

Berghei Infected Wistar Rats; the Role of Herbal Cocktail on the Spleen.

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Abstract: Nigeria is largely affected by malaria and transmissions occur all year round. Medicinal plants are used to treat malaria in various regions of the country where access to standard antiplasmodial drugs are not readily available. This study evaluates the histoarchitecture and haematological features in *Plasmodium berghei* infected wistar rats treated with a herbal cocktail used in some Nigeria communities. Thirty-five Wistar rats weighing an average of 200 g were divided into seven groups consisting of five rats each. The cocktail and aqueous extracts of *Mangifera indica*, *Carica papaya*, and *Citrus limon* were orally administered to the infected Wistar rats at doses of 100 mg/kg/day for seven days while the aqueous leaf extract of *Azadirachta indica* was administered at a dose of 10 mg/kg/day. The therapeutic effects of the cocktail and the various extracts against *Plasmodium berghei* were investigated and the effects on the spleen were histologically assessed, complete blood count was also determined. Result revealed a hopeful significant reduction in the malaria associated splenic pathology of rats administered with the cocktail and extracts of *C. papaya*. The antiplasmodial features exhibited by these plants justify their usage in the local treatment of malaria. **Keywords;** *Carica papaya*, *Cocktail*, *Mangifera indica*, *Malaria*, *Plasmodium berghei*,

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I. Introduction

Malaria is the most important parasitic disease in the world [1]. It is caused by five species of parasite that affects humans. All the parasites belong to the genus *Plasmodium*: with *Plasmodium falciparum* being the deadliest and most prevalent in Africa [2]. Malaria infection has been reported to induce acute injuries to vital organs and the most pronounced changes inflicted by this disease involve the blood, spleen, liver and kidney of the infected host [3]. The disease primarily affects poor populations in tropical and subtropical areas, where the temperature and rainfall are suitable for the development of vectors and parasites [4]. Furthermore, parasite resistance to available antimalarial drugs and the absence of vaccines remains a major challenge in the control of malaria [5-6]. Due to inadequate and inaccessible health facilities, majority of people living in rural communities heavily depend on traditional medicine which involves the use of medicinal plants for the management of malaria [7]. About 219 million cases of malaria occurred globally with Africa being the most burdened region accounting for 92% (200 million) [8]. Five countries accounted for half the global malaria burden with Nigeria having 25%; Democratic republic of Congo 11%, Mozambique 5%, India 4% and Uganda 4%. Furthermore 80% of the global deaths in 2017 were concentrated in 17 countries with Nigeria accounting for the highest mortality [8]. Data from household surveys also indicated that anemia as a result of malaria is evident in endemic regions of Africa when compared with other regions impacted by this disease [8]. In the last decade, there has been resurgence in search for new lead compounds from plants to treat malaria [9]. Traditional herbal medicines have been used to treat malaria for thousands of years in various parts of the world [10]. Nigeria has rich flora diversity and many of the plant species are used by some indigenous people for medicinal purposes. In poor countries, the healthcare is often sustained by other practices based on cultural alternatives. In many developing countries, one-fifth of patients use indigenous herbal remedies to treat malaria [11]. Medicinal plants play a major role in many communities over the world in the treatment and prevention of disease and the

promotion of general health. Previous studies have shown that over 1200 medicinal plants from 160 families are used worldwide to treat malaria [11] and still many anti-malarial plant species remain to be discovered [2]. Traditional knowledge and the use of plant-based medicines remain important in the prevention and treatment of malaria especially in the rural areas of a developing countries like Nigeria. This is important because it is often quickly accessible and affordable to the rural dwellers [2]. Medicinal plants such as *A. indica*, *M. indica*, *C. papaya* have been reported to possess antiplasmodial properties [10, 2, 12] and are used in the local treatment of malaria in various African countries. Pharmacological studies have demonstrated in vitro antiplasmodial and/or in vivo antimalarial activities of extracts from 45 plant species used in Nigerian folk medicine with leaves of *A. indica*, *C. papaya*, *M. indica* confirmed to be inclusive [10]. Most of these plants are used in form of monotherapy, and only a few plants are taken together in combined therapies. An example is the multi-herbal extract referred to as herbal cocktail made-up of *A. indica*, *C. papaya*, *M. indica*, *C. limon* [12]. This study evaluates the splenic histology and haematological changes of *Plasmodium berghei* infected wistar rats treated with a herbal cocktail or its constituent extracts.

II. Materials and Methods

Experimental animals

The experiment was conducted at the University of Benin, Benin City, Nigeria. Six weeks old inbred Wistar rats, weighing an average of 200 g obtained from the animal house in the Department of Anatomy, University of Benin, Nigeria were used as experimental animals for this study. The rats were kept in ventilated cages in the animal house and were fed with growers' mash obtained from Edo Feeds and Flour Mill Limited, Ewu, Edo State, Nigeria throughout the duration of this study.

Parasite and infection

Chloroquine-sensitive *Plasmodium berghei*(NK 65 strain) was obtained from the Institute of Advance Medical Research and Training, University of Ibadan, Nigeria. Thirty-five adult wistar rats weighing an average of 200 g were divided into seven groups with each group consisting of five rats which were allowed to attain the requisite weights before commencement of the experiment.

The first group consisted of five non-infected rats (negative control) administered with distilled water only while the second group were constituted with rats infected with *Plasmodium berghei* but was administered with distilled water only (infected-untreated group). The infected rats in the third group were administered with the herbal cocktail while rats in the fourth, fifth, sixth and seventh group were administered with aqueous extracts of *A. indica*, *M. indica*, *C. papaya* and juice extract of *C. limon* respectively.

Inoculation procedure

Three donor mice with parasitemia $\geq 25\%$ were sacrificed and blood was collected into ethylene diamine tetra acetic bottle and diluted with phosphate buffer saline to 10^8 parasitized erythrocytes/mL. Healthy wistar rats in the test groups were inoculated intraperitoneally with preparation of the parasitised red cells [14].

Blood cytology evaluation

Giemsa stained blood films obtained from the tail of the infected rats were reviewed with oil immersion objectives [15]. Percentage parasitaemia was determined for each infected rat [16]. Rats with parasitaemia $\geq 25\%$, were treated with the cocktail or either of the constituent extract on the seventh post-infection day.

Cocktail and individual extract

Plant samples were identified by a plant Taxonomist from Plant Biology and Biotechnology Department of the University of Benin, Benin City, Nigeria as *A. indica* (Meliaceae), *M. indica* (Anacardiaceae), *C. papaya* (Caricaceae), and *C. limon* (Rutaceae) [17]. The cocktail, *C. limon* juice extract and aqueous leaf extracts of *M. indica* and *C. papaya* were administered to respective groups at a dose of 100 mg/kg/bw, while aqueous leaf extract of *A. indica* was administered at 10 mg/kg/bw. The chemotherapeutic effect of the extract against *P. berghei* was investigated after seven days of administration.

Histopathology studies

The spleen was excised and immediately transferred into 10% neutral buffered formalin and processed for light microscopic study, using an automatic tissue processor machine (Thermo Scientific Spin Tissue Processor, STP120, Frankfurt, Germany). Five microns thick paraffin sections were obtained, which were finally stained using the Hematoxylin and Eosin staining procedure and the sections mounted with DPX for microscopic examination [18].

Haematological studies

Blood samples obtained were analyzed using a hematological auto analyzer (Mindray BC-5300).

Statistical analysis

Data analysis was performed using statistical package for social sciences version 19.0. Data were expressed as mean \pm standard deviation. Test of significance was calculated using paired Student's t-test. $p < 0.05$ was considered to be significant.

III. Results

Histopathological findings

Histological sections of the spleen of the uninfected rats showed no pathology "Figure 1". The infected untreated rats showed the presence of severe malaria pigment with focal areas of necrosis within the spleen "Figure 2". Splenic histology of rats treated with the cocktail showed reduction in the malaria pigments on comparison with the infected untreated rats, however congestion was seen in an area with the trabeculae observed to be thickened 'Figure 3'. Sections of rats administered with the aqueous leaf extract of *A. indica* showed significantly reduced haemozoin on comparison with the infected untreated group and group treated with the cocktail; however, there was a mild degeneration in the red pulp while the white pulp "Figure 4". Rats administered with aqueous extract of *M. indica* was observed to have a moderately inflamed spleen with moderately thickened walled vessel and perivascular infiltration "Figure 5". Rat administered with aqueous extract of *C. papaya* showed no pathological lesions as the splenic histology revealed a normal trabecula with normal red and white pulp "Figure 6". Rats administered with aqueous juice extract of *C. limon* showed the presence of severe deposits of malaria pigments in various regions of the spleen. The red pulp housed the malaria pigment and the sinuses are disorganized. The trabeculae are mildly thickened and moderately thickened walled vessels are seen "Figure 7".

Haematological studies.

Table 1 shows the mean and standard deviation of the haematocrit, haemoglobin values, total and differential white cell counts among rats in various groups. Statistical variations were observed in the haematocrit values among the rats in various groups on comparison with the uninfected control group ($p < 0.00002$), however rats treated with the extracts of *A. indica*, *C. papaya* and *C. limon* were not statistically significant ($p > 0.05$) "Table 2.0a". Significant variation was observed in the group treated with either the cocktail or the extracts when compared with the infected untreated group ($p < 0.003$) "Table 2.0a". Statistically significant variations were observed in the haemoglobin (Hb) values in rats treated with the extract of *M. indica* and the cocktail ($p < 0.0001$) on comparison with the negative control, however, the rats treated with aqueous extracts of *A. indica* or *C. papaya* showed no significant variations in their haemoglobin ($p > 0.05$) "Table 2.0a". Comparative analysis of rats administered with the cocktail or either of the contained extracts with the infected untreated rats showed no statistically significant variation ($p > 0.05$) however, rats administered with extracts of *A. indica* and *C. limon* were observed to be significant ($p < 0.0003$) "Table 2.0a". Statistical variation was observed in rats administered with either extracts of *A. indica* or *C. limon* on comparison with the rats treated with the cocktail ($p < 0.0005$) "Table 2.0b". Significant variation was observed among rats in various groups when compared with the negative control ($p < 0.00001$) however, rats administered with either of the cocktail, aqueous extract of *A. indica* or extract of *C. limon* showed no statistically significant variation in their white cell count ($p > 0.005$) "Table 2.0a". Comparison of the various extracts and cocktail with the infected untreated rats showed statistically significant variations in the total white blood cell counts (TWBC) ($p < 0.00001$) with the exception of the rat administered with extracts of *M. indica* ($p > 0.05$) "Table 2.0a". Neutrophil counts were significantly raised in various groups on comparison with the negative control ($p < 0.0001$). Comparison of the cocktail and various extracts with the infected untreated group also revealed a statistically significant variation in the neutrophil count ($p < 0.0003$) however the rats administered with the extract of *M. indica* or *C. papaya* showed no significant variation in their neutrophil count ($p > 0.05$) "Table 2.0a". Lymphocyte values was significantly raised in various groups administered with the extracts or cocktail ($p < 0.00001$), however the group administered with extracts of *A. indica* showed no significant variation ($p > 0.05$) on comparison with the negative control "Table 2.0a". Lymphocyte value in rats administered with extracts of *M. indica* and *C. papaya* showed no significant variation on comparison with the infected untreated group ($p > 0.05$) however, rats administered with the cocktail and other extracts were observed to be statistically significant ($p < 0.0003$) "Table 2a". Eosinophil counts was significantly raised in rats of various groups on comparison with the uninfected group ($p < 0.00013$) however, rats administered with the extracts of *M. indica* showed no statistically significant variation in their eosinophil counts ($p > 0.05$) "Table 2.0a". Eosinophil count in rats administered with the cocktail or either of the extracts showed statistically significant variation on comparison with the infected untreated group ($p < 0.0002$) "Table 2.0a". Monocyte values were apparently

normal in all groups on comparison with the negative control, however a significantly raised value was observed in the infected untreated rats and rats administered with the cocktail ($p < 0.03$) "Table 2.0a". Comparison of monocyte values in rats administered with either of the extracts or cocktail with the infected untreated group was statistically significant with the exception of rats administered with aqueous extracts of *A. indica* or *M. indica* ($p < 0.0004$) "Table 2.0a". No significant variation was observed in the basophil counts in all the groups ($p > 0.05$) "Table 2.0a, b".

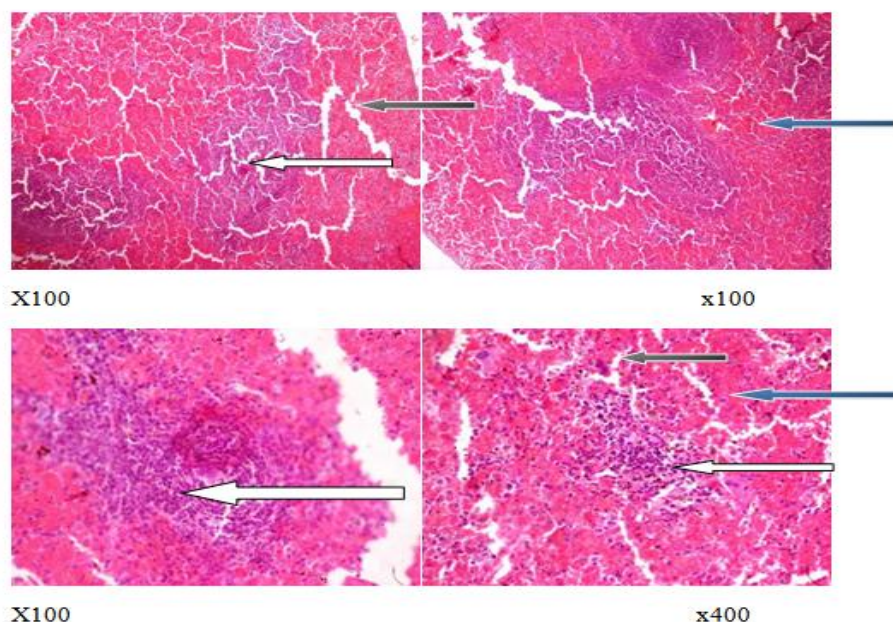


Figure 1. Photomicrographs of spleen of uninfected rats administered with distilled water only [negative control group] stained with Haematoxylin and Eosin showing normal trabeculae [black arrow], there are normal red pulp [blue arrow] and white pulp [white arrow] seen.

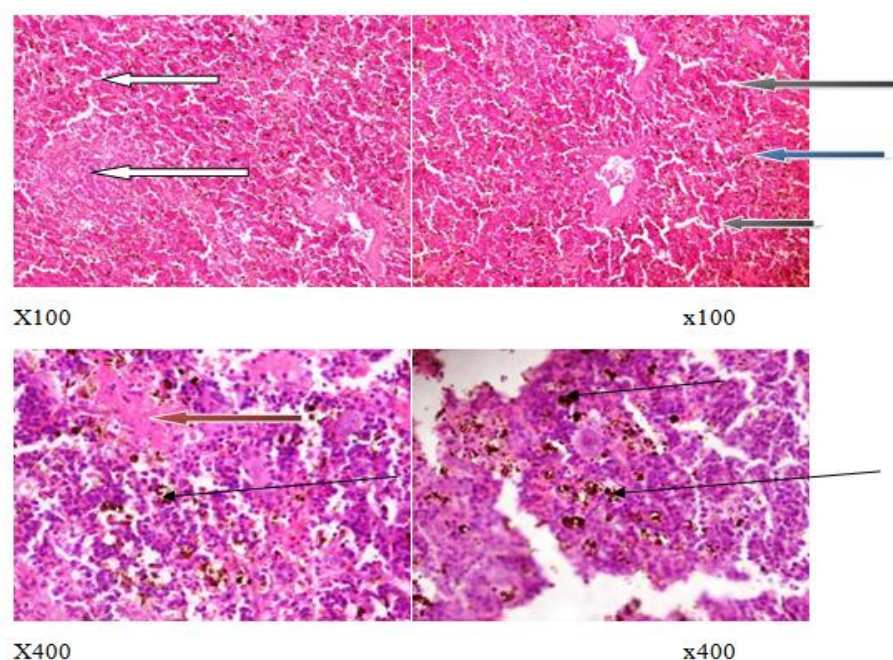


Figure 2. Photomicrographs of a splenic section of the infected untreated rats stained by Haematoxylin and Eosin showing splenic tissue with moderate to severe malaria pigments [slender arrow], there is a focal area of necrosis noted [red arrow]. The red [blue arrow] and white pulp [white arrow] seen appear normal. The trabeculae seen are normal [black arrow].

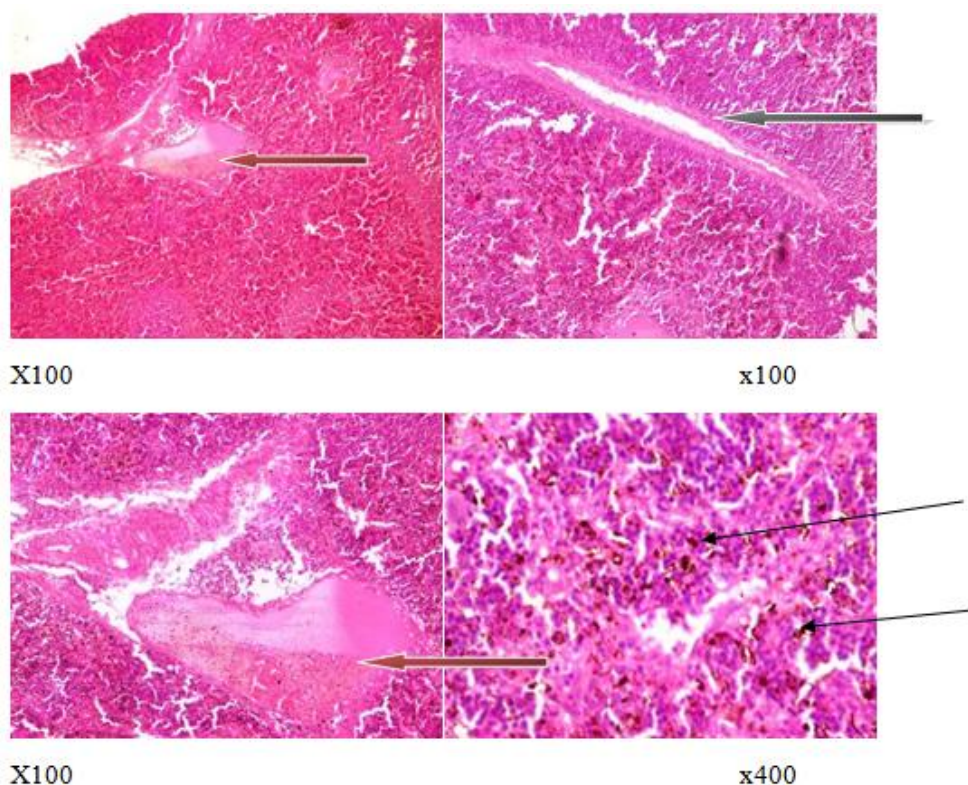


Figure 3 Photomicrographs of a splenic section of rats treated with the herbal cocktail, stained by Haematoxylin and Eosin showing splenic tissue with pigmentation suspected to be malaria pigments [slender arrow], there is an area with moderate congestion [red arrow]. The red pulp and white pulp seen appear normal. Mildly thickened trabeculae seen [black arrow], the sinuses are not well organized.

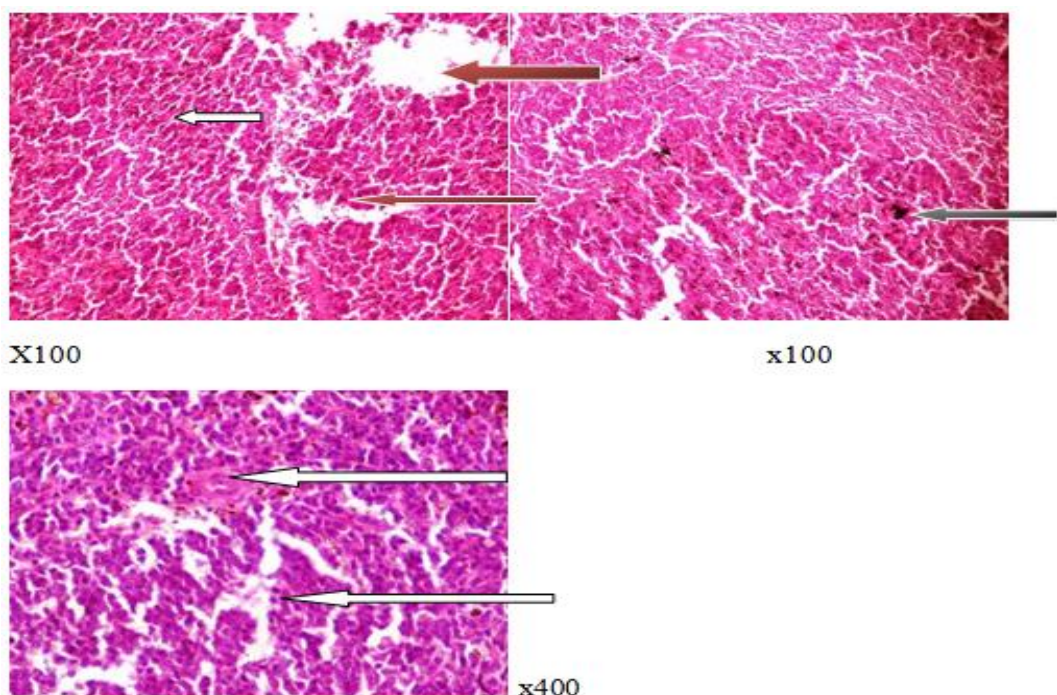


Figure 4. Photomicrographs of a splenic section of rats administered with aqueous extract of *A. indica*, stained by Haematoxylin and Eosin showing splenic tissue with moderately normal sinuses, there is mildly degenerated area of the red pulp seen [red arrow]. The white pulp [white arrow] seen appear normal. The trabeculae seen appear normal [black arrow].

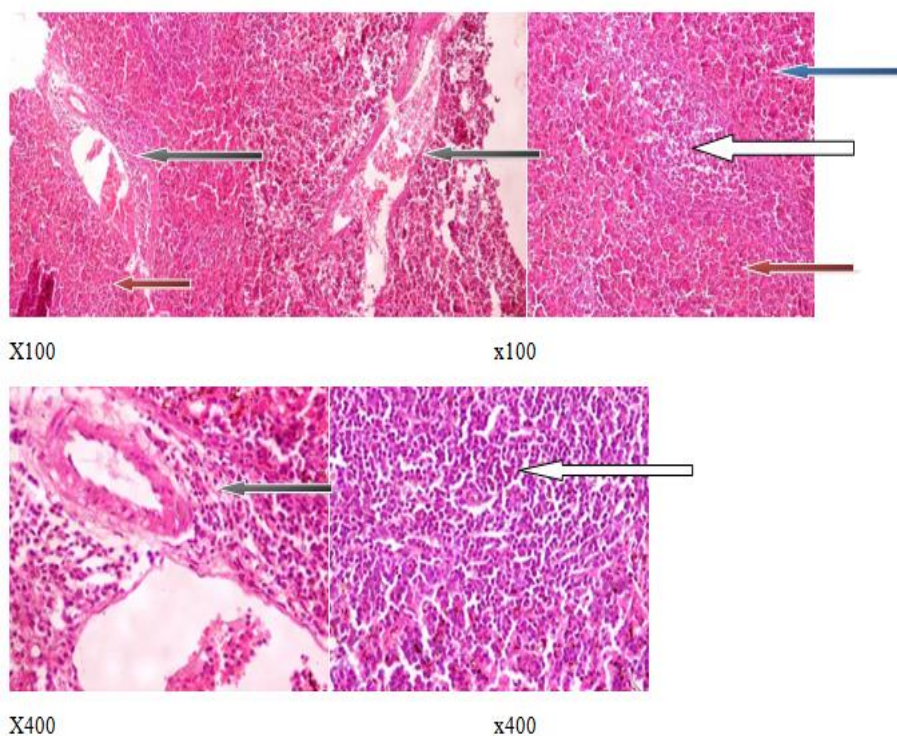


Figure 5. Photomicrographs of a splenic section of rats treated with *M. indica*, stained by Haematoxylin and Eosin showing moderately inflamed splenic tissue with moderately thick-walled vessel and perivascular infiltration [black arrow], the vessels are dilated, the red [blue arrow] and white pulp [white arrow] seen appear normal. The trabeculae are normal.

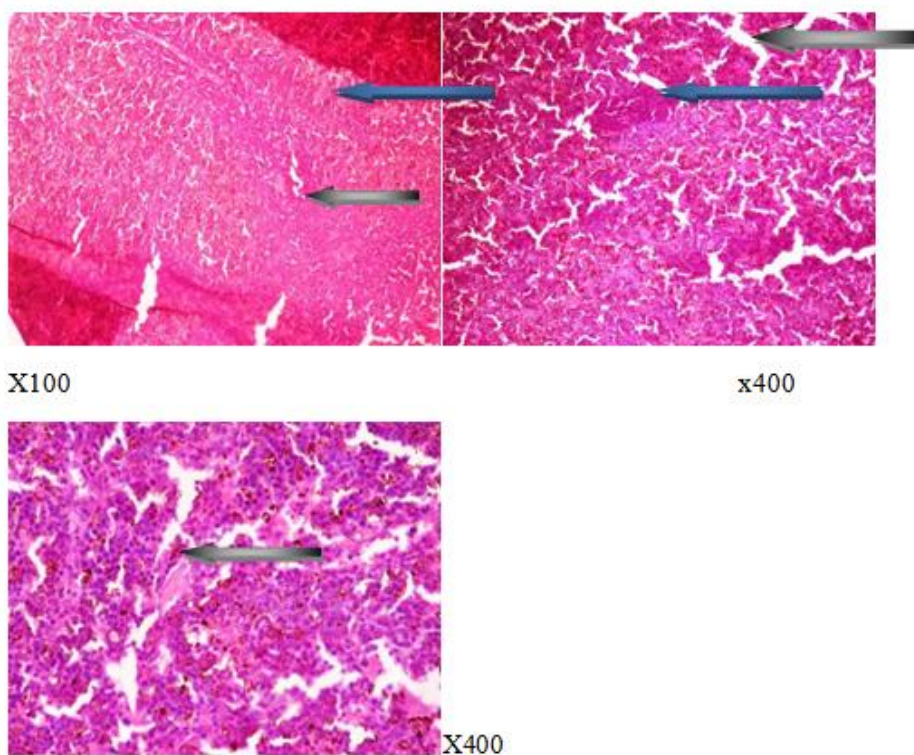


Figure 6. Photomicrographs of a splenic section of rats treated with extracts of *C. papaya*, stained by Haematoxylin and Eosin showing splenic tissue with normal trabeculae [black arrow], there are normal red pulp [blue arrow] and white pulp [white arrow] seen appear normal. There is mild malarial pigment seen.

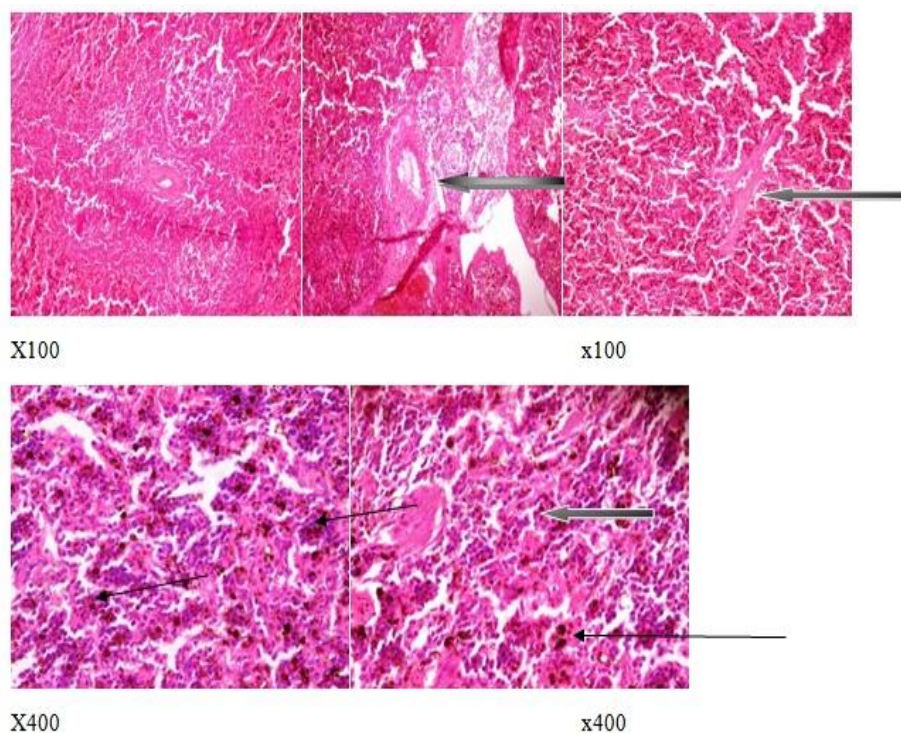


Figure 7. Photomicrographs of a splenic section of rats treated with *C. limon* juice extract, stained by Haematoxylin and Eosin showing splenic tissue with severely deposited malaria pigments [slender arrow]; the red pulp housed the malarial pigments, the sinuses are disorganized, and the trabeculae mildly thickened [black arrow]. There are moderately thick-walled vessels seen.

Table 1.0. Showing the haematological parameters among rats in the various groups.

Parameter	Negative control	Infected untreated group	Cocktail	<i>Azadirachta indica</i>	<i>Mangifera indica</i>	<i>Carica papaya</i>	<i>Citrus limon</i>
HCT	45±1.50	26±2.0	34±1.6	40±4.0	37±3.7	41±6.6	39±3.0
HB	12±0.5	7±0.4	8.2±0.4	11±0.7	9.2±1.1	10±1.2	11±0.5
TWBC	8080±428	17040±535	10040±1808	10820±1607	13000±2200	11680±1541	7560±827
Neutrophil	59±1.4	26±0.9	34±2.8	40±2.5	32±5.0	32±2.8	39±1.8
Lymphocyte	52±1.3	69±0.9	64±2.2	57±2.7	67±4.7	65±2.6	60±1.6
Eosinophil	4±0.2	4.4±0.6	0.3±0.7	1±0.2	1±0.3	2.2±0.8	1±0
Monocyte	1±0.2	0.2±0.2	0.2±0.2	1±0.2	1±0.2	1±0.4	0.4±0.2
Basophil	0±0	0.2±0.2	0±0	0±0	0.2±0.2	0±0	0±0

Note: HCT= haematocrit; Hb= haemoglobin; TWBC= Total white blood cell count.

Table 2.0a. Comparative analysis of haematological parameters between rats in different groups.

parameter	Negative control vs Infected untreated group	Negative control Vs Cocktail	Negative control Vs <i>Azadirachtaindica</i>	Negative control Vs <i>Mangiferaindica</i>	Negative control Vs <i>Carica papaya</i>	Negative control Vs <i>Citrus limon</i>	Infected untreated group Vs Cocktail	Infected untreated group Vs <i>Azadirachtaindica</i>	Infected untreated group Vs <i>Mangiferaindica</i>	Infected untreated group Vs <i>Carica papaya</i>
HCT	8.06	5.24	1.09	2.02	0.56	1.71	-3.14	-3.07	-2.68	-2.27
"p"	0.00002**	0.0004**	0.15	0.04*	0.3	0.06	0.006**	0.008**	0.01*	0.03*
Hb	6.22	5.84	1.43	2.21	1.64	0.57	-1.26	-3.77	-1.52	-1.63
"p"	0.0001**	0.0001**	0.09	0.03*	0.07	0.29	0.12	0.003**	0.08	0.07
TWBC	-13.07	-1.05	-1.64	-3.2	-2.25	0.56	3.71	3.67	1.78	3.28
"p"	0.00001**	0.16	0.07	0.03**	0.03*	0.3	0.003**	0.003**	0.07	0.006**
NEUT	12.81	4.39	2.35	2.88	4.85	3.55	-2.61	5.34	-1.27	-2.03
"p"	0.0001**	0.001**	0.02*	0.01*	0.0006**	0.004**	0.01*	0.0003*	0.12	0.38
LYM	-11.12	-4.74	-0.187	-2.79	-4.61	-3.89	2.41	4.22	0.84	1.44
"p"	0.00001**	0.0007**	0.05	0.01*	0.0009**	0.002	0.02*	0.001**	0.21	0.09
EOSIN	-6.17	-3.05	-2.53	-1.5	-2.15	-2.45	1.99	5.06	5.01	2.2
"p"	0.00013**	0.008*	0.02*	0.09	0.03*	0.02*	0.04*	0.0005**	0.0005**	0.03*
MONO	2.12	2.12	-1.41	-1.41	0.45	1.26	0	-3.53	-3.53	-0.61
"p"	0.03*	0.03*	0.1	0.1	0.33	0.12	0.5	0.004**	0.004**	0.25
BASO	-1	0	0	-1	0	1	1	0	1	1
"p"	0.17	0.5	0.5	0.17	0.5	0.17	0.17	0.5	0.17	0.17

Note: HCT= haematocrit; Hb= haemoglobin; TWBC= Total white blood cell count; NEUT=neutrophil;LYM=lymphocyte; EOSIN=eosinophil;MONO=monocyte;BASO=Basophil.

Table 2.0b Comparative analysis of haematological parameters between rats in different groups.

Parameter	Cocktail Vs <i>A. indica</i>	Cocktail Vs <i>M. indica</i>	Cocktail Vs <i>C. pa papaya</i>	Cocktail Vs <i>Citrus limon</i>	<i>A.indica</i> vs <i>Mangifera indica</i>	<i>A. indica</i> vs <i>Carica papaya</i>	<i>A. indica</i> vs <i>Citrus limon</i>	<i>M. indica</i> vs <i>Carica papaya</i>	<i>M. indica</i> vs <i>Citrus limon</i>	<i>Carica papaya</i> Vs <i>Citrus limon</i>
HCT	-1.43	-0.83	-1.15	-1.69	0.56	-0.15	0.15	-0.58	-0.5	0.28
"p"	0.09	0.21	0.15	0.06	0.3	0.44	0.11	0.28	0.32	0.4
HB	-3.1	-0.88	-1.07	-5.1	1.11	0.71	-0.94	-0.24	-1.86	-1.33
"p"	0.0008**	0.2	0.16	0.0005**	0.15	0.25	0.19	0.4	0.05	0.11
TWBC	-0.32	-1.03	-0.69	1.25	-0.8	-0.39	1.8	0.49	2.31	2.35
"p"	0.38	0.16	0.25	0.12	0.22	0.35	0.05	0.32	0.02*	0.02
Neutrophil	-1.8	0.21	0.41	-1.7	0.39	2.22	0.39	0.07	-1.29	-2.17
"p"	0.05	0.42	0.35	0.06	0.35	0.02*	0.35	0.47	0.12	0.03*
Lymphocyte	1.84	-0.31	-0.47	1.5	-1.48	-2.12	-0.77	0	1.13	1.83
"p"	0.05	0.38	0.33	0.09	0.09	0.03*	0.23	0.5	0.15	0.05
Eosinophil	1.98	2.13	0.38	2.36	0.53	0.53	-1.21	-1.39	0	1.5
"p"	0.04*	0.03*	0.36	0.02*	0.3	0.3	0.13	0.1	0.5	0.09
Monocyte	-3.53	-3.53	-0.61	-0.63	0	1.34	2.5	-0.61	-0.63	0.42
"p"	0.004**	0.004**	0.25	0.27	0.5	0.11	0.02*	0.25	0.27	0.34
Basophil	0	0	1	1	0	1	0	1	1	0
"p"	0.5	0.5	0.17	0.17	0.5	0.17	0.5	0.17	0.17	0.5

Note: HCT= haematocrit; Hb= haemoglobin; TWBC= Total white blood cell count.

IV. Discussion

Medicinal plants play a significant role in the treatment and prevention of disease and promotion of general health [2]. Traditional knowledge and the use of plant-based medicines remain widespread in rural regions of Nigeria as it plays an important role in the prevention and treatment of malaria. Findings in this study revealed the extracts of *C. papaya* as the most potent of the extracts while aqueous extracts of *M. indica* and juice extract of *C. limon* were observed to be the least potent. Photomicrograph of the spleen of uninfected rats showed no pathology. However, the splenic histology of the infected untreated group showed severe deposit of malaria pigment, a focal area of necrosis was also observed; however, the trabeculae, red and white pulp appeared normal. Antiplasmodial potential of *C. papaya* has been earlier reported [12] with findings in this study further buttressing this claim as the rats administered with the extracts of *C. papaya* showed no observable malaria associated splenic pathology. Furthermore, the use of *C. papaya* among indigenous Brazilians in the treatment of malaria [19] further authenticates the antiplasmodial potential of *C. papaya* reported in this study. *C. papaya* has been reportedly used in form of decoction in the effective treatment of malaria [10]. Reduced antiplasmodial potential of *M. indica* has been earlier reported [12] which is in tandem with the findings in this study thus corroborating the earlier claims that *M. indica* was less effective in ameliorating Plasmodial berghei infection in adult wistar rats [12]. The use of Citrus species as a medicinal plant used in treating malaria has been reported [19] however, findings in this study revealed a low antiplasmodial potential on comparison with extracts of *C. papaya* or *A. indica* however it was observed to be more effective than the extracts of *M. indica*. Rats administered with the juice extracts of *C. limon* also displayed significant amount of haemozoin in the splenic section however it was not as evident as that in the rats treated with extract of *M. indica* [12,19]. These pathological changes were less evident in the groups administered with aqueous leaf extract of *A. indica* or the cocktail. Antiplasmodial activity of *A. indica* has been attributed to a substantial oxidative stress during malaria infection which affects all stages of maturation of the gametocyte [20]. Photomicrographs of rats administered with the cocktail showed significant reduction in malaria associated splenic pathology on comparison with the infected untreated rats. Significant ameliorative effect was observed in rat administered with extracts of *A. indica* on comparison with those administered with the cocktail as there was significant reduction in malarial pigment deposit which further authenticates the claims on the antiplasmodial capabilities of *A. indica* in the traditional treatment and management of malaria [21,12]. Haematological findings revealed a significant reduction in the haematocrit values of the infected untreated rats on comparison with the negative control. This implies that infection by this rodent malaria parasite induces anemia in the rats. However, the rats administered with the cocktail or either of the contained extracts recovered from anemia with a significant increase in their haematocrit with groups administered with the cocktail and extract of *A. indica* having the highest haematocrit values in the treated group. Various mechanism of action postulated for isolation of antimalaria compound from plant origin include the inhibition of haemozoin polymerization in the parasite [22-23]; inhibition of *P. falciparum* lactate dehydrogenase, an integral enzyme for duplication and energy generation of energy within the parasite [24]; alkylation, [25] or mitotic spindle and microtubules interference thus inhibiting formation of the microgametes [26-27]; Inhibition of parasite derived cysteine protease which prevents sporozoite invasion of host cell [28]. The Hb values significantly reduced in the infected untreated group on comparison with the uninfected group. Rats administered with the extracts of *A. indica* showed an elevated Hb value on comparison with the infected untreated group. There was an observable increase in TWBC in the infected untreated group on comparison with the negative control. Rats administered with the extracts of *A. indica* and *C. papaya* displayed a significant reduction in the TWBC counts on comparison with the infected untreated group while statistically significant changes were not observed in rats treated with the cocktail or extracts of *M. indica* thus buttressing earlier report on the reduced antiplasmodial effect of *M. indica* in *Plasmodium berghei* infected rats [12, 29]. Statistically significant changes were observed in the neutrophil counts of rats treated with the cocktail and extracts of *A. indica* when compared with the infected untreated group. There was also observable reduction in the eosinophil count in the rats administered with the cocktail and various extracts on comparison with infected untreated group thus indicating the antiplasmodial effects of these plants. Statistically significant difference was also observed in the monocyte count in rats administered with extracts of *A. indica* and *M. indica* while the basophil counts among the infected untreated group and the various test groups were statistically insignificant on comparison with the uninfected group.

V. Conclusion.

Nigeria possesses a large fauna containing various medicinal plants with massive Phyto medical potentials. Findings in this study further buttress the claim of local medical practitioner on the antiplasmodial properties of these plants. Furthermore, the therapeutic potential of these plants could be harnessed to address the menace of malaria and other communicable disease in Nigeria and endemic region of the world.

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