorter relative to that of chloroquine (29.83 ± 0.16 d; Table 1).

3.3 Effect of Stem Extract and Fractions of *Telfairia occidentalis* on Haematological Indices *Plasmodium berghei*-Infected Mice

Treatment of *P. berghei*-infected mice with *T.occidentalis* stem extract and fractions did not caused any significant ($p>0.05$) effect on RBC, Hb concentration, PCV percentages, lymphocytes, monocytes, basophils and platelets counts relative to the control. White blood cell count was significantly ($p<0.05$) elevated in groups treated with middle and high doses (400 and 600 mg/kg), hexane and methanol fractions as well as chloroquine, while neutrophils percentages were significantly ($p<0.05$-0.001) elevated in groups treated with low and middle doses (200 and 400 mg/kg) and methanol fraction (Table 2).

3.4 Effect of Stem Extract and Fractions of *Telfairia occidentalis* on Liver Function Parameters of *Plasmodium berghei*-infected Mice

Treatment of *P. berghei* infected mice with *T.occidentalis* (200 - 600 mg/kg) stem extract and fractions had no significant ($p>0.05$) alteration of liver function indices studied (total protein, albumin, total and conjugated bilirubin,AST, ALT, ALP,) when compared to those of the control untreated *P. berghei* - infected mice (Table 3).

3.5 Effect of *Telfairia occidentalis* stem Extract and Fraction on Liver Oxidative Stress Markers of of Mice Infected with *Plasmodium berghei*

The oxidative stress indices (GSH, SOD, GPX, CAT and MDA) of mice infected with *Plasmodium berghei* did not change significantly ($p>0.05$) when administered with stem extract and fractions *of T.occidentalis* (200 - 600 mg/kg)

Table 1. Curative activity and mean survival time of mice treated with stem extract and fractions of *Telfairia occidentalis* during established *Plasmodium berghei* infection

Treatment	Dose (mg/kg)	Parasitaemia					MST
		Day3	Day 4	Day 5	Day 6	Day 7	
Control	-	12.33±0.92	14.56±0.48	22.66±1.36	24.04±0.91	33.78±1.16	7.20±0.13
Extract	200	12.24±0.35	13.14±0.19	19.10±0.16	21.22±0.32	23.06±0.15^{c}	10.60±1.20
	400	13.35±0.49	14.55±1.12	16.31±0.68	18.39±0.28	20.20±0.55^{c}	11.20±0.80^{c}
	600	13.26±0.81	12.20±0.17	13.88±0.18^{c}	12.11±0.33	10.22±1.54^{c}	16.10±1.76^{c}
n-hexane	400	15.38±1.22	13.29±1.04	12.76±1.68	10.33±0.44	10.24±0.24^{c}	15.60±2.06^{b}
Dichloromethane	400	13.21±0.51	13.22±0.56	15.33±0.96	18.0±0.55	22.24±1.29^{c}	11.40±0.50^{c}
Ethyl acetate	400	13.25±1.44	15.15±1.53	18.66±0.20	25.28±1.14	31.28±1.02	7.40±0.24
Methanol	400	14.71±0.62	15.90±1.16	18.13±0.63	21.34±0.26	24.54±1.38^{c}	8.60±1.60
Chloroquine	5	14.62±0.43	11.44±0.23	7.66±0.18^{c}	5.12±0.22^{c}	2.44±0.28^{c}	29.83±0.16^{c}

Values are expressed as mean ± SEM. Significant relative to control. $^{c}p<0.001$. n = 6

Table 2. Effect of *Telfairia occidentalis* stem extract/fractions on haematological parameters of *P. berghei*-infected mice

Treatment	WBC ($x10^{9}$/L)	LYM (%)	NEUT (%)	MONO (%)	ESINO (%)	BASO (%)	RBC ($x10^{12}$/L)	HGB (g/dL)	PCV (%)	PLT ($x10^{9}$/L)
Control	7.72±1.36	39.36±8.61	32.26±1.22	2.46±0.83	1.26±0.76	1.30±1.06	6.83±0.37	11.30±0.83	36.40±2.93	133.0±11.71
Extract (200 mg/kg)	6.40±0.46	54.60±2.12	43.73±1.81^{a}	1.00±0.45	0.66±0.17	0.00±0.00	4.45±1.51	6.0±2.65	22.56±9.36	141.6±2.60
Extract(400mg/kg)	7.37±0.98	47.63±2.75	48.23±1.23^{b}	1.76±0.80	1.86±0.34	0.50±0.45	5.96±0.64	11.23±1.18	36.66±1.93	199.30±68.68
Extract (600 mg/kg)	12.34±4.12^{a}	59.46±8.43	39.23±1.53	1.43±0.62	1.33±0.39	0.03±0.03	4.62±1.12	7.06±1.75	24.86±4.80	129.0±42.72
Hexane fraction	18.55±1.14^{a}	59.90±3.61	37.83±3.34	5.70±2.32	1.16±0.96	0.03±0.03	4.23±1.00	8.13±3.13	25.83±8.53	96.66±23.69
Dichloromethane fraction	6.49±1.15	60.86±2.98	37.43±3.16	0.76±0.21	0.83±0.28	0.10±0.05	4.77±0.79	8.76±2.02	30.03±5.88	131.0±58.85
Ethyl acetate fraction	8.29±1.23	67.60±5.33	22.86±4.35	3.26±0.96	3.56±2.69	0.26±0.26	5.71±1.08	11.56±0.89	40.16±3.32	137.3±13.34
Methanol Fraction	12.19±2.00^{a}	12.63±2.86	82.16±0.73^{c}	1.73±0.21	1.73±1.03	0.20±0.15	4.62±0.05	11.96±0.66	41.46±0.84	196.0±26.40
Chloroquine	18.46±4.30^{a}	64.93±3.51	32.26±9.22	2.46±0.83	1.26±0.76	0.00±0.00	4.34±0.63	5.70±3.48	25.66±6.00	121.0±13.20

All values are presented as mean±S.E.M. for six rats in each group.compared with control group $^{a}p<0.05$, $^{b}p<0.01$, $^{c}p<0.001$

Table 3. Effect of stem extract and fractions of *Telfairia occidentalis* on liver function parameters of mice infected with *Plasmodium berghei*

Treatment	Dose (mg/kg)	Liver Function Parameters						
		AST (IU/L)	ALT(IU/L)	ALP(IU/L)	Total Protein (g/L)	Albumin (g/L)	Total Bilirubin (µmol/mL)	Conjugated bilirubin (µmol/mL)
Control	-	46.0±0.57	44.66 ±2.60	56.0±1.73	78.66±2.02	47.33 ±1.20	8.76±0.5	6.20±0.17
Extract	200	51.33±1.18	48.66±0.10	54.66±4.33	71.33±1.76	44.66 ±2.02	10.16±0.63	7.50±0.40
	400	48.0±0.57	45.0±1.15	63.66±0.88	72.66±1.45	43.0±2.08	9.63±0.08	6.43±0.17
	600	46.33±1.76	43.38±0.88	61.33±2.02	67.0±5.50	41.0±1.52	9.13±0.23	6.60±0.40
n-hexane	400	47.66±3.18	55.0±4.61	57.0±1.15	72.33±3.84	43.33 ±2.72	9.46±0.50	6.80±0.51
Dichloromethane	400	47.0±1.15	51.66±2.60	65.66±1.76	70.66±2.33	43.0 ±2.08	9.16±0.08	6.50±0.17
Ethyl acetate	400	45.0±1.15	46.0±0.57	57.33±2.02	60.0±2.30	35.33±1.45[a]	9.23±0.14	6.53±0.14
Methanol	400	42.0±0.57	42.0±1.73	60.00±1.15	75.66±1.85	46.33±1.20	8.30±0.11	5.63±0.17
Chloroquine	5	44.38±0.88	44.0±0.57	59.0±1.15	75.66±1.85	48.0±0.57	8.80±0.20	6.06±0.14

Values are expressed as mean ± SEM. Significant relative to control. [a]p<0.05; [b]p<0.01; [c]p<0.001. n = 6

Table 4. Effect of stem extract and fractions of *Telfairia occidentalis* on liver antioxidant enzymes of mice infected with *Plasmodium berghei*

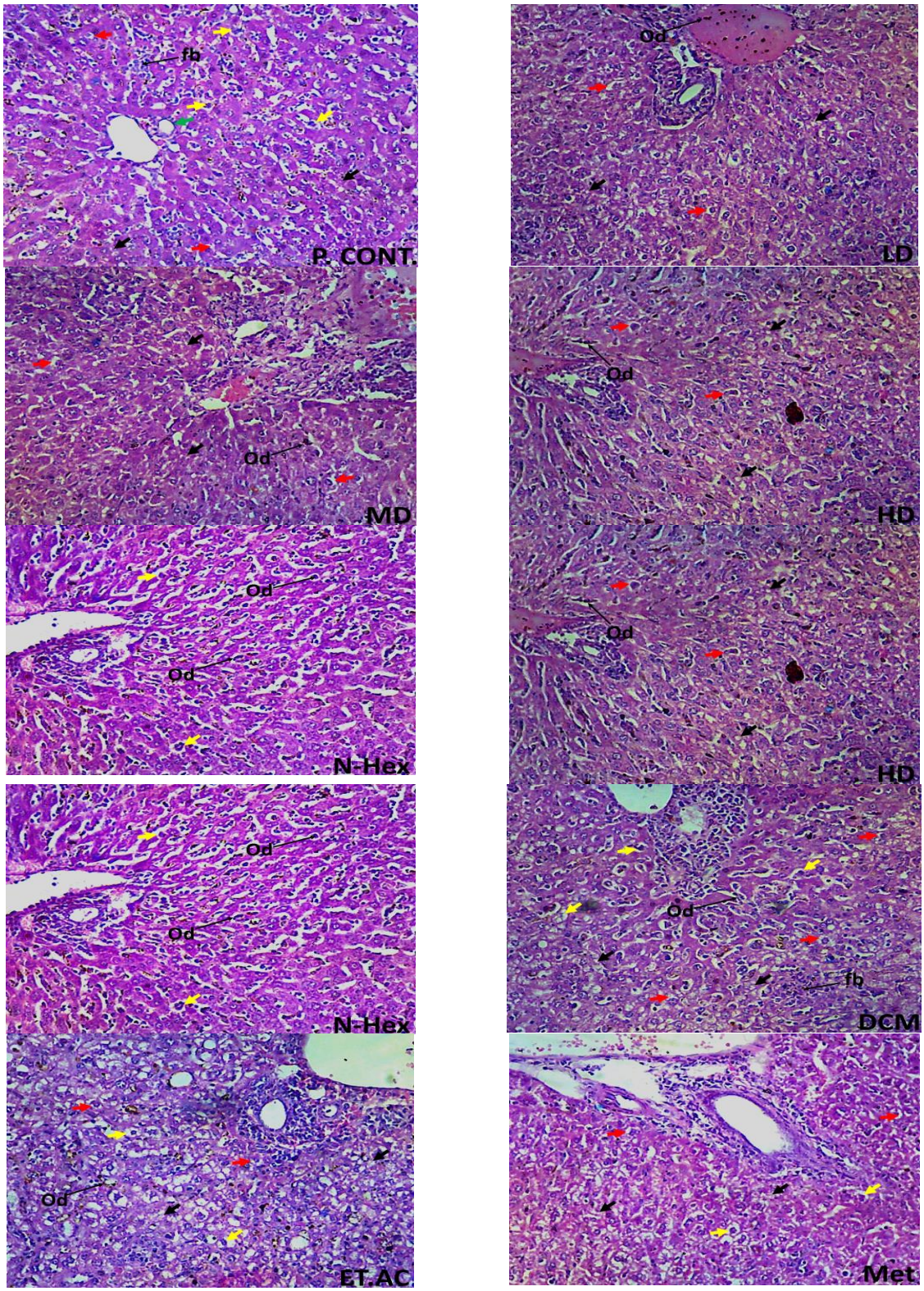
Treatment	Dose (mg/kg)	Antioxidant Parameters					
		GSH(µg/mL)	SOD(µg/mL)	CAT(µg/mL)	GPX (µm/mL)	MDA (µmol/mL)	Liver weight (g)
Control	-	1.89±0.29	0.43 ±0.02	3.65 ±0.27	0.056 ±0.008	0.35±0.02	2.91±0.12
Extract	200	2.02±0.34	0.36±0.07	5.61±0.66	0.059±0.009	0.42±0.07	2.46±0.11
	400	1.83±0.35	0.36±0.03	1.83±0.48	0.054±0.01	0.43±0.03	2.33±0.10
	600	2.20±0.54	0.36±0.05	3.89±0.64	0.065±0.01	0.44±0.05	2.28±0.13
n-hexane	400	1.55±0.07	0.33±0.07	3.58±0.19	0.056±0.01	0.46±0.07	2.41±0.14
Dichloromethane	400	2.35±0.57	0.37±0.01	4.44±0.98	0.068±0.01	0.44±0.01	2.20±0.15
Ethyl acetate	400	2.26±0.19	0.40± 0.01	2.55±0.23	0.067 ±0.005	0.39±0.01	2.40±0.16
Methanol	400	2.46±0.39	0.36± 0.03	3.50± 0.20	0.075 ±0.01	0.60±0.13	2.31±0.14
Chloroquine	5	1.70 ±0.07	0.32 ±0.05	2.88 ±0.81[c]	0.049±0.001	0.35±0.02	2.21±0.16

Values are expressed as mean ± SEM. Significant relative to control. [c]p<0.001. n = 6

Table 5. Effect of stem extract and fractions of *Telfairia occidentalis* on lipid profile of mice infected with *Plasmodium berghei*

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	4.46± 0.33	1.58± 0.20	1.68± 0.17	3.50± 0.26	0.71± 0.09
Crude extract	200	3.63±0.31	1.34± 0.10	1.39± 0.08	2.84± 0.32	0.61± 0.05
	400	4.40± 0.17	1.55± 0.09	1.88± 0.08	3.42± 0.12	0.70± 0.03
	600	4.13± 0.36	1.18± 0.05	1.21± 0.08	3.45± 0.36	0.53± 0.02
n-hexane	400	3.20± 0.47	1.32± 0.21	1.36± 0.16	2.43± 0.43	0.54± 0.09
Dichloromethane	400	4.43± 0.37	1.64± 0.10	1.75± 0.08	3.43± 0.34	0.74± 0.04
Ethyl acetate	400	3.83± 0.17	1.54± 0.11	1.58± 0.08	2.95± 0.22	0.70± 0.05
Methanol	400	4.13± 0.61	1.43± 0.05	1.49± 0.21	3.28± 0.54	0.65± 0.07
Chloroquine	5	4.36± 0.53	1.69± 0.11	1.70± 0.16	3.50± 0.26	0.71± 0.09

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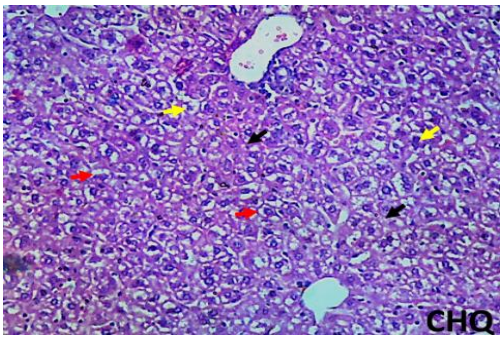

Fig. 1. Histologic Liver sections of *Plasmodium berghei*-infected mice untreated with normal saline (CONT), stem extract of *T. occidentalis*, 200 mg/kg (LD), 400 mg/kg (MD), 600 mg/kg (HD), n-hexane fraction(N-HEX), DCM fraction (DCM), ethyl acetate fraction (ETAC), methanol fraction MET) and chloroquine, 5 mg/kg (CHQ) at magnification X400. Keys: degenerated hepatic cells (yellow arrow) increased degenerating and vacoulated hepatocytes (red arrow), widespread micro-vesicular steatosis (black arrow), fibrosis of connective tissues (fb), shrinked ductal cells (green arrow) and organic deposits (Od) inter-hepatic fibrosis (fb)

relative to control. Although there were observable increases and/or decreases in the enzymes and molecules levels in the respective treatments, these changes were not significantly different ($p>0.05$) relative to control. (Table 4).

3.6 Effect of *Telfairia occidentalis* Stem Extract and Fraction on Lipid Profile of Mice Infected with *Plasmodium berghei*

Oral administration of *Plasmodium berghei*-infected mice with *T.occidentalis* stem extract and fractions (200-600 mg/kg) had no significant ($p>0.05$) effect on lipid profile parameters (total cholestrol, triglycerides, HDL, LDL, and VLDL) when compared to control. Although there were slight changes in the levels of these indices in the respective treatments, these changes were statistically insignificant ($p>0.05$) relative to that of the control (Table 5).

3.7 Effect of Stem Extract and fractions of *Telfairia occidentalis* on the Histology of Liver of Mice Infected with *Plasmodium berghei*

Histological examination of H&E stained livers' sections of mice administered various doses of *T. occidentalis* stem extract and fractions (200-600 mg/kg) at magnification (x100) revealed that untreated infected mice in group 1(control) given distilled water (10 mL/kg) had liver tissue demonstrating a severely altered hepato-architecture with areas of degenerated hepatic cells, inter-hepatic fibrosis, increased degenerating and vacoulated hepatocytes, widespread micro-vesicular steatosis, and shrinked ductal cells within the hepatic portal area. Extract low dose (200 mg/kg) treated infected mice in group 2 had liver tissue showing moderately altered hepato-architecture with increased degenerating and vacoulated hepatocytes, widespread micro-vesicular steatosis and organic deposits within the blood vessel and hepatic lobules. Group 3 infected mice treated with the extract dose (400 mg/kg) had liver tissue demonstrating a moderately altered hepato-architecture with increased degenerating and vacoulated hepatocytes and widespread micro-vesicular steatosis and organic deposits within the hepatic lobules. Mice in group 4 infected with *P. berghei* and treated with high dose extract (600 mg/kg) had liver tissue demonstrating a moderately altered hepato-architecture with increased degenerating and vacoulated hepatocytes, widespread micro-vesicular steatosis and organic deposits within the hepatic lobules. Liver tissues of infected mice in group 5 treated with n-hexane fraction showed moderately altered hepato-architecture with areas of degenerated hepatic cells and organic deposits within the hepatic lobules. Dichloromethane fraction treated infected mice in group 6 had liver tissue showing a severely altered hepato-architecture with areas of degenerated hepatic cells increased degenerating, vacoulated hepatocytes, widespread micro-vesicular steatosis, fibrosis of connective tissues and organic deposits within the hepatic lobules. Ethyl acetate fraction treated infected mice in group 7 had liver tissue demonstrating a severely altered hepato-architecture with areas of degenerated hepatic

cells increased degenerating and vacoulated hepatocytes, widespread micro-vesicular steatosis, and organic deposits within the hepatic lobules. Methanol fraction treated mice had liver tissue demonstrating a severely altered hepato-architecture with areas of degenerated hepatic cells increased degenerating and vacoulated hepatocytes, widespread micro-vesicular steatosis, and organic deposits within the hepatic lobules. Chloroquine treated mice had liver tissue demonstrating a severely altered hepato-architecture with areas of degenerated hepatic cells increased degenerating and vacoulated hepatocytes, widespread micro-vesicular steatosis and organic deposits within the hepatic lobules. (Fig. 1).

4. DISCUSSION

The stem of *T. occidentalis* are used locally as vegetable and to treat diverse diseases including malaria, diabetes, fever, arthritis and pain among others. This work was designed to investigate its effect on parasitaemia, hematological parameters, oxidative stress, lipid profile, liver functions and liver histology in mice infected with *Plasmodium berghei* employing curative test method to confirm its antimalarial activity and lay credence to its use locally as malaria remedy. *T. occidentalis* stem extract and fractions were evaluated for schizonticidal potentials against *P. berghei* infection in mice using standard *in vivo* method. The results revealed that the stem extract and fractions significantly decimated the parasitaemia in the established *P. berghei* infection model evaluated, in a dose-dependent manner with n-hexane fraction followed by dichloromethane fraction demonstrating significant *in vivo* activity corroborating the antimalarial activity of the stem extract. Prolongation of the MST of the mice by the extract and fractions was also observed, though with minimal degree of protection to the treated mice as most of the treated animals eventually died from infection. This activity suggests possible involvement of plasmodicidal or plasmodistatic actions of the phytoconstituents of the extract and fractions. Thus, corroborating previously reported antiplasmodial activities of the leaf, seed and root extracts of *T. occidentalis* (Okokon et al., 2007; Okokon et al*.,* 2009; Ebong et al*.*, 2020) in which these parts were shown to have exerted significant antimalarial activities. These findings confirm the local use of the stem extract decoctions to treat malaria.

The curative effects of the extract/fractions on the progression of *Plasmodium berghei* parasitaemia as observed in the present study, suggest that the extract and fractions have effect on the erythrocytic stages of the parasite (Waako et al*.,* 2005). The inability of the extract/fractions to provide complete protection to the infected animals in most cases could have resulted from low doses (200 -600 mg/kg) used, short half life/ duration of action of the extract/fractions due to rapid biotransformation processes and subsequent elimination (Waako et al*.,* 2005). Thus, resulting in further development and multiplication of the parasites as well as short MST of the extract/fractions- treated mice as observed.

Enin et al*.* (2024) had reported the presence of some pharmacologically active compounds in the extract and fractions understudy including tannins, flavonoids, alkaloids, terpenes and polyunsaturated fatty acids (PUFAs) among others. This revelation suggests the implication of these compounds in the observed pharmacological properties recorded in this study. A number of secondary metabolites (Kirby et al*.*, 1989; Philipson and Wright 1991; Christensen and Kharazmi 2001) and polyunsaturated fatty acids in the active hexane fraction as reported by Enin et al*.* (2024) to be present in the extract/fractions, have been implicated in antiplasmodial activities of plants. Antiplasmodial activities of these PUFAs are enhanced according to their unsaturation degree (Suksamrarn et al*.,* 2005; Attioua et al*.*, 2007; Melariri et al*.,* 2011, 2012; Enyiekere et al*.,* 2024a). Flavonoids demonstrate pronounced antiplasmodial activities against both chloroquine sensitive and resistant strains of human malaria parasite, *P. falciparum* (Attioua et al*.,* 2011; Ganesh et al*.,* 2012; Ezenyi et al*.*, 2014). Also, the free radical scavenging activities of these PUFAs mentioned above (Kohno et al., 1995; Ponnamma and Manjunath, 2012; Khan and Siddique, 2019) could contribute to the observed antiplasmodial activities. Moreso, antioxidant properties of flavonoids have been correlated with their antiplasmodial potentials (Cimanga et al*.,* 2009; Ganesh et al*.,* 2012), as elevated free radical levels are observed to be the cardinal factor in the pathogenesis of severe malaria complications. Significant free radical scavenging activities of the stem extract and fractions reported by Enin et al*.*, (2024) may account for the observed antiplasmodial activity in the current study and can be attributable to the presence of flavonoids and PUFAs. Similar study on *Justicia insularis*, a vegetable, had reported that poly unsaturated fatty acids bind to

Plasmodium falciparum Serine Hydroxymethyl Transferase (PFSHMT) and *Plasmodium falciparum* Erythrocyte Membrane Protein 1 (PFEMP-1) Proteins to cause inhibition of DNA synthesis, and apoptosis, whereas monoterpenes from the plant inhibited PfEMP-1, reversed the attachment of parasitized red blood cells to micro-vascular endothelium as their mechanism of action for parasitemia clearance (Enyiekere et al., 2024a). The stem extract/fractions may likely act through any of these pathways. These observations suggest that stem extract and fractions of *T. occidentalis* have the potentials to mitigate malarial infection and further support the claim of its antiplasmodial potentials.

Large quantities of free radicals are produced during plasmodial infections which primarily result in oxidative stress leading to pathogenesis and development of complications (Guha et al., 2006; Fabbri et al., 2013; Ojezele et al., 2017; Sarr et al., 2017), probably due to inflammatory responses by the body immune system (Becker et al., 2004; Percario et al., 2012). This results in reduced body antioxidants levels (enzymatic and non enzymatic) (Asagba et al., 2010) resulting in elevated lipid peroxidation and malondialdehyde levels as obvious in severe malaria cases (Asagba et al., 2010; Adil et al., 2013). These indices have been used as indicators of malaria severity. The activities of the enzymes and MDA levels in the extract/fractions treated groups, in the present study,were found to be insignificantly different from that of the untreated control mice. This demonstrates that the plant extract and fractions were unable to reduce parasitaemia significantly to curb the prevalent high amount of free radicals generated by the activities of the body immune system against the parasites perhaps due to the low doses used in this study.

However, moderate pathological signs were observed in liver histologies of groups treated with extract (400 and 600 mg/kg) and hexane fraction, whereas other treatment groups had severe pathologies compared to that of the control. This shows that the stem extract and fractions at the doses (200 - 600 mg/kg) used in this study could minimally protect the liver and improve the endogenous antioxidant enzymes and molecules to curb the generated oxidative stress. This could have been due to low doses administered in this study. However, studies on effect of leaf extracts of *J. insularis* and *S. officinarum* on oxidative stress status and liver function indices of *P. berghei* infected mice had shown improvement of these parameters and liver protective potentials (Edem et al., 2022; Enyiekere et al., 2024b).

Injuries on the liver by the parasites and oxidative stress condition lead to leakages of cellular enzymes such as transaminases and compromise the secretory functions of the liver causing hyperbilirubinemia (Onyesom and Onyemakonor, 2011). This could have been the case in this study which the extract/fractions were unable to ameliorate effectively.

Besides, plasmodial infection alters lipoproteins levels thereby aggravating oxidative stress and progression of malaria complications (Nathawut et al., 2004; Krishna et al., 2009). The alteration in lipid metabolism is due to the body immune response (Memon et al., 2000). However, administration of *T.occidentalis* extract and fractions did not cause any significant ($p>0.05$) effect on serum concentration of total cholesterol (TC), triglyceride (TG), LDL,VLDL and HDL in the parasitized treated mice. The results suggest that the plant may not possess significant hypolipidemic potential at the doses administered in this study

Haematological alterations resulting in the decrease of blood parameters are often observed in infected mice as common signs of anaemia (Surve et al., 2017). During malaria infection, *Plasmodium* invades the host cells and destroys RBC through its activities (Buffet et al., 2010; Saganuwan et al., 2011). Administration of stem extract and fractions to *P. berghei*-infected mice did not alter significantly the blood parameters of the treated mice relative to untreated control, except significant elevation of WBC count of the treated mice. This implies that the stem extract has no erythropoietic effect perhaps due to the low doses administered. The significant increase in WBC observed in infected untreated mice was earlier reported in malaria infection (Guyton, 2007) and attributed to the body's immunogenic response to the parasite and its metabolic products (Malaguarnera et al., 2002). This suggests the immunostimulatory activities of some constituents of the stem extract and fractions (Bero and Quertin-Leclercg, 2009), thereby offering certain degree of protection to the infected mice.

5. CONCLUSION

The findings of this investigation demonstrate that the stem extract and fractions of *T.*

occidentalis possess considerable antimalarial potential with minimal antioxidative stress and liver protective potentials which is attributable to the roles of its phytochemical constituents.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

Adil, R., Sumit, K., Varshney, M., Shahid, M. K., Haris, M., Ashraf Malik, A., Abbas, F. S., & Mahdi, A. (2013). Lipid peroxidation in cerebral malaria and role of antioxidants. *IOSR-PHR, 3*(1), 15-18.

Asagba, S. O., Eriyamremu, G. E., George, B. O., & Okoro, I. (2010). Biochemical indices of severity in human malaria. *Journal of Medical Sciences, 10*(4), 87-92.

Atanu, F. O., Idih, F. M., Nwonuma, C. O., Hetta, H. F., Alamery, S., & Batiha, G. E. (2021). Evaluation of antimalarial potential of extracts from *Alstonia boonei* and *Carica papaya* in *Plasmodium berghei*-infected mice. *Evidence-Based Complementary and Alternative Medicine, 2021*, Article ID 2599191, 11 pages.

Attioua, B., Weniger, B., & Chabert, P. (2007). Antiplasmodial activity of constituents isolated from *Croton lobatus*. *Pharmacology & Biology, 45*, 263–266.

Becker, K., Tilley, L., Vennerstrom, J. L., Roberts, D., Rogerson, S., & Ginsburg, H. (2004). Oxidative stress in malaria parasite-infected erythrocytes: Host-parasite interactions. *International Journal for Parasitology, 34*, 163–189.

Bero, J., & Quetin-Leclercq, J. (2011). Natural products published in 2009 from plants traditionally used to treat malaria. *Planta Medica, 77*(06), 631–640.

Buffet, P. A., Safeukui, I., Deplaine, G., Brousse, V., Prendki, V., Thellier, M. (2010). The pathogenesis of *Plasmodium falciparum* malaria in humans: Insights from splenic physiology. *Blood, 4*, 202911.

.

Centers for Disease Control and Prevention. (2023). Parasites-Malaria. Centers for Disease Control and Prevention. Available from: https://www.cdc.gov/parasites/malaria/index.html.

Christensen, S. B., & Kharazmi, A. (2001). Antimalarial natural products. In C. Tringali (Ed.), *Bioactive compounds from natural sources: Isolation, characterization and biological properties* (pp. 379–432). Taylor & Francis.

Cimanga, R. K., Tona, G. L., Kambu, O. K., Mesia, G. K., Muyembe, J. J., & Apers, S. (2009). Antimalarial, antiamoebic, and cytotoxic activities of some extracts and isolated constituents from the leaves of *Morinda morindoides* (Baker) Milne-Redh. (*Rubiaceae*). *Rec Progr Medicinal Plants, 25*, 225–242.

Drury, R. A., & Wallington, E. A. (1980). *Carleton's Histological Techniques* (5th ed.). Oxford University Press.

Ebong, A. S., Eseyin, O. A., Etim, E. I., & Okokon, J. E. (2020). *Telfairia occidentalis* potentiates antiplasmodial activity of artemisinin and amodiaquine combination therapy. *Anti-Infective Agents, 18*(2), 152–159. https://doi.org/10.2174/2211352517666190206160812

Edem, U. A., Okokon, J. E., Bassey, A. L., & Okokon, P. J. (2022). Antioxidative stress and hepatoprotective activities of leaf extract and fractions of *Saccharum officinarum* in *Plasmodium berghei*-infected mice. *Journal of Current Biomedical Research, 2*(4), 317–337.

Ellman, G. L. (1959). Tissue sulfhydryl groups. *Archives of Biochemistry and Biophysics, 82*, 70–77.

Enin, G. N., Okokon, J. E., Odokwo, B. O., & Antia, B. S. (2023). Preliminary phytochemical screening and in vivo inhibitory study of *Telfairia occidentalis* Hook f. seeds extract on alpha-amylase and alpha-glucosidase of rats. *Journal of Science and Technology Research, 5*(4),

26–35. http://dx.doi.org/10.5281/zenodo.10425980

Enyiekere, V. J., Asanga, E. E., Okokon, J. E., Ekeleme, C. M., Anagboso, M. O., & Ise, U. P. (2024a). Fatty acid esters and acyclic monoterpenoid from *Justicia insularis* leaf fractions attenuated malaria pathogenesis through docking with PFSHMT and PFEMP-1 proteins. *Natural Products Communication, 19*(7), 1–14.

Enyiekere, V. J., Essien, G. E., Anagboso, M. O., Ise, U. P., Ebong, N. O., & Okokon, J. E. (2024b). Effect of leaf extract and fractions of *Justicia insularis* on parasitemia, liver function indices, liver oxidative stress markers, and liver histology of *Plasmodium berghei*-infected mice. *Biology, Medicine, and Natural Product Chemistry, 13*(2), 351–359.

Esterbauer, H., & Cheeseman, K. H. (1990). Determination of aldehydic lipid peroxidation products: malonaldehyde and 4-hydroxynonenal. *Methods in Enzymology, 186*, 407–421.

Ezenyi, I. C., Salawu, O. A., Kulkarn, I. 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.

Fabbri, C., de Cássia Mascarenhas-Netto, R., Lalwani, P., Melo, G. C., Magalhães, B. M., Alexandre, M. A., Lacerda, M. V., & Lima, E. S. (2013). Lipid peroxidation and antioxidant enzymes activity in *Plasmodium vivax* malaria patients evolving with cholestatic jaundice. *Malaria Journal, 12*, 315.

Ferreira, J. F., Luthria, D. L., Sasaki, T., & Heyerick, A. (2010). Flavonoids from *Artemisia annua* L. as antioxidants and their potential synergism with artemisinin against malaria and cancer. *Molecules, 15*(5), 3135–3170.

Friedewald, W. T., Levy, R. I., & Fredrickson, D. S. (1972). Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clinical Chemistry, 18*, 499–502.

Ganesh, D., Fuehrer, H., Starzengruber, P., Swoboda, P., Khan, W. A., Reismann, J. A., Mueller, M. S., Chiba, P., & Noedl, H. (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.

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*(8), 439–449.

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

Khan, S. L., & Siddiqui, F. A. (2020). Beta-Sitosterol: As immunostimulant, antioxidant and inhibitor of SARS-CoV-2 spike glycoprotein. *Archive of Pharmacology and Therapeutic Journal, 2*(1), 12–16.

Kirby, G. C., O'Neill, M. J., Phillipson, J. D., & Warhurst, D. C. (1989). In vitro studies on the mode of action of quassinoids against chloroquine-resistant *Plasmodium falciparum*. *Biochemistry and Pharmacology, 38*, 4367–4374.

Kohno, Y., Egawa, Y., Itoh, S., Nagaoka, S., Takahashi, M., & Mukai, K. (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.

Lawrence, R. A., & Burk, R. F. (1976). Glutathione peroxidase activity in selenium-deficient rat liver. *Biochemical and Biophysical Research Communications*, 71, 952-958.

Lorke, D. (1983). A new approach to practical acute toxicity testing. *Archives of Toxicology*, 54, 275-286.

Malaguarnera, L., & Musumeci, S. (2002). The immune response to *Plasmodium falciparum* malaria. *Lancet Infectious Diseases*, 2(8), 472-478.

Marklund, S., & Marklund, G. (1974). Involvement of superoxide anion radical in the autooxidation of pyrogallol and a convenient assay for superoxide dismutase. *European Journal of Biochemistry*, 47, 469–474.

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. *Advances in Studies in Biology*, 4, 333–349.

Memon, R. A., Staprans, I., Noor, M., Holleran, W. M., Uchida, Y., Moser, A. H., Feingold, K. R., & Grunfeld, C. (2000). Infection and inflammation induce LDL oxidation in vivo. *Arteriosclerosis, Thrombosis, and Vascular Biology*, 20, 1536-1542.

Nathawut, S., Paveena, Y., Srivicha, K., Wattana, L., Gary, B., Sornchai, L., & Rachane, U. (2004). Increased fluidity and oxidation of malarial lipoproteins: Relation with severity and induction of endothelial expression of adhesion molecules. *Lipids in Health and Disease*, 3, 15.

Ojezele, M. O., Moke, E. G., & 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 Sciences*, 6(3), 260-265.

Okokon, J. E., Ekpo, A. J., & Eseyin, O. A. (2007). Antiplasmodial activity of ethanolic root extract of *Telfairia occidentalis*. *Research Journal of Parasitology*, 2(2), 94-98.

Okokon, J. E., Ekpo, A. J., & Eseyin, O. A. (2009). Evaluation of in vivo antimalarial activities of ethanolic leaf and seed extracts of *Telfairia occidentalis*. *Journal of Medicinal Food*, 12(3), 649–653. https://doi.org/10.1089/jmf.2008.0099

Okokon, J. E., Mobley, R., Edem, U. A., Bassey, A. L., Fadayomi, I., Horrocks, P., Drijfhout, F., & Li, W. W. (2022). In vitro and in vivo antimalarial activities and chemical profiling of sugarcane leaves. *Scientific Reports*, 41598, Article No. 14391. https://doi.org/10.1038/s41598-022-14391-8

Okokon, J. E., Okokon, P. J., & Sahal, D. (2017). In vitro antiplasmodial activity of some medicinal plants from Nigeria. *International Journal of Herbal Medicine*, 5(5), 102-109.

Okokon, J. E., Utibe, A., Edem, U., Obang, J. A., & Bankhede, H. (2019). Antimalarial and antipyretic activities of cornsilk extract and fractions of *Zea mays*. *Discovery Phytomedicine*, 6(4), 143-150.

Okokon, J. E., Nkemnele, A. B., & Mohanakrishnan, D. (2017). Antimalarial, antiplasmodial, and analgesic activities of root extract of *Alchornea laxiflora*. *Pharmaceutical Biology*, 55(1), 1022-1031.

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

Percário, S., Moreira, D. R., Gomes, B. A. Q., Ferreira, M. E. S., Gonçalves, A. C. M., & Laurindo, P. S. O. C. (2012). Oxidative stress in malaria. *International Journal of Molecular Sciences*, 13(12), 16346–16372.

Philipson, J. D., & Wright, C. W. (1991). Antiprotozoal compounds from plant sources. *Planta Medica*, 57, 553–559.

Ponnamma, S. U., & Manjunath, K. (2012). GC-MS analysis of phytocomponents in the methanolic extract of *Justicia wyaadensis* (NEES) T. Anders. *International Journal of Pharma and Bio Sciences*, 3, 570-576.

Saganuwan, S., Onyeyili, P., Ameh, E., & Etuk, U. (2011). In vivo antiplasmodial activity by aqueous extract of *Abrus*. *Review of Latino American Medicine*, 39(1-2), 32-44.

Sarr, D., Cooper, C. A., Bracken, T. C., Martinez-Uribe, O., Nagy, T., & Moore, J. M. (2017). Oxidative stress: A potential therapeutic target in placental malaria. *ImmunoHorizons*, 1(4), 29-41.

Sinha, A. K. (1972). Colorimetric assay of catalase. *Analytical Biochemistry*, 47, 389–394.

Suksamrarn, A., Buaprom, M., Udtip, S., Nuntawong, N., Haritakun, R., & Kanokmedhakul, S. (2005). Antimycobacterial and antiplasmodial unsaturated carboxylic acid from the twigs of *Scleropyrum wallichianum*. *Chemical and Pharmaceutical Bulletin*, 53, 1327–1329.

Surve, K., Kulkarni, A., Rathod, S., & Bindu, R. (2017). Study of haematological parameters in malaria. *International Journal of Research in Medical Sciences*, 5(6), 2552-2557.

Tietz, N. W. (1990). *Clinical guide to laboratory tests* (2nd ed.). Sanders Company.

United Nations International Children's Emergency Fund (UNICEF). (2024). UNICEF data: Monitoring the situation of children and women. UNICEF. Available from: https://data.unicef.org/topic/child-health/malaria

Usunomena U and Okpiabhele A.(2023). *Telfairia occidentalis* Hook f. mitigates Carbon Tetrachloride induced Nephrotoxicity in Rat. *Journal of Research in Applied and Basic Medical Sciences* 9 (3) :130-137.

Waako, P. J., Gumede, B., Smith, P., & Folb, P. I. (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.

World Health Organization. (2020). World malaria report. https://www.who.int/publications/m/item/WHO-HTM-GMP-2020.08

World Health Organization. (2021). *World malaria report*. World Health Organization.

World Health Organization. (2022). *World malaria report*. World Health Organization.

World Health Organization. (2023). *World malaria report*. World Health Organization.

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