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IN VIVO ANTIMALARIAL ASSESSMENT AND PHYTOCHEMICAL COMPOSITION OF *Carica papaya* LEAF EXTRACT IN *Plasmodium berghei*-INFECTED MICE

***[1]Alozieuwa, Uchenna B. and [2,3]Ogbadoyi, Emmanuel O.**

[1]Department of Biochemistry, Veritas University Abuja, Bwari, FCT-Abuja, Nigeria
[2]Department of Biochemistry, Federal University of Technology, Minna, Nigeria
[3]Centre for Genetic Engineering and Biotechnology, Federal University of Technology, Minna, Nigeria.

*Corresponding authors' email: alozieuwau@veritas.edu.ng

ABSTRACT

Malaria is one of the most devastating illnesses of all time and the most common parasitic disease in sub-Saharan Africa. Increasing resistance of *Plasmodium* species to currently used antimalarial drugs has necessitated the search for novel and more effective anti-malarial drugs from medicinal plants used in traditional medicine. This study aimed to quantify the phytochemical constituents and evaluate the antimalarial activity of the leaf extract of *Carica papaya* in established *Plasmodium berghei* infection in mice. Quantitative phytochemical analysis was carried out using conventional biochemical techniques. Mice infected with *Plasmodium berghei* were administered orally with the extract at doses of 100-500 mg/kg/day for five consecutive days. The extract significantly ($p < 0.05$) suppressed parasitemia in a dose-dependent manner, with the highest parasite inhibition (59.89%) observed at 500 mg/kg. Mean survival time (MST) ranged from 17.00 ± 1.51 to $21.0\,0 \pm 1.53$ days in extract-treated groups, compared to 27.00 ± 0.16 days and 27.33 ± 1.00 days in chloroquine- and artesunate-treated groups, respectively, and 10.33 ± 0.46 days in the untreated control. Quantitative phytochemical screening revealed appreciable levels of alkaloids (119.0 ± 2.0 mg/g), saponins (42.0 ± 0.67 mg/g), tannins (26.33 ± 0.56 mg/g), flavonoids (24.50 ± 0.79 mg/g), and phenols (6.52 ± 0.24 mg/g). These findings demonstrate that *C. papaya* leaf extract possesses significant antiplasmodial activity, likely attributable to its rich alkaloid content, and suggest its potential as a promising source of alternative antimalarial agents.

Keywords: *Carica papaya*, *Plasmodium berghei*, Antimalarial activity, Phytochemical, Traditional medicine

INTRODUCTION

Malaria is a parasitic disease caused by the parasite *Plasmodium*. Malaria has proven to be one of the most devastating illnesses of all time and is the most common parasitic disease in sub-Saharan Africa. It was reported that 263 million cases of malaria occurred worldwide in 2023 and the disease led to 597 000 deaths particularly in children under 5 years of age (WHO, 2024). Nigeria accounted for highest malaria burden and death sharing the global malaria cases of 26% and 31% death in 2023 (WHO, 2024). In Nigeria, malaria accounts for 60% morbidity, 30% childhood death and 11% maternal mortality (NFMH, 2014; Odoko *et al*., 2020; Alozieuwa *et al.*, 2022). Nigeria accounted for 39.3% of global malaria deaths in children aged under 5 years (WHO, 2024). The human and economic costs associated with declining quality of life, treatment, prevention, loss of man-hours, consultations, hospitalizations and other events related to malaria are huge and often lead to low productivity and lost incomes estimated at 132 billion Naira annually (Odoko *et al*., 2020). Malaria is considered as not only a public health problem but also a deterrent to the socioeconomic growth of a country. The main problem of global malaria control efforts at the present is the development of partial resistance to artemisinin evidenced by delayed parasite clearance following treatment with artemisinin-containing drugs and resistance to the partner drugs used in artemisinin-based combination therapies (ACTs) (WHO, 2024). Meanwhile, the effectiveness of malaria control strategies is increasingly compromised by the growing resistance of mosquito vectors to commonly used insecticides (WHO, 2024). Additionally, the high cost and potential adverse effects associated with some existing antimalarial drugs limit their widespread use.

Traditional medicines for malaria treatment are already in popular use in developing countries including Nigeria. To address the challenges associated with malaria chemotherapy, such as drug resistance and limited efficacy, there is a renewed emphasis on exploring natural products as potential sources for antimalarial drug development (Uzor *et al*., 2020). Notably, plant-derived compounds like artemisinin and quinine have historically played pivotal roles in malaria treatment (Uzor *et al*., 2020; Kingston and Cassera, 2022). Recent studies continue to investigate various natural products, including those from marine organisms and medicinal plants, for their antimalarial properties and potential to inspire novel therapeutic agents (Negm *et al*., 2022). It is widely recognized that only about 10% of all plant species have been thoroughly investigated for their phytochemical constituents. This underscores the importance of exploring the phytochemicals present in medicinal plants used in traditional medicine, as these compounds may serve as valuable leads for the development of novel antimalarial drugs

Carica papaya, commonly known as pawpaw, belongs to the plant family Caricaceae. Various parts of the *C. papaya* plant, including the fruit, leaves, seeds, and bark, have been extensively studied for their therapeutic potentials. It has been used widely in folk medicine for many ailments including fever, asthma, colica, beriberi and jaundice (Singh *et al*., 2020). Extracts from *Carica papaya* have been reported to exhibit a wide range of pharmacological activities, including antibacterial, antiplasmodial, immunomodulatory, anti-dengue, hypoglycemic, gastroprotective, and anticancer

effects (Norahmad *et al*., 2019; Singh *et al*., 2020; Atanu *et al*., 2021). Despite these promising findings, the phytochemical composition alongside evaluation of in vivo therapeutic efficacy of *C. papaya* leaf extract in established malaria infection remain insufficiently characterized. The present study sought to investigate the phytochemical constituent of the leaf extract of *Carica papaya* and to evaluate its therapeutic potential in established *Plasmodium berghei* infection in mice, a widely accepted model for studying human malaria.

MATERIALS AND METHODS

Plant Samples

Fresh leaves of *Carica papaya* was collected in July from Minna, Niger State, Nigeria. The plant was identified by a taxonomist at the Botany Department of University of Nigeria Nsukka, Enugu state. A voucher number of UNH NO 602[a] was assigned and deposited at the herbarium of Botany Department of University of Nigeria, Nsukka, Enugu State, Nigeria.

Parasites

Plasmodium berghei (NK65) was obtained from the National Institute of Pharmaceutical Research and Development (NIPRD), Abuja, Nigeria. It was maintained in mice by serial blood passage.

Animals

The experiment was conducted in compliance with the internationally accepted principles for laboratory animal use and care as contained in the Canadian Council on Animal Care (CCAC) guidelines on animal use protocol review (1997). Adult Swiss albino mice, male and female, weighing between 22-28g were obtained from National Veterinary Research Institute (NVRI), Vom, Plateau State of Nigeria and were acclimatized in the experimental animal house for two weeks. Animals were fed with standard mice pellet diet (Vital feeds, Nigeria) and water *ad libitum*.

Preparation of Crude Extract

Fresh leaves of *Carica papaya* were collected, washed, air dried at room temperature to a constant weight and pulverised using a blender. Fifty grams of the powdered sample was extracted under reflux in 400ml of 70% methanol for 2 h with medline extraction mantle according to the method of Ugwu *et al*. (2011). The extracts were filtered hot through muslin cloth, and then concentrated to dryness under reduced pressure using a rotary evaporator. The extracts were transferred into sterile universal tubes, and kept in the refrigerator until required for use.

Qualitative Phytochemical Analysis

Qualitative phytochemical screening of the crude extracts was carried out using standard procedures to identify the secondary metabolites, as outlined by (Evans, 1989; Sofoworo, 1993).

Quantitative Phytochemical Determination

Total alkaloid was determined using gravimetric method of Harborne described by Sharma *et al*. (2021) and colorimetric method as described by Win *et al*. (2019) was used in the determination of total flavonoids. The total phenolic content was estimated by Folin- Ciocalteu photometric method (Seglab *et al*., 2021) while AOAC (1990) method was used for tannin determination. Gravimetric method as described by Murtiningsih *et al*. (2023) was used for the determination of saponin.

Parasite Inoculation

Blood was collected from the tail vein of a donor mouse previously infected with *Plasmodium berghei* and exhibiting a parasitemia level of 20–30%. The blood was then diluted with 0.9% normal saline to prepare a suspension containing 10^7 parasitized red blood cells per 0.2 ml. Each healthy mouse was subsequently inoculated intraperitoneally with 10^7 *P. berghei*-infected RBCs, following the method described by Alozieuwa *et al*. (2018).

Antimalarial activity of the Aqueous Methanol extract of *Carica papaya*

The curative potential of plant extracts was assessed using the method described by Ryley and Peters (1970). On day 0, 18 albino mice (both sexes) were intraperitoneally infected with a standard inoculum of Plasmodium berghei-infected red blood cells. After 72 hours (day 3), following confirmation of parasitemia, the infected mice were randomly divided into seven groups of three. The groups were orally treated with the plant extract at doses of 100 (Group A), 300 (Group B), and 500 (Group C) mg/kg/day. Positive control groups (E & F) received 5 mg/kg/day chloroquine and 50 mg/kg/day artesunate, respectively, while Group G (negative control) was given 0.2 ml of distilled water. Group D, which was not infected, received 500 mg/kg of the extract. Treatments continued for 5 days. Giemsa-stained (10%) thin blood films, prepared from blood samples drawn from the tail vein of each mouse, were microscopically examined. The percentage of parasitemia and inhibition were calculated using the formula described by (Peters *et al*., 1975):

$$\% \text{ parasitemia} = \frac{\text{Number of parasitized erythrocytes}}{\text{Number of erythrocytes}} \times 100$$

$$\% \text{ inhibition} = \frac{A-B}{A} \times 100$$

Where:

A = Parasitaemia in negative control

B = Parasitaemia in test group

Weight Determination

The body weight of each mouse in all the groups was taken before infection (day 0) and after treatment (day 8) using a sensitive weighing balance.

Determination of Packed Cell Volume

Blood samples were obtained from the tail of each mouse using heparinized microhematocrit capillary tubes. The tubes were filled to about three-quarters of their capacity with blood and sealed using crystal seal. The tubes were then positioned in a microhematocrit centrifuge with the sealed ends facing outward and centrifuged for 5 minutes at 11,000 rpm. The packed cell volume (PCV) percentage was recorded both before parasite inoculation (day 0) and after the treatment period (day 8).

Determination of Mean Survival Time

The experimental mice were monitored daily for mortality, and the number of days from *Plasmodium berghei* infection to death was recorded for each mouse in both the treated and control groups throughout the study period.

Mean Survival Time (MST) of all mice were determined using the formula:

$$\text{MST} = \frac{\text{Sum of survival time of all mice in a group (days)}}{\text{Total number of mice in that group}}$$

Statistical Analysis

The study results were presented as mean ± SEM. Data analysis was conducted using SPSS software (Version 19.0) for Windows. To assess statistical significance, a one-way

analysis of variance (ANOVA) followed by Duncan's post-hoc test was performed. A p-value of less than 0.05 at a 95% confidence level was considered statistically significant.

RESULTS AND DISCUSSION

Qualitative Phytochemical Analysis

Phytochemical screening of the crude leaf extract of C. papaya identified the presence of alkaloids, flavonoids, steroids, tannins, phenols, glycosides, and saponins, whereas terpenoids were not detected.

Quantitative Phytochemical Analysis

Quantitative phytochemical analysis of the extract showed that alkaloids composition was highest while phenol was the least (Table 1).

Table 1: Phytochemical constituents of the aqueous methanol extract of *C. papaya*

Phytochemicals	Concentration (mg/g)
Flavonoids	24.50±0.79
Tannins	26.33±0.56
Phenols	6.52±0.24
Alkaloids	119.0±2.0
Saponins	42.0±0.67

Values represent the mean ± SEM, n = 3.

Antiplasmodial Activity of Crude Leaf Extract of *Carica papaya*

All three doses of the *Carica papaya* leaf extract exhibited significant ($p < 0.05$) antiplasmodial activity in a dose-dependent manner compared to the negative control (Figure 1). The highest parasite growth inhibition (59.89%) was observed in the group administered 500 mg/kg of the extract (Table 2). Although there was a reduction in body weight among the extract-treated groups, it was not statistically significant ($p > 0.05$), unlike the negative control group, which showed a significant weight loss (Figure 2). Changes in packed cell volume (PCV) were not significant ($p > 0.05$) across all groups, except in the group treated with 100 mg/kg of the extract and the untreated control, where significant reductions were noted (Figure 3). A dose-dependent prolongation of mean survival time was observed in all extract-treated groups (Table 2).

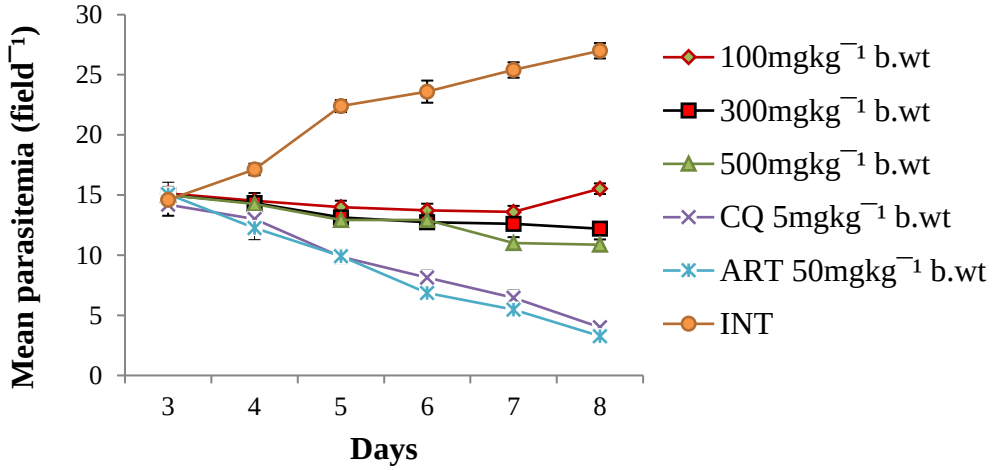

Figure 1: Antiplasmodial activity of aqueous methanol leaf extract of *Carica papaya*
Data are mean ± SEM; CQ = chloroquine, ART = artesunate, INT = infected but not treated (untreated control)

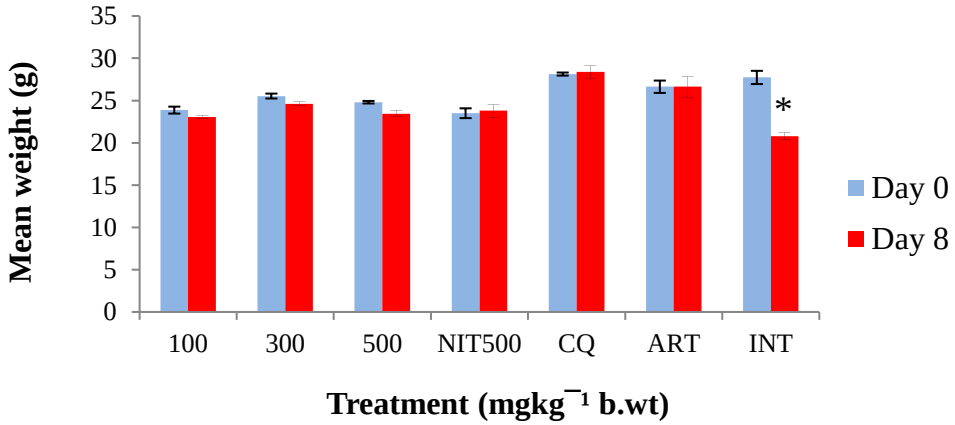

Figure 2: Effect of aqueous methanol leaf extract of *C. papaya* on body weight of *P. berghei* infected mice
Data are mean ± SEM; CQ = chloroquine, ART = artesunate, NIT = Not infected but treated, INT = infected but not treated, * = significant difference between the body weight before and after treatment.

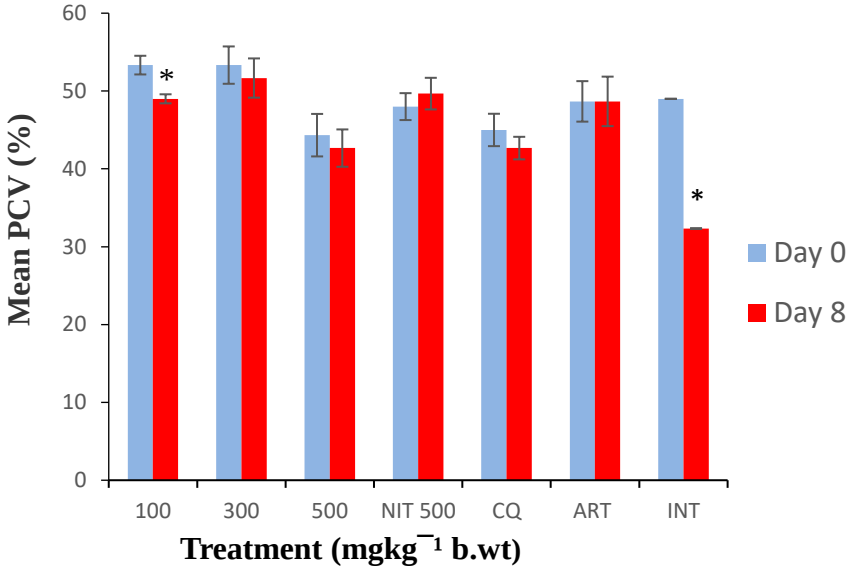

Figure 3: Effect of aqueous methanol extract of *Carica papaya* on PCV of *P. berghei* infected mice
Data are mean ± SEM; CQ = chloroquine, ART = artesunate, NIT = Not infected but treated, INT = infected but not treated, * = significant difference between the %PCV before and after treatment.

Table 2: Effect of aqueous methanol extracts of *Carica papaya* on the % inhibition of parasite growth and survival time of *P. berghei* infected mice

Dose (mg/kg)	% Parasitemia (day 8)	% Inhibition	Mean survival time (days)
100	15.53±0.44	42.69	17.00±1.51
300	12.20±0.31	54.98	18.67±1.76
500	10.87±0.44	59.89	21.00±1.53
CQ 5	4.00±0.07	85.24	27.00±0.16
Art 50	3.27±0.24	87.93	27.33±1.00
INT	27.10±0.64	0	10.33±0.46

Values are expressed as mean ± SEM, n = 3, Art = Artesunate, CQ = Chloroquine diphosphate, INT = untreated control

Discussion

Traditional medicines for malaria treatment are already in popular use in developing countries including Nigeria. Notably, plant-derived compounds like artemisinin and quinine have historically played pivotal roles in malaria treatment (Uzor *et al*., 2020; Kingston and Cassera, 2022). The results indicate that the crude extract of leaf of *Carica papaya* has significant antimalarial activity which was apparent from the inhibitory effect of the extract in established infection (Fig. 1, Table 2). Previous report revealed that the crude aqueous leaf extract of *C. papaya* significantly suppressed parasites when administered to *P. berghei* infected mice in early infection (Atanu *et al*., 2021). However, Amazu *et al*. (2009) noted in the curative study that *Carica papaya* seed extract at doses of 200, 100 and 50 mg/kg/day failed to inhibit malaria parasite. The observed antiplasmodial activity of *C. papaya* leaf extract in established infection in our study, suggests that the active ingredients against *P. berghei* parasite is resident in the leaf of *C. papaya* rather than the seed. Although our study evaluated the single-plant extract of *Carica papaya*, related research has shown that a decoction containing *Psidium guajava*, *Mangifera indica*, and *C. papaya* leaves also ameliorated the effects of *P. berghei* infection in a dose-dependent manner, likely due to phytochemical synergy (Bashir *et al*., 2024).

Rodent malaria is known to cause a reduction in packed cell volume (PCV), typically occurring around 48 hours after infection (Nardos and Makonnen, 2017; Alozieuwa *et al*., 2022). Mice infected with *Plasmodium berghei* often develop anaemia due to the destruction of red blood cells, which can result from the parasite's replication or the action of reticuloendothelial cells in the spleen. The presence of numerous abnormal erythrocytes stimulates the spleen to generate increased numbers of phagocytic cells (Somsak *et al*., 2016). These processes collectively contribute to malaria-induced anaemia in both mice and humans (Lamikanra et al., 2007; Alozieuwa *et al*., 2018). The crude extract of *C. papaya* leaf extract significantly prevented body weight loss and PCV reduction associated with increase in parasitemia level. However, a significant reduction in PCV was observed in the group treated with the lowest dose (100mg/kg/day) of the extract (Fig. 3). This could mean that the active secondary metabolites may not be in a sufficient concentration in that dose to significantly reduce parasite load and prevent parasite-induced fall of PCV.

The mean survival time of extract-treated mice was significantly longer than the untreated control mice (Table 2), indicating that the extract inhibited the growth of *P. berghei* and reduced the overall pathologic effect of the parasite on the mice. In the untreated control, a daily rise in parasitemia was observed until the death of the animals.

A reduction in body weight gain and packed cell volume (PCV) is recognized as a sensitive marker in toxicological evaluations following exposure to potentially harmful agents (Nardos and Makonnem, 2017). In this study, administration

of the crude extract to healthy, non-infected mice at a dosage of 500 mg/kg body weight daily for five days did not produce any noticeable toxic effects (Fig. 2). This was evidenced by a slight increase in both body weight and PCV, along with 100% survival throughout the 30-day observation period. These findings suggest that the crude extract is relatively safe for oral use at the tested dosage.

The quantitative phytochemical analysis showed that alkaloid is in appreciable quantity (Table 1) and alkaloids have been implicated in antiplasmodial activities of many plants as reported by other investigators (Ghildyal *et al*., 2010; Otimenyin and Umar, 2012; Tadesse and Wubneh, 2017; Uzor, 2020). Alkaloids are believed to interfere with protein synthesis in *Plasmodium falciparium* (Nergz and Otles, 1993; Okokon *et al*., 2011; Ettebong *et al*., 2012). Therefore, the possible active compounds responsible for the antimalarial activity of *C. papaya* may be alkaloids. Isolation and characterisation of alkaloids from *C. papaya* for antimalarial agent is recommended.

CONCLUSION

This study demonstrated that the aqueous methanol leaf extract of *Carica papaya* contains several phytochemicals, with alkaloids being the most abundant, followed by saponins, tannins, flavonoids, and phenols. The extract exhibited significant, dose-dependent antiplasmodial activity in *Plasmodium berghei*-infected mice, with the highest activity observed at 500 mg/kg (59.89% inhibition). Additionally, the extract mitigated malaria-induced weight loss and reduction in packed cell volume, prolonged survival, and showed no observable toxicity. These findings suggest that *C. papaya* leaf extract possesses considerable inherent antimalarial activity in established rodent infection, which can be attributed to its phytochemical compounds. Therefore, isolation of these phytochemicals may lead to an alternative antimalarial.

REFERENCES

Alozieuwa, U. B., Mann, A., Kabiru, A. Y., & Ogbadoyi, E. O. (2018). Antimalarial Activity of the Crude Extract and Fractions of *Phyllanthus amarus* in *Plasmodium berghei*-infected mice. *European Journal of Medicinal Plants*, *24*(3):1-11. https://doi.org/10.9734/EJMP/2018/38578

Alozieuwa, U. B., Mann, A., Kabiru, A. Y., & Ogbadoyi, E. O. (2022). In vivo antimalarial efficacy of *Psidium guajava* leaf crude ex tract and fractions in *Plasmodium berghei* infected mice. *AROC in Natural Products Research*, *2*(1): 28-37. https://doi.org/10.1155/2022/1215097

Amazu, L. U., Ebong, O. O., Azikiwe, C. C. A., Unekwe, P. C., Siminialayi, M. I., Nwosu, P. J. C., Ezeani, M. C., Obidiya, O. S. and A. O. Ajugwo (2009). Effects of the methanolic seeds extract of *Carica papaya* on *Plasmodium berghei* infected mice. Asian Pacific Journal of Tropical Medicine, 2(3), 1-6.

AOAC (Association of Official Analytical Chemists). (1990). Official methods of analysis. *Assoc Anal Chem*, *62*, 2742-2744.

Atanu, F. O., Idih, F. M., Nwonuma, C. O., Hetta, H. F., Alamery, S., & El-Saber Batiha, G. (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*(1), 2599191. https://doi.org/10.1155/2021/2599191

Bashir, A., Bello, M. I., Mahmoud, S. J., & Zailani, A. H. (2024). Antimalarial Activity of Decoction from *Carica Papaya*, *Psidium Guajava* and *Mangifera Indica* Leaves in Mice Infected with *Plasmodium Berghei* (Nk-65). *Fudma Journal of Sciences*, *8*(3), 477-486.

CCAC (1997). Guidelines on Animal use and Protocol review. http://www.ccac.ca/Documents/Standards/Guidelines/Protocol_Review.pdf.

Ettebong, E. O., Nwafor, P. A. J. E., & Okokon (2012). In vivo antiplasmodial activities of ethanolic exract and fractions of *Eleucine indica. Asian Pacific Journal of Tropical Medicine*, *2*, 673-676. https://doi.org/10.1016/S1995-7645(12)60105-9

Evans, W. C. (2009). Trease and Evans' pharmacognosy. Elsevier Health Sciences.

Ghildyal, P., Grønhaug, T. E., Rusten, A., Skogsrud, M., Rolstad, B., Diallo, D., ... & Paulsen, B. S. (2010). Chemical composition and immunological activities of polysaccharides isolated from the Malian medicinal plant *Syzygium guineense*. *Journal of Pharmacognosy and Phytotherapy*, *2*(6), 76-85.

Harborne, A. J. (1998). Phytochemical methods a guide to modern techniques of plant analysis. springer science & business media.

Kingston, D. G., & Cassera, M. B. (2022). Antimalarial natural products (pp. 1-106). Springer International Publishing.

Lamikanra, A. A., Theron, M., Kooij, T. W., & Roberts, D. J. (2009). Hemozoin (malarial pigment) directly promotes apoptosis of erythroid precursors. *PloS one*, *4*(12), e8446. https://doi.org/10.1371/journal.pone.0008446

Murtiningsih, T., Supriningrum, R., & Nurhasnawati, H. (2023). Identification and determination of saponin content from extract of *Embelia borneensis* bark. *Al Ulum: Jurnal Sains Dan Teknologi*, *9*(3), 117-126.

Nardos, A., & Makonnen, E. (2017). In vivo antiplasmodial activity and toxicological assessment of hydroethanolic crude extract of *Ajuga remota*. *Malaria Journal*, *16*, 1-8. https://doi.org/10.1186/s12936-017-1677-3

Negm, W. A., Ezzat, S. M., & Zayed, A. (2023). Marine organisms as potential sources of natural products for the prevention and treatment of malaria. *RSC advances*, *13*(7), 4436-4475. https://doi.org/10.1186/s12936-017-1677-3

Nergiz, C., & Ötleş, S. (1993). Chemical composition of *Nigella sativa* L. seeds. *Food chemistry*, *48*(3), 259-261. https://doi.org/10.1016/0308-8146(93)90137-5

Nigeria Federal Ministry of Health (2014). National Malaria Control Programme Strategic plan 2014–2020: a road map for malaria control in Nigeria, Abuja: Yaliam Press Ltd, Federal Ministry of Health.

Norahmad, N. A., Mohd Abd Razak, M. R., Mohmad Misnan, N., Md Jelas, N. H., Sastu, U. R., Muhammad, A., & Syed Mohamed, A. F. (2019). Effect of freeze-dried *Carica papaya* leaf juice on inflammatory cytokines production during dengue virus infection in AG129 mice. *BMC complementary and alternative medicine*, *19*, 1-10. https://doi.org/10.1186/s12906-019-2438-3

Odoko, J. O., Nwose, E. U., Nwajei, S. D., Agege, E. A., Moyegbone, J. E., & Igumbor, E. O. (2020). Epidemiology of malaria as it relates to utilization of insecticide treated nets among pregnant women and under five years children in South-South Nigeria. *International Journal of Community Medicine and Public Health*, *7*(10), 7. https://doi.org/10.18203/2394-6040.ijcmph20204390

Okokon, J. E., Etebong, E. O., Udobang, J. A., & Obot, J. (2012). Antiplasmodial and antiulcer activities of *Melanthera scadens*. *Asian Pacific Journal of Tropical Biomedicine*, *2*(1), 16-20. https://doi.org/10.1016/S2221-1691(11)60182-2

Otimenyin, O., & Umar, M. (2012). Anti-inflamatory and analgesic activities of the ethanolic extract of the leaf of *Syzygium guineense* in rats and mice. *International organisation of Scientific Research Journal of Pharmacy*, *2*, 33–36.

Peters, W., Portus, H. and L. Robinson (1975). The four-day suppressive in vivo antimalarial test. *Annal of Tropical Medicine and Parasitology*, *69*, 155–171.

Ryley, J, F. and W. Peters (1970). The antimalarial activity of some quinoline esters. Annal of Tropical Medicine and Parasitology, *84*, 209-222.

Seglab, F., Hamia, C., Khacheba, I., Djeridane, A., & Moyse, F. (2021). High in vitro antioxidant capacities of Algerian *Cleome arabica* leaves' extracts. *Phytothérapie*, *19*(1), 16-24. https://doi.org/10.3166/phyto-2019-0179

Sharma, T., Gamit, R., Acharya, R., & Shukla, V. J. (2021). Quantitative estimation of total tannin, alkaloid, phenolic, and flavonoid content of the root, leaf, and whole plant of *Byttneria herbacea* Roxb. *AYU (An International Quarterly Journal of Research in Ayurveda)*, *42*(3), 143-147. https://doi.org/10.4103/ayu.AYU_25_19

Singh, S. P., Kumar, S., Mathan, S. V., Tomar, M. S., Singh, R. K., Verma, P. K., & Acharya, A. (2020). Therapeutic application of *Carica papaya* leaf extract in the management of human diseases. *DARU Journal of Pharmaceutical Sciences*, *28*, 735-744. https://doi.org/10.1007/s40199-020-00348-7

Sofoworo, A. (1993). Medicinal plants and traditional medicine in Africa. *J Altern Complement Med*, *13*, 195-238.

Somsak, V., Polwiang, N., & Chachiyo, S. (2016). In vivo antimalarial activity of *Annona muricata* leaf extract in mice infected with *Plasmodium berghei*. *Journal of pathogens*, *2016*(1), 3264070. https://doi.org/10.1155/2016/3264070

Tadesse, S. A., & Wubneh, Z. B. (2017). Antimalarial activity of *Syzygium guineense* during early and established *Plasmodium* infection in rodent models. *BMC complementary and Alternative Medicine*, *17*, 1-7. https://doi.org/10.1186/s12906-016-1538-6

Ugwu, U. B., Okogun, J. I., Kabiru, A. Y. and E. O. Ogbadoyi (2011). Evaluation of Therapeutic Potentials of Stem Bark Extracts of *Annona senegalensis* in Experimental *Trypanosoma brucei brucei Infection* in Mice. *British Journal of Pharmacology and Toxicology*, *2*(2), 63-70.

Uzor, P. F. (2020). Alkaloids from plants with antimalarial activity: a review of recent studies. *Evidence-Based Complementary and Alternative Medicine*, *2020*(1), 8749083. https://doi.org/10.1155/2020/8749083

Uzor, P. F., Prasasty, V. D., & Agubata, C. O. (2020). Natural products as sources of antimalarial drugs. *Evidence-based Complementary and Alternative Medicine: eCAM*, *2020*, 9385125. https://doi.org/10.1155/2020/9385125

WHO (2024). World Health Organization Fact sheet on the World Malaria Report https://www.who.int/teams/global-malaria-programme/reports/world-malaria-report-2024

Win, A., Thida N. A., Lwin, K.M. (2019). Determination of total flavonoid content of commonly consumed commercial tea. *International Journal of Scientific and Research Publications (IJSRP)*, *9*(5).