A Study of Malaria Infection Suppression by Bioactive Compounds from Extracts of a Tropical Medicinal Plant

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Abstract: The properties exhibited by natural products from plants, including the medicinal activity, can be ascribed to the type and nature of the biologically active compounds they contain. The scientific study of these bioactive compounds is a very important step in the research to discover new molecules for potential drugs development from such natural products. This study was carried out with the aim of investigating the malaria infection suppression effect of bioactive compounds from *Alstonia boonei,* a plant that is used in the traditional treatment of malaria in many African countries. There is scarcity of studies that have evaluated bioactive from *Alstonia boonai* with most of them ending only on evaluating the extracts, thus the importance of this study. **T**he bioactive compounds present in the ethanolic and aqueous leaf, stem bark and root extracts of *Alstonia booei* were determined and identified by Gas Chromatography and Mass Spectrometer methods using the Angilent 6890 series GC-MS equipment. Antimalarial tests were carried out on the bioactive compounds using white albino mice infected with *Plasmodium berghei* and microscopy method to investigate their malaria infection suppression activity. The antimalarial tests carried out on these bioactive compounds revealed that they possess significant suppressive activity (at $p < 0.05$) against malaria parasite infection in mice which was dose-dependent. From this result it was concluded that the plant is a potential source for new antimalarial molecules and should be further investigated for antimalarial drug development.

Key word: Antimalarial drug, bioactive compounds, malaria infection**,** *Plasmodium berghei,* suppression

INTRODUCTION

Malaria is a devastating and infectious disease that kills millions of people worldwide especially in Africa where it still remains endemic (Imieje and Amafili, 2025). *Plasmodium falciparum* causes the majority of morbidity and mortality of the malaria disease (Afolabi and Abejide, 2020). The rise of resistance cases to commonly used antimalarial drugs is a threat to the human race (Lestari *et al.,* 2023). There is therefore a very pressing need to identify and develop new antimalarial compounds (Noronha *et al.,* 2020). The burden of this disease is getting worse, mainly due to the increasing resistance of *Plasmodium falciparum* against the widely available antimalarial drugs (Lestari *et al.,* 2023; Dibua *et al.,* 2013). There is an urgent need for new, more affordable and accessible antimalarial agents possessing original modes of action (Nanven *et al.,* 2024). Products from nature, such as those from plants, play important roles as leads for the discovery and development of new drugs **(**Madhiri and Vijayalakshini, 2018; Muhseen *et al.,* 2021*). Alstonia boonei* belongs to the family Apocynaceae and is an herbal medicinal plant of West African origin (Olanlokun *et al.,* 2021).

Alstonia boonei parts have been used for the complementary treatment of malaria and other forms of diseases in West African Countries (Iyiola *et al.,* 2011; Moronkola and Kunle, 2012). Tropical rain forest plants have high amounts of bioactive concentrations of natural chemical diversity, and as a result they are potential sources of new molecules (Farooqi *et al.,* 2024). Thus studies on plants from the African region is encouraged since the burden and impact of malaria is heaviest in the African region (Adebayo and Krettli, 2011). Hence, the aim of this study which is to investigate the early malaria infection suppression effect of bioactive compounds from *Alstonia boonei*. This study is important as it has the potential of yielding new molecules for the development of new antimalarial drugs with novel modes of action different from that of the currently used drugs against malaria. This would contribute immensely in combating drug resistant malaria which has been on the increase in recent times.

MATERIALS AND METHODS

Plant Parts Collection, Cleaning and Extraction Methods: The leaf, stem bark and root of *Alstonia boonei* were collected from Obukpa village in Nsukka Local Government Area in Enugu State, Nigeria. The plant parts were identified and authenticated by a botanist expert in the Department of Plant Science and Biotechnology, University of Nigeria Nsukka, Enugu State, Nigeria. They were prepared by using the modified methods described in an earlier study (Afolabi and Abejide, 2020). The plant parts were separately cut into small pieces, washed and air dried for two weeks under room temperature. The dry samples were then ground into powder with well cleaned and dry grinding machine. Aqueous and ethanolic extraction were then carried out using modified procedures described in a previous study (Amole and Ilori, 2010). For the aqueous extraction, a quantity of 500 g of the ground fine powder obtained was percolated in 1600 mL of water for 72h after which it was filtered. This was followed by evaporating the filtrate collected to dryness using a temperature-regulated water bath pre-set at 40^{o}C to yield the extract concentrate which was stored in the refrigerator at 4^{o}C before use (Amole and Ilori, 2010). For the ethanolic extraction, another quantity of 500 g of the ground plant material powder was measured and dispersed in 2.5 L of ethanol. The mixture was shaken with a mechanical shaking machine (GFL shaker No. 3017 MBH, Germany) for 72 h after which it was vacuum filtered. The resultant extract was then concentrated using a rotary evaporator at a temperature not exceeding 400^{o}C. The concentrate was then heated over a temperature-regulated water bath pre-set at 40^{o}C to obtain a solvent-free extract. The extract thus obtained was stored in the refrigerator at 4^{o}C until use (Amole and Ilori, 2010).

Animals for the Study: White albino mice of both sexes were used for this study. They were obtained from the Animal Department, Nigerian Institute of Pharmaceutical Research and Development, Idu, Abuja, Nigeria. Animal tests were carried out according to the National Institute of Health (NIH) Guide for the Care and Use of Laboratory Animals. Approval for all protocols was obtained for all animal experiments from the University of Nigeria Ethical Committee on the use of laboratory animals for research with approval number UNN/ZEB/22/6804.

Plasmodium Parasite for the Study: The parasite used for this study was *Plasmodium berghei* NK65 strain, which was sensitive to artesunate. The parasite was obtained from the Animal Department, Nigerian Institute of Pharmaceutical Research and Development, Idu, Abuja, Nigeria.

Gas Chromatography-Mass Spectrometer (GC-MS) Analysis of the Extracts for Bioactive compounds: **T**he bioactive compounds present in the ethanolic and aqueous leaf, stem bark and root extracts of *Alstonia booei* were determined and identified by Gas Chromatography and Mass

Spectrometer methods using (Angilent 6890 series) equipment following the procedures described by Eswarainh *et al.,* 2019. The equipment had a HP-5MS column mass spectrometer operated at initial column temperature of 30°C and heated up to 300°C at the rate of an increase of 10°C/min and maintained for 10 min. Injection port temperature was ensured at 250°C and Helium flow rate at 1.5 ml/min. The ionization voltage was 70 ev. The samples were injected in split mode of 10:1. Mass spectral scan range was set at 40-700 m/z. The ion source temperature was maintained at 230°C and Interface temperature was set at 240°C. The MS start time was 3 min and end time was 40 min with solvent cut time of 3 min. The identification of the compounds was done based on retention time, integral area of peaks and by using the database of National Institute Standard and Technology (NIST). The similarity of compounds matched with >70% listing based on NIST 59 library search. The most active isolated compounds thus analyzed and identified were stored in the refrigerator at 4^{o}C until use for antimalarial tests.

Tests for Malaria Infection Suppression Activity by the Bioactive compounds: The bioactive compounds underwent antimalarial tests in *P. berghei* infected mice in order to determine their suppressive effects on malaria infection using methods described by Dibua *et al.,* 2013, which was modified to suit the aim of this study. On the first day a set of mice of both sexes were randomly selected and infected with 10 ^{7} *Plasmodium berghei*. Three hours later blood was taken from the tail of each infected mice and examined under the microscope to determine parasitaemia of early infection and then the infected mice were each treated orally with 100, 200 and 400 mgkg^{-1} body weights of the bioactive compounds or 5mgkg^{-1} body weight of artesunate, the positive control, separately using corn meal as the vehicle. Another set of infected mice, the negative control, were given 5mlkg^{-1} distilled water. Treatment was continued for four consecutive days and on the fifth day, blood was again taken from the tail of each mice and examined for suppression of parasitaemia and the values were recorded accordingly.

Statistical Analysis: The values obtained was analyzed using Statistical Package for the Social Science (SPSS) version 20. The mean values of the parameters studied were compared using one-way Analysis of Variance (ANOVA) at 95% confidence interval, and separated using Turkey-b post hoc comparison. Probability values of $p < 0.05$ were considered significant. Results were expressed as mean +/- standard error of mean (SEM).

RESULTS

This study was carried out in order to investigate the malaria infection suppression activity by bioactive compounds from extracts of *Alstonia boonei* plant. The GC-MS analysis of the extracts yielded important bioactive compounds and the most active ones according to the activity spectra were identified and tested for their malaria infection suppression activity. The results of the tests are stated below. Bioactive compounds found in the extracts belong to different classes including phenols, esters, carboxylic acids, glycocids, terpenoids, acridones and lactones. The selected most active compounds based on the spectra from the products of the GC-MS analysis are 1,3,5-Triazine, 2-methylamino-4,6-bis(nonafluoro-tert-butyl), a compound from aqueous leaf extract of *A. boonei*, androsta--2,4,16-triene-3,6,17-triol, tri-TMS, a compound from aqueous stem bark extract of *A. boonei*, 1-Oxo-forskolin, a compound from aqueous root extract of *A. boonei*, Aldosterone, N-methoxy-tri-TMS, a compound from ethanolic leaf extract of *A. boonei*, Hecogenin acetate, a compound from ethanolic stem bark extract of *A. boonei* and βeta-Dithiodilactic acid, a compound from ethanolic root extract of *A. boonei*. The antimalarial tests for *Plasmodium berghei* infection suppression effect of the bioactive compounds revealed a significant dose-dependent antimalarial activity as indicated in the suppressions recorded as shown in Tables 1, 2, 3. 4, 5 and 6 below.

Table 1: Suppression activity of 1,3,5-Triazine, 2-methylamino-4,6-bis(nonafluoro-tert-butyl), a compound from aqueous leaf extract of *A. boonei* and artesunate in mice infected with *Plasmodium berghei*

Treatments	Suppression (%)
Distilled water 5mlkg^{-1}	0.00 ± 0.00^{a}
Compound 100mgkg^{-1}	34.31 ± 0.64^{b}
Compound 200mgkg^{-1}	48.45 ± 0.63^{c}
Compound 400mgkg^{-1}	65.87 ± 0.64^{d}
Artesunate 5mgkg^{-1}	86.73 ± 0.63^{e}

[1]All values expressed as mean ± standard error (±SE).
[2]Different superscript letters indicated significance difference ($p < 0.05$) in mean values among different treatments using Turkey's b *post hoc* comparison.

Table 2: Suppression activity of androsta-2,4,16-triene-3,6,17-triol, tri-TMS, a compound from aqueous stem bark extract of *A. boonei* and artesunate in mice infected with *P. berghei*

Treatments	Suppression (%)
Distilled water 5mlkg^{-1}	0.00 ± 0.00^{a}
Compound 100mgkg^{-1}	48.71 ± 0.64^{b}
Compound 200mgkg^{-1}	59.64 ± 0.64^{c}
Compound 400mgkg^{-1}	68.31 ± 0.63^{d}
Artesunate 5mgkg^{-1}	94.52 ± 0.63^{e}

[1]All values expressed as mean ± standard error (±SE).
[2]Different superscript letters indicated significance difference ($p < 0.05$) in mean values among different treatments using Turkey's b *post hoc* comparison.

Table 3: Suppression activity of 1-Oxo-forskolin, a compound from aqueous root extract of *A. boonei* and artesunate in mice infected with *Plasmodium berghei*

Treatments	Suppression (%)
Distilled water 5mlkg^{-1}	0.00 ± 0.00^{a}
Compound 100mgkg^{-1}	44.15 ± 1.21^{b}
Compound 200mgkg^{-1}	50.63 ± 0.64^{c}
Compound 400mgkg^{-1}	62.31 ± 0.64^{d}
Artesunate 5mgkg^{-1}	94.22 ± 1.21^{e}

[1]All values expressed as mean ± standard error (±SE).
[2]Different superscript letters indicated significance difference ($p < 0.05$) in mean values among different treatments using Turkey's b *post hoc* comparison.

Table 4: Suppression activity of Aldosterone, N-methoxy-tri-TMS, a compound from ethanolic leaf extract of *A. boonei* and artesunate in mice infected with *Plasmodium berghei*

Treatments	Suppression (%)
Distilled water 5mlkg^{-1}	0.00 ± 0.00^{a}
Compound 100mgkg^{-1}	41.19 ± 1.79^{b}
Compound 200mgkg^{-1}	49.92 ± 0.59^{c}
Compound 400mgkg^{-1}	62.05 ± 0.92^{d}
Artesunate 5mgkg^{-1}	91.03 ± 1.44^{e}

[1]All values expressed as mean ± standard error (±SE).
[2]Different superscript letters indicated significance difference ($p < 0.05$) in mean values among different treatments using Turkey's b *post hoc* comparison.

Table 5: Suppression activity of Hecogenin acetate, a compound from ethanolic stem bark extract of *A. boonei* and artesunate in mice infected with *Plasmodium berghei*

Treatments	Suppression (%)
Distilled water 5mlkg^{-1}	0.00 ± 0.00^{a}
Compound 100mgkg^{-1}	40.77 ± 1.67^{b}
Compound 200mgkg^{-1}	55.13 ± 0.59^{c}
Compound 400mgkg^{-1}	69.34 ± 0.75^{d}
Artesunate 5mgkg^{-1}	94.33 ± 0.73^{e}

[1]All values expressed as mean ± standard error (±SE).
[2]Different superscript letters indicated significance difference ($p < 0.05$) in mean values among different treatments using Turkey's b *post hoc* comparison.

Table 6: Suppression activity of βeta-Dithiodilactic acid, a compound from ethanolic root extract of *A. boonei* and artesunate in mice infected with *Plasmodium berghei*

Treatments	Suppression (%)
Distilled water 5mlkg^{-1}	0.00 ± 0.00^{a}
Compound 100mgkg^{-1}	46.88 ± 0.58^{b}
Compound 200mgkg^{-1}	57.44 ± 1.16^{c}
Compound 400mgkg^{-1}	60.22 ± 1.73^{c}
Artesunate 5mgkg^{-1}	90.16 ± 0.59^{d}

[1]All values expressed as mean ± standard error (±SE).
[2]Different superscript letters indicated significance difference ($p < 0.05$) in mean values among different treatments using Turkey's b *post hoc* comparison.

DISCUSSION

This study emphasized on the investigation of malaria infection suppression activity by bioactive compounds from extracts of *Alstonia boonei*, a traditional medicinal plant in African countries.The malaria infection suppression test of 1,3,5-Triazine, 2-methylamino-4,6-bis(nonafluoro-tert-butyl), a methoxymethylbutyl class of compound from aqueous leaf extract of *A. boonei* indicated that the minimal suppression of parasitaemia was found in the lowest administered dose of 100mg/kg body weight while the optimal suppressionc effect was in the highest administered dose of 400mg/kg body weight as shown in Table 1. In a study on methoxymethylbutyl class of compound, Wong *et al.,* 2021 evaluated the antimalarial activity of a methoxymethylbutyl class of compound from the crude extract of *Dendrocalamus asper* using chromatographic methods and reported a significant antimalarial activity.

From the malaria infection suppression test of androsta-2,4,16-triene-3,6,17-triol, tri-TMS, a lactone class compound from aqueous stem bark extract of *A. boonei* (Table 2), it showed that the minimal suppression effect was found in the lowest administered dose of 100mg/kg body weight while the optimal suppression effect was in the highest administered dose of 400mg/kg body weight as shown in Table 2. Other similar studies have reported the antimalarial effects of lactones from plants. Chea *et al.,* 2006 in their study on lactone class compounds found out that lactones exhibited significant antimalarial activity as shown in this study also.

The result from the malaria infection suppression test of 1-Oxo-forskolin, a diterpenoid compound from aqueous root extract of *A. boonei* revealed that the lowest suppression effect was in the lowest administered dose of 100mg/kg body weight while the highest suppression effect was in the highest administered dose of 400mg/kg body weight as recorded in Table 3. Saleh *et al.,* 2019, carried out a study on the therapeutic potential of the Labdone Diterpenoid Forskolin class of compound and also recorded a significant antimalarial activity by the compound which agrees to this study.

The malaria infection suppression test of Aldosterone, N-methoxy-tri-TMS, an acridone class compound from ethanolic leaf extract of *A. boonei* showed that the lowest suppression effect was found in the lowest administered dose of 100mg/kg body weight while the highest suppression effect was in the highest administered dose of 400mg/kg body weight as shown in Table 4. The antimalarial potential of acridone compounds have been reported in other studies. Winter *et al.,* 2006 reported significant antimalarial activity of acridone class of compound.

From the malaria infection suppression test of Hecogenin acetate, a diosgenin compound from ethanolic stem bark extract of *A. boonei* (Table 5), it was revealed that the lowest suppression effect was found in the lowest administered dose of 100mg/kg body weight while the highest suppression effect was in the highest administered dose of 400mg/kg body weight. Omonirri *et al.,* 2021 studied the antimalarial activity of a diosgenin class of compound and reported that the compound exerted significant

antimalarial activity on the tested murine model which is akin to the result of this study.

The result from the malaria infection suppression test of βeta-Dithiodilactic acid, a compound from ethanolic root extract of *A. boonei* revealed that the lowest suppression effect was in the lowest administered dose of 100mg/kg body weight while the highest suppression effect was in the highest administered dose of 400mg/kg body weight as recorded in Table 6. This result revealed that the suppression antimalarial effect of this compound is significant.

CONCLUSION

The results from this study revealed that the tested bioactive compounds exhibited significant dose-dependent suppression effect on early *Plasmodium berghei* infection in mice. These bioactive compounds from the ethanolic and aqueous extracts of leaf, stem bark and root of *Alstonia boonei* possess significant antimalarial activity and thus, they should be further investigated for antimalarial drug development. Further recommended studies on the bioactive compounds in this present study are established malaria tests, human malaria parasite (*Plasmodium falciparum*) isolate tests, pharmacological tests, pharmacokinetic tests, determination of their modes of action and appropriate dosage and determination of any interactions with other drug formulations.

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REFERENCES

Adebayo, J. O. and Krettli, A. U. (2011) Potential antimalarial from Nigerian plants: A review. *J. Ethnopharmacol*.; 133: 289–302.

Afolabi, O. J. and Abejide, A. E. (2020) Antiplasmodial activities of *Morinda lucida* (Benth) and *Alstonia boonei* (De wild) in mice infected with *Plasmodium berghei*. *BNRC*; 44: 85–91.

Amole, O. O. and Ilori, O. O. (2010) Antimicrobial activity of the aqueous and ethanolic extracts of the stem bark of *Alstonia boonei*. *Int. J. Pharmacol*.; 1(2): 119–123.

Chea, A., Hout, S., Long, C., Marcourt, L., Faure, R., Azas, N. and Elias, R. (2006) Antimalarial Activity of Sesquiterpene Lactones from *Vernonia cinerea*. Chem. *Pharm. Bull*.; 54: 1437–1439.

Dibua, U. M. E., Okeke, C. C., Ugwu, C., Kenechuckwu, F. C. and Okorie, A. (2013) *In vivo* antimalarial and cytotoxicity activity of ethanolic stem bark of *Petersianthus macrocarpus* and leaf of *Alstonia boonei* in experimental mice model. *Int. J. Curr. Microbiol. Appl. Sci*.; 2: 354–368.

Eswaraiah, G., Peele, K. A., Krupanidhi, S., Indira, M., Kumar, R. B. and Venkateswarulu, T. C. (2019) GC-MS analysis for compounds identification in leaf extract of *Lumnitzera racemosa* and evaluation of its *in vitro* anticancer effect against MCF7 and Hela cell lines. *J. King Saud Univ. Sci*.; 32: 780–783.

Farooqi, S. S., Naveed, S., Quamar, F., Sana, A., Farooqi, S. H., Sabir, N., Mansoor, A. and Sadir, H. (2024) Phytochemical analysis, GC-MS characterization and antioxidant activity of *Hordeun vulgare* seed extract. *Heliyon*; 10: e27297.

Imieje, V. O. and Amafili, L. N. (2025) GC-MS Profiling, *In* Vitro Antimalarial, and Antimicrobial activity of *Ricinodendron heudelotii* Seed Extracts. *Sci. Phytochem*.; 1: 1–8.

Iyiola, O. A., Tijani, A. Y. and Lateef, K. M. (2011) Antimalarial Activity of Ethanolic Stem Bark Extract of *Alstonia boonei* in Mice. *Asian J. Biol. Sci*.; 4: 235–243.

Lestari, E., Setyaningrum, E., Wahyuningsih, S., Rosa, E., Nurcahyani, N. and Kanedi, M. (2023) Antimalarial tests and GC-MS analysis of ethanol and ethyl acetate extract of snake plant (*Sansevieria trifasciata* Prainj. *WJBPHS*; 15(2): 91–97.

Madhiri, R. and Vijayalakshini, G. (2018) A Review of Phytochemical Composition and Pharmacological Aspects of the Genus *Alstonia*. *Pharma Tutor*; 6: 50–55.

Moronkola, D. O. and Kunle, O. E. (2012) Essential Oil Compositions of leaf, stem bark and root of *Alstonia boonei* de Wild (Apocynaceae). *IJBPR*; 3: 51–60.

Muhseen, Z. T., Hameed, A. R., Al - Bhadly, O., Ahmad, S. and Li, G. (2021) Natural products for the treatment of *Plasmodium falciparum* malaria: An integrated computational approach. *Comput. Biol. Med*.; 134: 104415.

Nanven, A. F., Nannim, N., Monday, E. A., Mark, S., Wilson, N. B. and Anthony, D. (2024) Gas Chromatography-Mass Spectrometry Analysis and Antimalarial Activity of *Salix ledermanni* ethanol leaves extracts. *TJNPR*; 8(11): 9121–9130.

Noronha, M., Pawar, V., Prajapati, A. and Subramanian, R. B. (2020) A literature review on traditional herbal medicines for malaria. *S. Afr. J. Bot*.; 128: 292–303.

Olanlokun, J. O., Olowofolahan, A. O., Bodede, O., Adegbuyi, A. T., Princeloo, G., Steenkamp, P. and Olorunsogo, O. O. (2021) Anti-inflammatory potential of the n-hexane fraction of *Alstonia boonei* stem bark in Lipopolysaccharide-induced inflammation in wistar rats. *J. Inflamm. Res*.; 14: 3905–3920.

Omonirri, M. A., Madubuogwu, N. U., Oraekei, D. I.., Ataihire, J. U., Ofili, C. C. and Mbata, U. C. (2021) Antimalaria and hypoglycemic activities of diosgenin on alloxan induced diabetic wistar rats. *GSCBPS*; 15: 73–79.

Saleh, B., Staniak, M., Czopek, K., Stepien, A., Dua, K. and Sharifi-Rad, J. (2019) The therapeutic potential of the Labdane Diterpenoid Forskolin. *Appl. Sci*.; 9: 4089.

Winter, R. W., Kelly, J. X., Smikstein, M. J., Dodean, R. and Bagby, G. C. (2006) Evaluation and lead optimization of antimalarial acridones. *Exp. Parasitol*.; 114: 47–56.

Wong, K., Osman, H., Parumasivam, T., Abdullah, J., Hassan, M. and Azmi, M. (2021) Antimalarial Evaluation of the Chemical Constituents Isolated from *Dondrocalamus asper*. *JOTCSA*; 8: 9951002.