The Journal of Medical Research

Research Article

JMR 2019; 5(4): 134-139
July- August
ISSN: 2395-7565

www.medicinearticle.com
Received: 20-07-2019
Published: 15-08-2019

Evaluation of Arthemether-Lumefantrine Effectiveness in Malaria Treatment in Nnewi, Nigeria

Madubogwu NU[1], MA Omoirri[2], Chukwurah IB[3], Iloh ES[1], Nnekwe PC[4]

1 *Department of Pharmacology and Toxicology, Faculty of Pharmaceutical Sciences, Chukwemeka Odumegwu Ojukwu University, Igbariam, Anambra State*

2 *Department of Pharmacology and Toxicology, Faculty of Pharmaceutical Sciences, Nnamdi Azikiwe University, Akwa, Anambra State, Nigeria*

3 *Department of Pharmacology and Toxicology, Faculty of Medicine, Nnamdi Azikiwe University, Akwa, Anambra State, Nigeria*

4 *Department of Pharmacology and Therapeutics, College of Medicine, Nnamdi Azikiwe University, Awka, Anambra State, Nigeria*

Abstract

As an acute and chronic mosquito-borne disease of man, malaria is characterized by chills and fever, anaemia, splenomegaly and damage to other vital organs such as liver and brain. With reportedly increasing incidence of its lethargy in sub-Sahara Africa, current study was thus designed to investigate the effectiveness of one of malaria's management pharmacological variety, Arthemether-Lumefantrine amongst residents of Nnewi community of Anambra State, Nigeria. Hundred (100) human subjects from the General Outpatients Department (GOPD) of the Nnamdi Azikiwe University Teaching Hospital, Nnewi, who showed signs and symptoms of malaria, were recruited for the study. After gaining subjects' consent and co-operation, Artemether-Lumefantrine combination (combination therapy) was then orally administered to the patients; with Blood samples collected 10 min before, and on days 4, 8, 10 and 14 after drug administration. Efficacy evaluation of parasitological cure rates was also determined after the 14th day. In addition to cure rate, fever clearance time (FCT), as the time from drug administration till axillary temperature fell below 37.5^{o}C and remained so for at least 48 hours was also determined. In any case, obtained data were analysed using appropriate descriptive statistical (mean, standard deviation, frequency and percentage). The Chi square distribution test was performed to ascertain the goodness of fit of obtained variables. p-value was determined at 95% confidence interval, with significance level set at $p < 0.05$. Upon analysis, Study found after treatment at day 4, that cure rate for patients <16 years (paediatrics) was 52%, with those >16 years (adults) being 72%. On day 8 after treatment, cure rates for patients <16 years (paediatrics) was 89%, while that of those >16 years (adults) was 94%, while on day 10 and 14 the cure rate for patients <16 years became 98% while that of those >16 years was 100%.

Keywords: Maleria, Anti-MaleriaCure Rate, Effectiveness.

INTRODUCTION

As an acute and chronic mosquito-borne disease that affects man, malaria is commonly characterized by chills and fever, anaemia, splenomegaly and damage to several vital organs like the liver and brain. Though it is historically an ancient disease, it currently ranks as Africa's most lethargic and leading cause of health problems, including death [1-2]. Malaria is the most important parasitic disease in the tropics and remains a high public health menace of utmost importance.

Available records posit that approximately 90% of global known cases of malaria deaths occur in sub-Sahara Africa, with an estimated one million people reportedly dying yearly and most of which are children under five years old [3]. In Nigeria, malaria transmission is holoendemic, with over 90% of her populace reportedly lives in areas with stable malaria. This flags it as a major cause of morbidity and mortality in most parts of developing countries [4]; maintaining it as one of the major infectious diseases in the world with about 2.8 billion (60%) of the world's population apparently living with it in endemic areas.

Recently, the United Nation's Children Fund (UNICEF) has noted that the greatest challenge in malaria control is that the cheapest anti malaria drug, chloroquine is rapidly losing its efficacy in many endemic regions of the world. With this, efforts of the World Health Organization (WHO) through its numerous eradication programmes has remained a vexatious health problem in the tropics as attested by the attendant high morbidity and usually high mortality, especially among children [5].

***Corresponding author:**
Madubogwu NU
Department of Pharmacology and Toxicology, Faculty of Pharmaceutical Sciences, Chukwemeka Odumegwu Ojukwu University, Igbariam, Anambra State
Email: osgiedeprof@yahoo.com

This has increased global malaria burden and is a major threat to malaria control. Resistance can be prevented, or its onset slowed considerably, by combining anti-malarials with different mechanisms of action and ensures very high cure rates through full adherence to correct dose regimens [6].

Combination therapy with antimalarial drugs is the simultaneous use of two or more blood schizonticidal drugs with independent modes of action and different biochemical targets in the parasite [7]. Combination therapies can be either fixed-combination of medicinal products, in which the components are co-formulated in the same tablet or capsule, or multiple drug therapy, in which the components are co-administered in separate tablets or capsules. This concept has been realized in multiple-drug therapy for leprosy, tuberculosis and cancer and more recently, in antiretroviral treatments. The best available treatment is a combination of drugs known as Artemisinin-based combination therapies (ACTS).

Recently, the World Health Organization (WHO) announced that the emergence of artemisinin resistant parasites at the Thai-Cambodia border could seriously undermine global malaria control efforts [2-3]. With no known effective alternatives to artemisinin for the treatment of malaria, drug resistance to commonly used anti-malarias has spread very rapidly. In order to avoid this for artemisinin, they are often used in combination as ACTS, and artemisinin monotherapy (use of one artemisinin drug versus the more effective combination pill) [8]. The less effective single-drug treatment increases the chance of the parasite to evolve and become resistant to the medicine [3]. This combination has proved as effective and better tolerated as artesunate plus mefloquine in the treatment of multi-drug resistant *plasmodium falciparum* when given as a six dose regimens over three days [9].

Objectively, treatment of uncomplicated malaria is geared toward curative solutions to the infection, but in severe malaria, the main objective is to prevent the patient from dying. Secondary objectives are prevention of recrudescence, transmission or emergence of resistance and prevention of disabilities [10-11].

Artemether-lumefantrine is the most viable artemisinin combination treatment available at the moment and so the study of the effectiveness of this combination would be quite timely. In view of this, current study was designed to evaluate the effectiveness and safety of Arthemether-lumefantrine in the treatment of malaria in Nnewi, Anambra state, Nigeria.

Aim of Study

Current study was designed to evaluate the effectiveness and safety of Arthemether-Lumefantrine in the treatment of malaria in Nnewi, Anambra state. Specifically, study determined the cure rate of Arthemether-lumefantrine therapy after day 14 of treatment. Study also ascertained the effect of Arthemether-lumefantrine on fever clearance time, plus investigated whether the drug is more effective in treating paediatrics than adults.

MATERIALS AND METHOD

Humans

One hundred subjects from the General Outpatients and Paediatrics Departments (GOPD) of the Nnamdi Azikiwe teaching Hospital, Nnewi and who showed signs and symptoms of malaria were ethically recruited for the study.

Drugs

Arthemether-lumefantrine (Lomal™) was purchased from Emzor Pharmaceuticals LTD, Lagos, Nigeria.

Ethical Clearance

Ethical Approval was obtained from the Ethical Committee of the Nnamdi Azikiwe University Teaching Hospital before the commencement of the study. Also, the consent of Physicians at GOPD and Paediatrics unit was obtained, with two of them participating in this study.

Informed Consent

Written and oral consent was obtained from participants and the caregiver of the paediatrics patients.

Sampling Technique

Non-probabilistic sampling technique of the purposive method was used. By this, subjects with clinical diagnosis of malaria and fever seen at the general Out-patient Department (GOPD) and Paediatrics Department of Nnamdi Azikiwe University Teaching Hospital were selected. This was done after due consultation with them, and their written consent gotten. Those that refused to give their consent were excluded.

Sample Size Determination

Sample size for the study was expected to be about ninety patients. This was arrived at using the 30% prevalence for malaria disease in the area of study. According to availed record, an estimated population size of 124 new malaria cases are reported (average monthly record of malaria from GOPD). The sample size was then calculated using;

$$nf = \frac{n}{1 + n/N}$$ [12].

Where, nf = the desired sample size when the population is less than 10,000. n = the desired sample size when the population is more than 10,000, N = the estimate of the population size

n can be calculated using the formula;

$$n = Z^2Pq/d^2$$

Where Z = the standard normal deviation usually set at 1.96 (which corresponds to 95% confidence level). P = prevalence q = 1 – p d = degree of accuracy desired usually set at 0.5

Therefore $n = \frac{(1.96)^2 \times 0.3 \times 0.7}{(0.05)^2}$

$$n = \frac{3.8416 \times 0.3 \times 0.7}{0.0025}$$

$$n = \frac{0.806736}{0.0025} = 323$$

Then $nf = \frac{n}{1 + n/N}$

Where n = 323

N = 124

Therefore $nf = \frac{323}{1 + 2.6} = \frac{323}{3.6} = 90$

nf = 90 (approximate)

Inclusion Criteria

(I) Subjects who presented with symptoms of malaria and have been diagnosed as such.
(II) Malaria sufferers who can take in oral medication.
(III) Subjects with axillary temperature > 37.5^{0}C.
(IV) Patients who gave informed consent and for paediatrics the consent of the caregivers was gotten.

Exclusion Criteria

(i) Patients treated with anti-malaria in the previous 24 hours.
(ii) Patients with symptoms and signs of severe malaria (i.e. patients with more than 5% of their red blood cells parasitized).
(iii) Patients with known serious underlying disease (example patients who are known hypertensives, diabetics, or those suffering from chronic heart failure or other conditions suspected to affect the result).
(iv) Allergy to any component of the drug combination.
(v) Patients who are pregnant and breast feeding.
(vi) Those who did not give informed consent.

Drug Administration and Collection

The drug, Artemether-Lumefantrine combination (combination therapy) was administered to subjects through oral route. Blood samples were collected 10 min before and on days 4, 8, 10 and 14 after drug administration. With gloved hands and application of tornique proximal to the site of sample collection, spirit swab was used to scrub the area of sample collection. Blood samples were collected with 2 ml syringe and then placed into Ethylene Diamine Tetracetic Acid (EDTA) bottle. The tornique was subsequently removed and pressure applied over the site of collection with sterile gauze. The sample was then carried to the haematology laboratory for staining and microscopic examination.

Identification of Parasites

Malaria parasites

Blood films for malaria parasites were prepared from anti-coagulated venous blood. Thin and Thick films were prepared from it to identify the presence of Plasmodium parasite while the actual counting of parasitemia and examination of the blood film was done in the haematology department of the hospital, examined by a single microscopist. Thin and Thick blood films were Giemsa stained within 30 min-1h of preparation.

Parasite count was obtained using thick blood films, counted as the number of parasite per 200 white blood cells (WBC) while thin film was used to determine the parasite specie.

Making of Thin and Thick Blood Film

Wash dry, grease-free, water free, and starch free slides were used for making smear and placed in template positions.

Thin smears were made using a smooth edged slide spreader on the drop of blood. Holding the slide and the "spreader "at suitable angle, the spreader was pushed along the slide, drawing the blood behind it, making sure the whole of the drop was smeared.

Thick film was prepared by dropping blood the slide with micro pipettes to fill large circle and the rear end of the pipette was used to spread the large drop to make a thick smear. The slides were labelled with pencils by writing the name, serial number of patients at the edge of the slide and dates written and the films were air dried with the slides in horizontal position and safe guarded against any contact with any object whatsoever until they were completely dry.

Giemsa Staining Technique

Romanoswsky stains using giemsa staining technique were employed. It is an alcohol –based stain that requires dilution in pH 7.1-7.2 buffered water before use. This technique allowed the opportunity of staining many films at a time. The films were allowed to dry overnight to get the best result and giemsa stain was usually diluted just before use by adding 1.5ml of giemsa to 50ml of buffered water (PH7.1) and gently mixed.

Dried films were placed facing downwards in a staining rack for immersion in a staining trough. Giemsa stain was run on the film with clean, water free Pasteur, pipette. Stain was then allowed to run on the film for 30 min after which the films were rapidly rinsed with buffered saline. Back of the slide was wiped clean, drained and stood to air dry.

Viewing Dry Film under Microscope

Magnifications of 40x and 100x objective lens were used for viewing after immersion oil was applied on the dried films. Parasite count was obtained using thick blood films, counted as the number of the parasites per 200 white blood cells (Warhurt and Willian, 1996). Parasitemia was calculated by the formula:

$$\text{No. of parasite per } \mu l = \frac{\text{No of parasite x 8000}}{\text{No of White Blood Cell Count (WBC)}}$$

(8000 leucocytes per µl is the world health recommended standard).

The parasite count in this study is grouped as follows:

1. Parasite count of <40/µl is considered negative (-).

2. Parasite count of 40-399/µl is considered one plus (+).

3. Parasite count of 400-3999/µl is considered two pluses (++).

4. Parasite count of 4000-39999/µl is considered three pluses (+++).

The specie of parasite seen was *P. falciparium*.

Efficacy Assessment

Efficacy evaluation was based on parasitological cure rates at days 4,8,10 and 14.Cure rates at days 4, 8, 10, and 14 were defined as the proportion of patients cleared of asexual parasitaemia within the specific time intervals of initiation of treatment with artemether-lumefantrine. In addition to cure rate, fever clearance time (FCT) was determined. FCT is defined as the time from drug administration until axillary temperature fell below 37.5^{o}C and remained so for at least 48 h.

Safety Evaluation

Safety assessment consisted of monitoring and recording of all the adverse events whether volunteered, discovered by questioning or detected by examination in addition to clinical significant changes in haematology. An adverse event is regarded as any undesirable sign, symptom or medical condition occurring after initiation of treatment with the study drug, whether the event is considered to be related to study drug or not. Any sign and symptom that appeared newly or worsened were recorded as adverse events. In the present study, no adverse or side effect to Arthemether-Lumefantrine was noted in any of the patient in the study population.

Analytical Approach

Obtained data were analysed using descriptive statistics (mean, standard deviation, frequency and percentage, as appropriate) by body

weight group and overall. The statistical method used was Chi square distribution using SPSS version 17. P value was determined at 95% confidence interval and values less than 0.05 were taken as statistically significant.

RESULTS

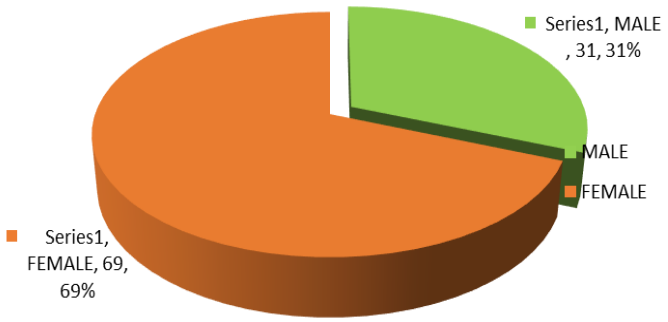

Figure I: Percentage distribution of Participants by Gender

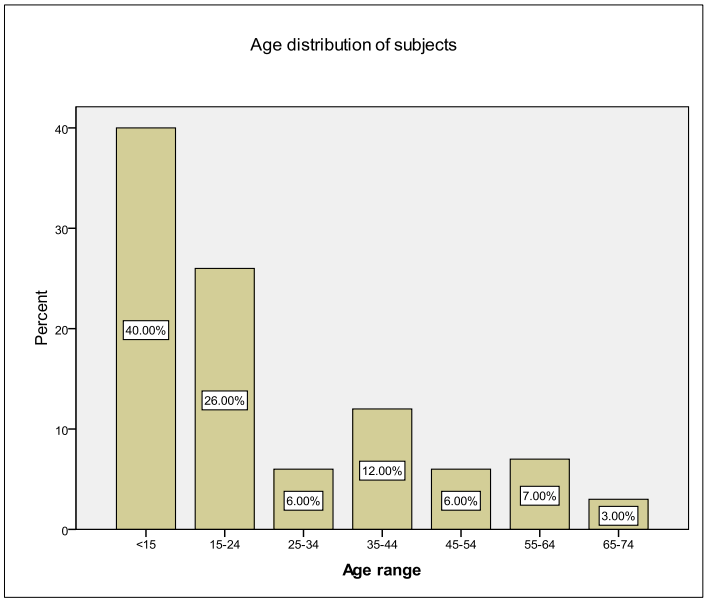

Figure II: Percentage distribution of Participants by Age

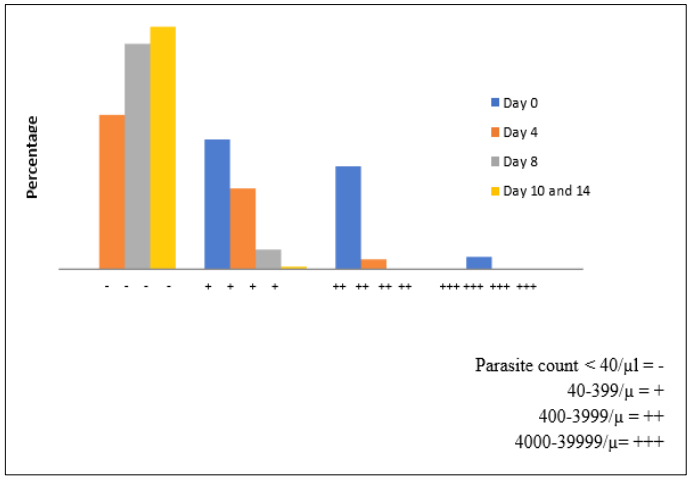

FIGURE III: Summary of Parasite Counts on Days 0, 4, 8, 10 and 14

Table 1: Effectiveness of Arthemeter-Lumefantrine on day 4 in adults and children

	Parasite count Day 4		Total
Age group	_	+	
<16 yrs	24(52%)	22(48%)	46
>16 yrs	39(72%)	15(28%)	54
Total	63	37	100

Table 2: Effectiveness of Arthemeter-Lumefantrine on day 8 in adults and children

	Parasite count Day 8		Total
Age group	_	+	
<16 yrs	41(89%)	5(11%)	46
>16 yrs	51(94%)	3(6%)	54
Total	92	8	100

Table 3: Effectiveness of Arthemeter-lumefantrine on day 10 and 14 in adults and children

	Parasite count Day 10 &14		Total
Age group	_	+	
<16 yrs	45(98%)	1(2%)	46
>16 yrs	54(100%)	0(0%)	54
Total	99	1	100

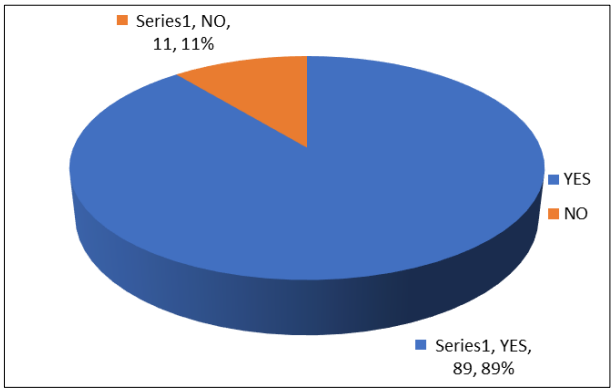

Figure IV: Pie chart showing the percentage of patients with fever on day O

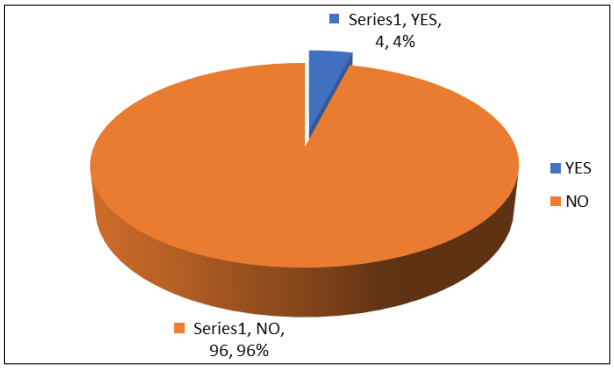

Figure V: Pie chart showing the effect of the drug on fever after day 3

DISCUSSION

Malaria, disease caused by infection with single-celled parasites of the genus *Plasmodium*. *Anopheles* mosquitoes transmit these parasites from one person to another in their bites. It is characterized by periodic bouts of severe chills and high fever. Serious cases of malaria can result in death if left untreated. More than a million people die of the disease each year, most of them in Africa, according to the World Health Organization (WHO) [2-3].

Malaria was once widespread in North America and other temperate regions. Today, the disease occurs mostly in tropical and subtropical regions, particularly in sub-Saharan Africa and Southeast Asia [13]. The disease is also found in Central and South America, Oceania, and on some Caribbean islands. Public health officials had hoped to wipe out malaria during the 20th century. However, malaria parasites have developed defenses against many antimalarial drugs. This response, known as drug resistance, makes the drugs less effective. In addition, the *Anopheles* mosquitoes that transmit the disease have become resistant to many insecticides. Malaria has continuously been a main public health problem in the world, especially in the majority of African countries [14].

Even though Chloroquine has been the drug of choice for its treatment, the introduction of new anti-malaria agents has become necessary due to chloroquine resistance by *P. falciparum,* which is now common in almost all malaria ravaged parts of the world. Interestingly however, Artemisinin-based combination therapy (ACT) is advocated as the way forward in malaria treatment to overcome the global spread of *P. falciparum* drug resistant [1]. Artemether-lumefantrine has been shown to be well tolerated in humans with very little significant toxic effects [15]. Howbeit, results from this investigation showed that 63% of the patients treated with artemether –lumefantrine were free of parasitaemia after four days of treatment.

Krimsner *et al*. (2004) had report that 80% of the population treated with Artemether-Lumefantrine were free of patent parasitaemia 4 days after initiation of therapy. Also on day 8 after treatment, current study observed 92% of the patient to be free of the patent parasitaemia, while on day 10 and 14 after treatment 99% of the patients were free of patent parasitaemia. Though this does not mean that the patients were parasitologically cured because of recrudescence; however, this is suggestive that only one patient out of one hundred patients treated with artemether-lumefantrine still had malaria parasite after treatment. Therefore the overall cure rate after 14 days of treatment was 99%. The reason for the effectiveness of artemether-lumefantrine therapy could be explained in terms of its mechanism of action, which is known to rapidly diminish parasite biomass, leading to clinical and parasitological cure while at the same time gametocytocidal activity might be able to reduce overall malaria transmission [16]. These results are consistent with those reported by Abdul-Aguye *et al.* 2000 [17] in a pre-registration study of the four dose-regimen in Zaria, Northern Nigeria which recorded 100% cure rate at day 14 among 50 patients aged 2 to 65 years suffering from acute uncomplicated malaria.

On the other hand, the efficacy results recorded in this study is superior to those obtained by Salako *et al*. (2000) [18] in which they reported cure rates of 87% and 73% on day 7 and 14 respectively among children aged 2 to12 years, while it is consistent with the works done by Ezedinachi *et al*. (2000) in which they reported 96% and 93%cure rates on days 7 and 14 from Calabar Nigeria [19]. Also working in Nigeria, Eke *et al*. 2000 [20] recorded a day 7 cure rate of 88% among 57 patients aged 2 and 16 years in Port Harcourt.

The superior efficacy of the six dose regimen used in this study when compared with that of the four –dose regimen is consistent with findings in similar studies that evaluated the four dose and six dose regimens in the western border of Thailand, in Tanzania, Kenya and the Gambia [21]. Meremikwu *et al.*(2006) [22] working in Calaber Nigeria recorded a day 14 cure rate of 87% among 54 children they studied, Robbin-Kobbe *et al*.(2008)reported a cure rate of 88.3% after treatment with artemether-lumefantrine whereas in this study a 99% cure rate was recorded [23]. The 87% cure rate recorded by Meremikwu *et al*. might be due to smaller population of 54 patients compared to 100 patients in this study. Catherine *et al*. 2008 in Ibadan [24], Nigeria reported a cure rate of 100% after day 14 treatment with artemether-lumefantrine and this was consistent with this study.

Again from this study, 89% of the patients manifested with fever (temp>37.5^{o}c) before initiation of treatment, while only 4% of the patient had fever after the third day of treatment (temp < 37.30^{o}C). No patient had fever after the day eight of treatment. This corresponds to the work done by Catherine *et al*. 2008 in which they recorded mean fever clearance time of 24.9h.

CONCLUSION

Results of this study suggest that if properly deployed, Arthemeter-Lumefantrine could lead to a reduction and may also contribute to a significant extent in halting the worsening morbidity and mortality from malaria on the African continent [25].

Recommendations

The Federal government of Nigeria recently changed its malaria treatment policy from chloroquine to artemisinin-based combination therapy as a first line drug with a preference for AL and artesunate plus amodiaquine in that order. It is important to design appropriate information, education and communication materials, which will educate patients, parents, guardians and health care workers alike on correct treatment regimen and the need to administer AL with food and that will encourage compliance as part of the efforts at prolonging the useful therapeutic life of this combination.

References

1. White NJ. Malaria. Mansons Tropical Disease (Edited by: Cook G.C, Zumala A., and Weir J). Philadelphia P.A, W.B Suanders: 1205-1295, 2003.
2. World Health Organization. stat – Q1992: World malaria situations division of control tropical diseases, 1990.
3. World Health Organization. World malaria situation in 1994, population at risk. Weekly epidemol Rec, 1994, 1997; 72(36):269-274.
4. Ramakrishna JW, Brieger WR, Adeniyi JO. Treatment of malaria and febrile convulsion. An educational diagnosis of Yoruba Beliefs: Int. Quatr. Common. Health Educ. 2009; 9:305-319.
5. Okonkwo PO, Okpala CO, Okafor HU, Mbah AU, Nwaiwu O. Compliance to correct dose of chloroquine in uncomplicated malaria correlates with improvement in the condition of rural Nigerian Children Trans. R-Soc. Trop. Med. Hyg. 2001; 95(3):320-324.
6. O' Neill PM, Posner GH. A medicinal chemistry perspective on artemisinin and related endoperoxides J. med. Chem. 2004; 47:2945-2964.
7. Price RN, Van Vugt M, Phaipun L. Adverse effects in patient with acute falciparium malaria treated with artemisinin derivatives, Am. j. Trop. med Hyg. 2009; (1999)60:547-555.
8. Novartis Pharma. New hope of winning the war against malaria with new fixed combination of artemether plus lumefantrine. Trop. Med. Int. Health 2004; 2:192-199.
9. Tamariya P. *In vitro* senisitvity of P. falciparum and clinical response to lumefantrine and artemether. British Journal of Clincial Pharmacology. 2000; 49:437-444.
10. Ogutu. The efficacy of pyrimathamine-sulfadoxine resistance of plasmodium falciparum malaria in Kenyan Children Trans. Soc. Trop. Med. Hyg. 2000; 94:83-84.
11. Okonkwo PO, Okpala CO, Okafor HU, Mbah AU, Nwaiwu O. Compliance to correct dose of chloroquine in uncomplicated malaria correlates with improvement in the condition of rural Nigerian Children Trans. R-Soc. Trop. Med. Hyg. 2001; 95(3):320-324.
12. Araoye MO. Resrearch Methodology with statistics for health and social sciences.First edition, Nathadox Publishers, Ilorin, Nigeria, 2004; 120.

13. Bosman A. Review Application for inclusion of a drug in the WHO Essential Drug List. Trop. Med. Int. Health. 2002; 2:192-198.
14. Price RN, Van Vugt M, Phaipun L. Adverse effects in patient with acute falciparium malaria treated with artemisinin derivatives, Am. j. Trop. med Hyg. 2009; (1999)60:547-555.
15. Breman JG. The ears of the hippopotamus: manifestations, determinants and estimates of the malaria burden. AM J. trop. Med. Hyg, 2005; 64(1-2 suppl):1-11.
16. Krimsner P, Krishna S. Antimalaria Combinations.Lancet, 2004; 364:285-294.
17. Abdu-Aguye I, Gebi UI, Wuyat A, Agbo M. An open-label uncontrolled trial confirming efficacy and safety of the 4-dose regimen of Coartem® in the treatment of acute Plasmodium falciparum malaria in patients aged > 2 years in Zaria Nigeria. Report of a Clinical Trial, Novartis Pharma AG Nigeria study Report, 2000.
18. Salako LA, Adewole TA, Afolabi BM, Mafe AG. An open-label uncontrolled trial confirming efficacy and safety of the 4-dose regimen of Coartem® in the treatment of acute Plasmodium falciparum malaria in patients aged > 2 years in Lagos Nigeria. Report of a Clinical Trial. Novartis Pharma AG Nigeria study Report, 2000.
19. Ezedinachi EN, Ekanem-Ephraim E, Ndifon N, Amaechi V, Alaribe AA, Bam AB. An open-label uncontrolled trial confirming efficacy and safety of the 4-dose regimen of Coartem® in the treatment of acute Plasmodium falciparum malaria in patients aged > 2 years in Calabar, 2000.
20. Eke F, Akhidue V, Ukiwo UE, Akosubo IJ, Izuora P, Akpan PU, *et al*. Report of an open-label uncontrolled trial confirming efficacy and safety of a 4-dose regimen of Coartem® [artemether-lumefantrine (benflumetol)] in the treatment of acute Plasmodium falciparum malaria in children in Port Harcourt. Novartis Pharma AG Nigeria study Report; 2000.
21. Falade C, Makanga M, Premji Z, Ortmann C-E, Stockmeyer M, Ibarra de Palacios P. Efficacy and safety of six dose artemether-lumefantrine (Coartem®) tablets (six-dose regimen) in African infants and children treatment of acute uncomplicated malaria. Trans R Soc Trop Med Hyg. 2004; 99:459-467.
22. Meremikwu M, Alaribe A, Ejemot R, Oyo-Ita A, Ekenjoku J, Nwachuckwu C, *et al*. Artemether-lumefantrine versus artesunate plus amodiaquine for treating uncomplicated malaria in Nigeria: randomized controlled trial. Malar J. 2006; 5:43.
23. Robin Kobbe, Philip Klein, Samuel Adjei, Solomon Amemasor, William Nana Thompson, Hanna Heidemann, *et al*. A randomized trial on effectiveness of artemether- lumefantrine versus artesunate plus amodiaquine for unsupervised treatment of uncomplicated P.falciparium malaria in Ghanain children. Malaria Journal. 2008; 7:261.
24. Catherine O Falade, Oluwatoyin O Agunkule, Hannah O Dada-Adegbola, Adegoke G Falade, Patricia Ibara de Palacios, Philip Hunt, *et al*. Evaluation of the efficacy and safety of arthemether-lumefantrine in treatment of acute uncomplicated Plasmodium falciparium malaria in Nigeria infants and children. Malaria journal. 2008; 7:246.
25. Brewer TG, Grate SJ, Peggins JO, Werna PJ, Petras JM. Fatal neurotoxicity of arteether and artemether. AM. J. Trop. Med. Hyg. 2014; 51:251-259.