EFFICACY, SAFETY AND TOLERABILITY OF ARTESUNATE – MEFLOQUINE (ARTEQUIN™) IN THE TREATMENT OF UNCOMPLICATED FALCIPARUM MALARIA IN 4 GEOGRAPHIC ZONES IN NIGERIA

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ABSTRACT

BACKGROUND: Artequin™, a combination of Artesunate and Mefloquine has been reported to be effective against multi-drug resistant *Plasmodium falciparum* malaria in other countries but not in Nigeria where we now report the results of multi-centre studies in four high malaria transmission areas. The objective was to evaluate the efficacy, safety and tolerability of the co-packaged drug in the treatment of malaria in children and adults weighing 15 -< 30kg and ≥30kg respectively.
Methods: Rural communities in the North East (Maiduguri), North Central (Jos), South West (Lagos) and South-South (Calabar) were used for the trial. The WHO protocol was followed. Outpatients having amongst other criteria, a pre-treatment parasite density of ≥ 1000µl of blood were enrolled. Ethical approval and Informed Consent were obtained by each site and the drugs were given on days 0, 1 and 2. Each patient was followed up to day 28 with the assessment of the parasitological, biochemical and haematological parameters on days 0, 1, 2, 3, 7, 14 and 28. Data generated from the four sites were entered into EPI- INFO Version 6.04, Microsoft Excel was used to plot simple graphs and various aspects of the data were subsequently analysed using SPSS statistical Package Version 11.

Results: of the 4,139 patients screened in the four sites, **446** met the enrollment criteria but **431** (**203** Adults and **228** children) completed the trial. The rates in adults on D1, D2, D3, D7, D14 and D28 were **40.6%**, **92.1%**, **99.5% 100%**, **100%**, **99.5%**, respectively. The rates in children were **31.3%**, **91.6%**, **100%**, **100%**, **100%**, and **97.8%** respectively. The mean parasite clearance times in adults and children were **40.1** h and **42.4** hrs respectively while the mean fever clearance times were **11.25** h and **13.25** h respectively. Artequin™ exhibited marked antigametocyte activity, with mean gametocyte clearance time of **42.0** h in adults and **45.6** h in children. There were no serious adverse events in all centres. The most common adverse events were headache, dizziness, vomiting and abdominal discomfort. There was also no significant derangement in the haematological and biochemical parameters in the treated patients.

Conclusion: We conclude that co-packaged artesunate + amodiquine (Artequin™) is highly efficacious, safe and well tolerated. Its use in the treatment of malaria is therefore recommended.

Key words: Artequin™, *Plasmodium falciparum,* malaria, ACTs.

BACKGROUND

Malaria remains one of the greatest causes of morbidity and mortality in the world. Global estimates show that there are about 300-500 million cases of clinical malaria every year, with 85% of these from Africa **[1]**. Currently, 1.5 to 2.7 million deaths are attributable to malaria annually, 90% of them in Africa **[1]**. In Nigeria, malaria is holoendemic and the risk of getting malaria is present all the time. It is the most common cause of outpatient hospital attendance in all age-groups and in all parts of Nigeria **[2]**. Children 6 months to five years and to a lesser extent 5-11 years are prone to severe illness which, if untreated, often leads to death. Many adults in these high transmission, rural areas are not spared.

The Drug Therapeutic Efficacy Test (DTET) conducted on chloroquine (CQ) and sulphadoxine pyrimethamine (SP) in Nigeria (2002) showed a national average of adequate clinical and parasitological response (ACPR) of 39.2% (range 3.7% to 77.3%) and 56.7% (range 8.5% to 94.2%) respectively **[3]**. This was too poor. The global malaria control strategy advocates prompt and adequate treatment as an essential measure to reduce the morbidity and mortality arising from the disease **[3]**. However, there has been a great concern on the increasing reports of resistance to the first and second-line drugs, namely, CQ and SP respectively [**4, 5**] . To overcome this problem, both the World Health Organization (WHO) and the Nigerian Federal Ministry of Health advocated a change to artemisinin - based combination drugs which have been found to be effective in other countries. The principle advanced is that the probability of resistance developing simultaneously to two drugs with independent mechanisms of action is extremely low **[5,6]**.

The Artemisinins are new drugs developed from the Chinese Warmwood (*Artemisia annua*) and the derivatives, namely, artemether, artesunate and dihydroartemisinin have now gained popularity as short acting drugs which could be used in combination with drugs which have long half-life **[7,8]** . The National Drug therapeutic Efficacy tests (DTET) conducted on two such combinations,namely, artesunate + amodiaquine and

artemether + lumefantrine in 2004 showed a national average of adequate clinical and parsitological response (ACPR) of 94.6% and 96.8% respectively **[3]**. The National Policy on Malaria Treatment then recommended the use of artemisinin–based combination therapy (ACT) since artemisinins have been found to be the most potent antimalarial drugs and they have an excellent safety profile **[3]**.

However, the problem of availability and affordability still existed. To improve better access to ACTs at affordable prices, Roll Back Malaria partners in the pharmaceutical industries were encouraged to prepackage ACTs which should be added to the market if found effective, approved and duly registered by the National Agency for Food and Drug Administration and Control (NAFDAC). One such combination drug is Artequin™, a combination of artesunate and mefloquine, manufactured by MEPHA Ltd (Aesch- Basel, Switzerland).

The rationale for adding mefloquine to artesunate is that due to the short half life of artesunate, a certain fraction of parasite may survive which is then exposed to long-term therapeutic concentration of mefloquine until complete extinction.

Mefloquine has been reported to consistently show high treatment efficacy in African Children **[9,10]** and in pregnant women [**11]** .This was in the era of antimalarial monotherapy. At that time, mefloquine, a 4-quinoline carbinol, was reported to be one the most effective drugs in the treatment of malaria in Nigeria **[12].** It was also found to be an effective suppressive prophylactic drug when administered weekly or fortnightly against drug-resistant *P. falciparum* **[13]**. The successful treatment of falciparum malaria with regimens of artemisinin derivatives plus mefloquine has been reported in other countries **[14,15,16,17]** The pharmacokinetics of mefloquine combined with artesunate in children with acute falciparum malaria in Thailand has also been studied **[18]**. Li *et al* **[19]** showed that artesunate has a broader stage – specificity of action than other antimalarial drugs. After oral artesunate, relative bioavailability of the drug was 82.0%. The parasite clearance time (PCT) and fever clearance time (FCT) were 6.5hr and 24hr respectively **[20]** and parasitaemia was reduced by 90% within 24hr after starting treatment.

Having reviewed the work done by workers in other countries, and knowing that results of therapeutic efficacy tests (DTET) could depend on various parameters in any particular country and/or geographical zone of one country, we deemed it pertinent to undertake the trial in 4 rural, high transmission communities of Nigeria. The objectives of the study were:

- To evaluate therapeutic efficacy of a combination of Artesunate + mefloquine (Artequin™) in adults and children using the modified WHO 7-day *in vivo* test extended to 14 and 28 day follow-up period.
- To determine the safety and tolerability of Artequin™ (Artesunate + Mefloquine) in the treatment of acute uncomplicated malaria.
- To estimate gametocyte carriage and its reduction during treatment.

The rationale was based on the convincing evidence that a combination of two or more schizontocidal drugs will not only improve cure rate but could help reduce the rate of development of parasite resistance to either of the drugs in the combination. Furthermore, a combination of short-acting artemisinin derivative (artesunate) and long acting mefloquine are said to present a good ACT combination. Since no two populations are exactly the same, it is important to determine the efficacy, safety and tolerability of this ACT among Nigerian population. There is also the need to provide more ACTs as armamentarium for malaria control in Nigeria.

METHODS:

STUDY SITES: The investigations were conducted in four geographical zones of Nigeria and each research team was led by a Principal Investigator (PI). A Primary Health Centre and a private Health Clinic owned by the National Power Holding Company of Nigeria (PHCN) all located in Ijede constituted one of the sites. Ijede is a rural community in Ikorodu Local Government Area (LGA) Lagos State, Southwest Nigeria..The second site was in Borno state, North Eastern Nigeria. The state Specialist Hospital, the General Hospital, Damboa and the Maiduguri Teaching Hospital all in Maiduguri, Borno State were the Health centres used. People visited the Health Centres

from rural areas. The third site was located in a Primary Health Centre in Ikot Ansa, Cross River State, South Eastern Nigeria. The fourth site, located in Jos, Plateau State, North Central Nigeria operated in three Health Centres, namely, ECWA Evangel Hospital, Jos University Teaching Hospital and Plateau State Specialist Hospital. All sites were considered to be homogenous and high malaria transmission areas, hence their suitability for trials of this nature.

Inclusion criteria:

Age stratified into 6 months – 15 yrs (15-30kg) and over 15 years (>30kg) , absence of severe malnutrition by clinical examination and weight for height measurement, mono-infection with *P. falciparum* parasitaemia with a parasitaemia in the range of 1000 to 250,000 asexual parasites per µl of blood, presence of axillary temperature $\geq$ 37.5^{o}C and/or history of fever in the preceding 24 hours, informed consent by parent / guardian (in the case of children),ability to come for the stipulated follow-up visits, and, easy access to the health facility.

Exclusion criteria:

Presence of general danger signs such as: not able to drink or breastfeed, vomiting everything, recent history of convulsion, lethargic or unconscious state, unable to sit or stand up and use of any drug known to influence cardiac function within 4 weeks before screening. Also excluded were those showing signs of severe and complicated falciparum malaria**,** namely**,** cerebral malaria (unarousable coma), severe anaemia (PCV <15% at day 0), renal failure (serum creatinine >3mg/dl), pulmonary oedema, hypoglycaemia (<40mg/dl), shock (systolic BP<70 in adults, 50 in Children), spontaneous bleeding, macroscopic haemoglobinuria, jaundice (serum bilirubin>3mg/dl), febrile conditions caused by diseases other than malaria, history of allergy to study drugs and pregnant women.

STUDY DESIGN

No existing medication was approved for a direct comparison, so, a descriptive, open label, non-comparative trial of three-day regimen of a combination of Artequin™ (artesunate + mefloquine) for efficacy, safety and tolerability was carried out within the four sites in Nigeria. Patients who met the criteria as earlier stated were asked to freely volunteer for the study. They or their guardians also gave written consent after explanatory notes. Day 0 was the day of screening, clinical assessment, initial malaria smears, and taking of blood for Packed Cell Volume (PCV). The participants in each arm (adults and children) also had their blood taken for studies on the laboratory indices of safety, namely, routine biochemistry (Liver Function Tests such as aspartate amino-transferase (AST),alaninie amino-transferase (ALT), total and conjugated bilirubin and renal function - urea). Haematological parameters such as full blood counts (FBC), WBC, Platelets, ESR, PCV and reticulocytes were also investigated. The patients were allocated to one or two bodyweight groups (15 to < 30kg and >30kg). They were called children and adults for convenience and given the first dose of Artequin™ (300/375 or 600/750 Lactab respectively) on day 0.

The drugs were administered under medical supervision and treated patients were observed for 60 minutes. If vomiting occurred within 30 minutes of administration of the drug, the same dose was repeated. However, if it occurred 30-60 minutes, half the dosage was given again. Participants who vomited a second time were excluded from the study and referred for treatment with appropriate parenteral antimalarial regimen. Any use of concomitant medications (including acetaminophen i.e. paracetamol) were documented in the Case Report Form (CRF).

After drug administration on day 0, the patients were asked to return on days 1 and 2 to complete the drug regimen and for clinical assessment. They were also given appointment papers for days 3, 7, 14 and 28 for clinical examination and blood smears. They were also asked to return to the clinic on days other than these if they developed any additional complaints, or any change in their condition compared to pre- administration of the drug. If a patient did not report at the Health Centre for

the scheduled visit, every effort was made by the field workers to locate his/her home address.

Discontinuation of treatment

Serious adverse events, loss of patient to follow-up, consent withdrawal or withdrawal as a result of treatment failure, were criteria for discontinuation. All discontinued patients were followed-up for 28 days for safety assessments, where possible.

Efficacy assessments

Primary treatment efficacy was determined based on parasitological cure rates on days 2, 3, 7, 14 and 28 and by the times to parasite and fever clearance and from the proportion of patients without gametocytes.The other outcomes assessed were early treatment failure (ETF), late clinical failure (LCF) and late parasitological failure (LPF).Recrudescence denoted clinical recurrence of malaria after the initial clearance of parasite from circulation. Parasite reappearance after day 14 was interpreted as either true recrudescence or a new infection. Thus, treatment efficacy for cure rates in our context were described as uncorrected since no DNA polymerase chain reaction (PCR) analysis was performed in the four sites.

Safety assessments

All adverse events were monitored and recorded on the case report forms (CRFs). Haematological parameters, liver enzymes and creatinine were assessed for the purpose of detecting abnormal laboratory features that constitute adverse events. Treatment-emergent symptoms of malaria were defined as adverse events occurring anew or worsening from baseline, but occurring before possible recurrence of parasitaemia.

Safety evaluation

Assessment of possible treatment-related adverse events during acute disease is difficult, due to the background dominance of malaria-related signs and symptoms. Malaria clinical features were therefore recorded at baseline, during treatment, and during follow-up visits (see study design).

Sample size calculation

The sample size of patients used for this drug trial was calculated from the table of anticipated proportion (WHO/HTM/RBM/2003) at 95% confidence level and 10% precision. Calculation was based on estimated cure rate for current artemisinin-based antimalarial drug treatment **[3]** With this combination drug having anticipated proportion of treatment failure of less than 5%, the sample size for the trial drug should be 18 (EPI-INFO version 6.04). However, since a minimum sample size of 50 is recommended by the World Health Organization (WHO), we decided to enroll about 50-55 patients on each arm (children and adults) for each site(i.e,200 - 220 each) thus making an estimated total of 400 – 440 patients for the drug therapeutic efficacy test as we had to adjust for losses and withdrawals.

Data Analysis

Data generated from this trial were entered into EPI-INFO version 6.04. Microsoft excel software was also used to plot simple graphs. Various aspects of the data were subsequently analysed using SPSS statistical package version 11. Descriptive statistics were produced for different parameters before figures representing various observations were compared using X^2 0r student t-test or analysis of variance (ANOVA) as appropriate. Pearson's correlation test was used to examine the relationship between selected variables.

Parasite counts:

At screening prior to enrolment, thick and thin blood films were examined. A second, Giemsa- stained thick film was examined with a binocular microscope with an oil immersion objective lens to quantify the parasitaemia. Parasitaemia was measured by counting the number of asexual parasites against a number of leukocytes in the thick blood film, based on a putative count of 8000 leukocytes per microlitre of blood or an adequate mean WBC in the population under investigation. The number of asexual parasites was counted against 200 leukocytes using a hand

tally counter. The number of parasites per microlitre of blood was calculated by using the formula:

$$\text{Parasite Density (parasites } \mu l^{-1}) = \frac{\text{number of parasites x WBC count (8000)*}}{\text{Number of leukocytes counted (200)}}$$

If *P.falciparum* gametocytes were seen, a gametocyte count was performed against 1000 leukocytes (WHO/MAL/82.988).

Temperature:

Axillary temperature was recorded using a digital electronic thermometer.

Ethical Issues:

This work was approved by the Ethics Review Boards (IRBs)/Health Research Ethics committees of the various states who gave written permission to carry out the study. The Heads of communities and authorities of the various Health Centres also consented to the conduct of the study. The study was carried out in accordance with the principles laid down by the World Health Assembly of 1975 on Ethics in Human experimentation and the Helsinki Declaration. The study adhered to Good Clinical Practices (GCP) and conformed to the TDR Standard Operating Procedures (SOP).

Each, participant was informed of the aims, methods, anticipated benefits and potential hazards of the study. Then, written informed consent was obtained by the investigators from every participant or parent / guardian of patients participating in the study. The subject was informed that he/she was at liberty to abstain from participation in the study and that he/she was free to withdraw the consent of participation at anytime. This was written in English and the local languages of the various communities.

RESULTS

General

A total of 4,139 patients were screened in the four sites because they complained of symptoms suggestive of malaria and had not taken any antimalarial medication within the previous seven days. Overall, 446 patients fulfilled the criteria for enrolment and were enrolled but a total of 431 (96.6%) patients completed the study.

The demographic and clinical characteristics of the patients are shown in Table 1.

Defaulters

A total of 431 (96.6%) of the 446 enrolled patients consisting of 203 adults weighing 30Kg and above and 228 children weighing 15 to <30kg completed the study. However, 15 patients defaulted as a result of withdrawal/loss to follow-up and/or protocol violation. The trial profile is shown in Fig. 1.

Efficacy of Artequin™

Parasite Clearance Rate, Profile and Time

Results showed that on DI, 83(40.9%) of the 203 enrolled adults and 72 (31.6%) of the 228 enrolled children no longer had any malaria parasites in their blood. The mean parasite densities in the remaining 120 adults and 156 children were drastically reduced. The clearance rate dramatically increased to 92.1% and 91.7% in adults and in children respectively on D2 until total clearance was achieved in the remaining 16 adults and 19 children on D3.

Adults.

In adults, the geometric mean parasite density of the 203 participants enrolled on D0 was 6,890.8 parasites/μL of blood which decreased to 30.74 parasites/μL on D1, therefore giving a percentage rate of 99.6% (Fig II). The rates on other days (i.e. using the geometric mean parasite densities) were as follows: D2 (99.98%), D3 (100%), D7 (100%) and D14 (100%). However, on day 28, one of the adults (in Lagos site) manifested low-grade parasite density of 360 parasite μL^{-1} blood. We

were unable to infer whether it was as a result of recrudescence or of new infection since no polymerase chain reaction (PCR) technique was used.

Time to parasite clearance (PCT) in 203 adults was determined from the spread sheet data (WHO/MAL/82.988). Parasitaemia completely cleared in 83 patients within 24hrs, 104 cleared in 48hrs while 16 patients were cleared in 72 hours. Time to parasite clearance was therefore calculated as follows:

$$\frac{(83 \times 24) + (104 \times 48) + (16 \times 72)}{203} = \frac{8,136}{203} = \frac{40.1}{\text{hrs}}$$

Children

In children, the geometric mean parasite density of the 228 enrolled patients on day 0 was 11,727.5 parasites/µ of blood which reduced to 54.2 parasites on D1 therefore giving a percentage rate of 99.2% on D1 (**Fig I**)

The rates on other days, using the geometric mean parasite density, were as follows: D2 (99.97%), D3 (100%), D7 (100%) and D14 (100%). As in adults, 6 children in 3 sites manifested low-grade parasitaemia on day 28 and had to be referred. Time to parasite clearance (PCT) in 228 children was also determined from the spread sheet. Here, parasitaemia had cleared in 72 children within 24hrs (D1), 137 children within 48hrs (D 2) and in the remaining 19 children within 72hrs (D3).

Time to parasite clearance was therefore calculated as follows:

$$\frac{(72 \times 24) + (137 \times 48) + (19 \times 72)}{228} = \frac{9672\text{hrs}}{228} = \frac{42.4}{\text{hrs.}}$$

Temperature Clearance Profile

Result showed that 109 of the 203 enrolled adult patients had temperatures below 37.5^{o}C reflecting a possible self-medication with analgesics before coming to the health centre with high parasitaemia. The mean temperature of the 94 adult patients with temperatures ≥ 37.5 ^{o}C was 38.44±0.75^{o}C. The temperature dropped to a mean value of 36.47± 0.67^{o}C in 24hrs. Therefore, the time to fever clearance (FCT= time from the first dose for the temperature to fall below 37

.5^{o}C and remain so for 3 consecutive days) was calculated to be **11.25** hrs (Fig III).

The mean temperature of 132 children who were febrile (T$\geq$37.5^{o}C) was 38.56 $\pm$ 0.79^{o}C. The temperature of these patients dropped to 36.57$\pm$ 0.81^{o}C by D1 (24hrs). The time to fever clearance was calculated to be 13.25 hrs.

Anti-Gametocyte Activity of Artequin™

Only three of the four sites investigated the gametocyte carriage rate and the changes in geometric mean gametocyte densities (GMGD) in the patients. The gametocyte carriage rate for adults and for children were 5.9% and 4.4% respectively.

In ten children where gametocytes were found, the mean gametocyte count dropped from pre-treatment levels on day 1. The gametocytes cleared in 4 patients in 24 hours, 3 were cleared in 48 hours and 3 in 72 hours. Time to gametocyte clearance in children was calculated (as in parasite clearance time) to be 45.6 hours.

In twelve adults where gametocytes were found, the gametocytes cleared in 5 patients in 24 hours. It cleared in 4 patients in 48 hours and in the remaining 3 patients in 72 hours. The gametocyte clearance time in adults was 42.0 hours.

Safety and Tolerability

Artequin™ was well tolerated. Adverse events were carefully investigated according to the protocol. No serious adverse event (SAE) was reported during the study in the four trial sites. Many AEs of the antimalaria drugs were most likely related to the underlying malaria disease. Of the 39 reports, 10 patients (2.3% of total patients treated) reported vomiting **(fig 4).** The others were as follows: headache (9, 2.1%), dizziness (12, 2.8%) and abdominal discomfort (4, 0.9%). There was one report (0.2%) each of sleeplessness, fast breathing, weakness and vertebral pain.

Laboratory Indices of Safety

The result of laboratory characteristics of patients at enrolment is shown in **Table 2.** The values in individual patients varied in accordance with the seriousness of the

infection. However, the mean values of all parameters were within normal limits on D0. The WBC count showed slight increase in both infected adults and children. However, the increase was more prominent in the lymphocyte count with corresponding decrease in neutrophils as treatment progressed.

The slight increase in lymphocyte count from 43.25 $\pm$ 13.19% on D0 to 56.15 $\pm$ 13.22% on day 14 in adults and 43.0±15.35% to 56,55±13.77% in children is consistent with manifestations of recovery in malaria infection. Here, immune mechanisms seemed to elicit cellular responses which rose against further infection.

The percentage recticulocyte count remained within normal limits for inhabitants of the various sites although normal values in some countries could be 0.2 – 2.0%. There was a slight decrease in mean haemoglobin value on days 7 and 14 before returning to normal on day 28. All these changes were not statistically significant ($P>0.05$). Only a few patients had PCV of over 40.0 % on day 0 in the sites used. The mean PCV was 39.4 $\pm$ 5.36 for adult men and 36.0 $\pm$ 5.05 for adult women; while in children it was 33.4±4.87% and 34.1±5.23% in males and females respectively. However, there were decreases in the mean values on D7 and D14 before rising to the normal mean values for that period of investigation. Other haematological parameters, viz platelets, eosinophils and monocytes had varying values which were not statistically different ($P > 0.05$) from the D0 values.

The clinical chemistry values on D0 are also shown in **Table 2.** The mean starting values for total bilirubin was 6.16 $\pm$ 6.35 µmol $^{-1}$ in adults and 5.12±5.63 µmol $^{-1}$. The level remained within this value from D0 to D28. There were also no significant decreases or increases ($P>0.05$) in the values of serum alanine aminotransferase (ALT, GOT), serum aspartate amonitransferase (AST, GPT) and alkaline phosphotase (ALK).

DISCUSSION

We have carried out the Therapeutic Efficacy Trial on artesunate + mefloquine (Artequin™) in four geographic areas of Nigeria. The main outcome of our study was

that a 3-d course with this artesunate + mefloquine blister pack was both highly effective and well tolerated in the treatment of acute uncomplicated *P. falciparum* malaria in all the sites.

The geometric mean parasite density was drastically reduced in adults and in children within 24h after treatment and completely cleared by day 3 in the remaining patients who completed the study. The parasitological response in both adults and children were 100% in D3, D7, and D14.

The rapid clinical response was shown by a drop in temperature to normal values (viz. below 37.5^{o}C) on the 2nd day. This rapid clinical and parasitological response confirmed the previous findings of others [**16, 17, 18, 19, 21, 22]** who have, for many years worked in countries where, as in Nigeria, multi-drug-resistant strains predominate.

Apart from the rapid clearance of asexual forms of *P. falciparum,* Artequin™ therapy was also beneficial in inducing significant reduction in gametocyte rate and density. . The data suggest that Artequin™ ultimately cleared gametocytes from peripheral blood. This shows that Artequin™ exhibits considerable gametocidal effect. The second observation was that patients with mixed infections of *P. falciparum and P. malariae* were cured parasitologically and clinically.

It was observed in the course of this trial, that parasitaemia consisting of young trophozoites appeared on day 28 in 1 of the adults and 6 of the children. Parasitaemia was not associated fever or other symptoms of malaria. Since we did not PCR analysis of the parasite genotypes, it was not possible to determine whether these were new infections or recruidescence. However, given that our trial was conducted in areas of malaria transmission, it is likely that the observed day 28 parasitaemia could be re-infections instead of recrudescence**[21]**. Falade et. al. **[23]**, by genotyping new infections seen between 2nd and 4th week post-therapy, attributed this phenomenon to new infections. There is therefore the need to perform more molecular genotyping of the parasite strains in subsequent trials to confirm this observation.

Artequin™ from the experience of this study is safe and well tolerated. The laboratory values were not significantly different pre-and post-treatment. The marginal variations in liver function test results may be related to stabilization of the liver following successful treatment. The same result was observed with mean PCV values which returned to normal after recovery. The slight reduction in mean platelet count was consistent with the reported findings of relative thrombocytopenia in 50 to 75% of patients with acute malaria **[24]**.

We conclude from this study that our results have confirmed the efficacy of Artequin™ in the treatment of *Plasmodium falciparum* malaria in four sites in Nigeria. It also exhibited significant gametocidal activity. As observed by other workers, the rapid parasitological response corresponded to the fast clinical response.

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Table 1: Demographic and Clinical characteristics of Patients at enrolment (Day 0)

Characteristics	Group 1 Adults (>30kg)	Group 2 Child 15-30kg
Sex ratio		
.. Male	78 (38.4%)	139 (61.0%)
Female	125 (61.6%)	89 (39.0%)
Mean age (Yrs)	22.5 ± 11.5	7.1 ± 2.7
Range	9-65	(3-13)
Mean weight (kg)	50.9 ± 14.1	20.5 ± 4.7
Range	30-94	15-30
Mean Height (cm)	156.8 ±10.7	115.5±14.4
Range	124-185	80.0-146.5
Mean Parasite Density (μl^{-1})	19,797.6 ±33,397.6	27,010.2 ± 35,704.4
Range	1000-220,000	1016 – 235,294
Geometric mean parasite density (μl^{-1})	6890.8	11,727.5
Mean axillary Temperature (^oC)		
<37.5^oC	36.6 ± 0.52 (n=109)	36.6 ± 0.39 (n=96)
≥ 37.5oC	38.4 ± 0.75 (n=94)	38.6 ± 0.79 (n=132)

Table 2: Laboratory Characteristics of Patients On Day 0 and Day 14

	Group 1 (Adults) (> 30 kg) Day 0	Day 14	Group 2 (Children) (15- 30kg) Day 0	Day 14
Packed Cell volume (%)				
Male	39.4± 5.36 (n=71)	37.47±5.82	33.4 ±_4.87 (n=137)	32.49±4.32
Female	36.0 ± 5.05 (n=120)	34.19±4.49	34.1 ± 5.23 (n=85)	33.03±4.61
Erythrocyte Sedimentation Rate (ESR)	23.8 ± 26.7 (n=53)	29.08 ± 17,955	42.7 ± 29.2 (n=49)	24.78 ± 17.708
Haemoglobin mg/L (n=113)	11.76 ±1.65	10.86 ± 1.63	10.65 ± 1.75	9.11 ± 1.26
Total White Blood Cells (WBC)	5,272.6± 1869.7 (n=113)	6,548.33 ±5292.095	7,252.8 ± 11125.2 (n=107)	7,466.67 ± 7,043650
WBC Differential (%) n=113				
Lymphocytes ..	43.25 ± 13.19	56.15 ± 13.22	43.0 ± 15.35	56.55 ± 13,77
Neutrophils ..	52.85 ± 13.35	38.20 ± 10,82	98.07 ± 16.37	41.62 ± 18.70
Monocytes ..	2.4 ± 3.28	1.02 ± 1.59	1.67 ± 1.82	.57± .844
Eosinophils ..	2.32 ± 2.61	3.95 ± 4.0	1.42 ± 1.75	2.85 ± 4.907
Alkaline Phosphatase ..(iu/L). ..	59.56 ± 121.58	105.10 ± 76.57	220.64 ± 85.33	178.124 ± 72.32
Aspartate Amino-transferase (iu/L)	29.12 ± 19.54	34.347 ± 68.82	38.58 ± 42.55	15.608 ± 18.05
Alanine aminotransferase(iu/L)	20.41 ± 24.94	27.538 ± 45.77	23.04 ± 20.40	10.850 ± 16.88
Total Bilirubin ..(µmol/L)	. 6.16 ± 6.35	4.295 ± 5.20	5.12 ± 5.63	5.800 ± 15.38
Conjugated bilirubin (µmol/L)	. 2.75 ± 3.25	2.5073 ± 2.56	0.49 ± 0.58	.3979 ± 2.53
Urea	.. 8.96 ± 7.14	16.935 ± 12.48	8.22 ± 5.95	14.457 ± 7.17
Creatinine	34.12 ± 31.81	.919 ± 0.33	21.97 ± 21.78	.759 ± .28

Table 3 Study outcome of trial participants

Characteristics / Outcome - Group A	Group A	Group B	Group A and B
	Artequin® (600/750) Adults >30kg	300/375 for children 15 - 30kg	All Participants
Number enrolled	208	238	446
Number completed	203	228	431
Withdrawal / loss to follow up	5(2.5%)	10(4.45)	15(3.5%)
Early treatment failure (ETF)	0	0	0
Late Clinical Failure (LCF)	0	0	0
Late Parasitological Failure (LPF)	0	0	0
Adequate Clinical and Parasitological Response (ACPR) Day 14	203(100%)	228(100%)	431(100%)
Adequate Clinical and Parasitological response (ACPR)* Day 28	202(99.5%)	222(97.4%)	424(97.45%)
Fever Clearance Time	11.25hrs	13.25hrs	12.25
Parasite Clearance Time	40.1 hrs	42.4hrs	41.3

*** Not Polymerase chain Reaction (PCR) corrected**

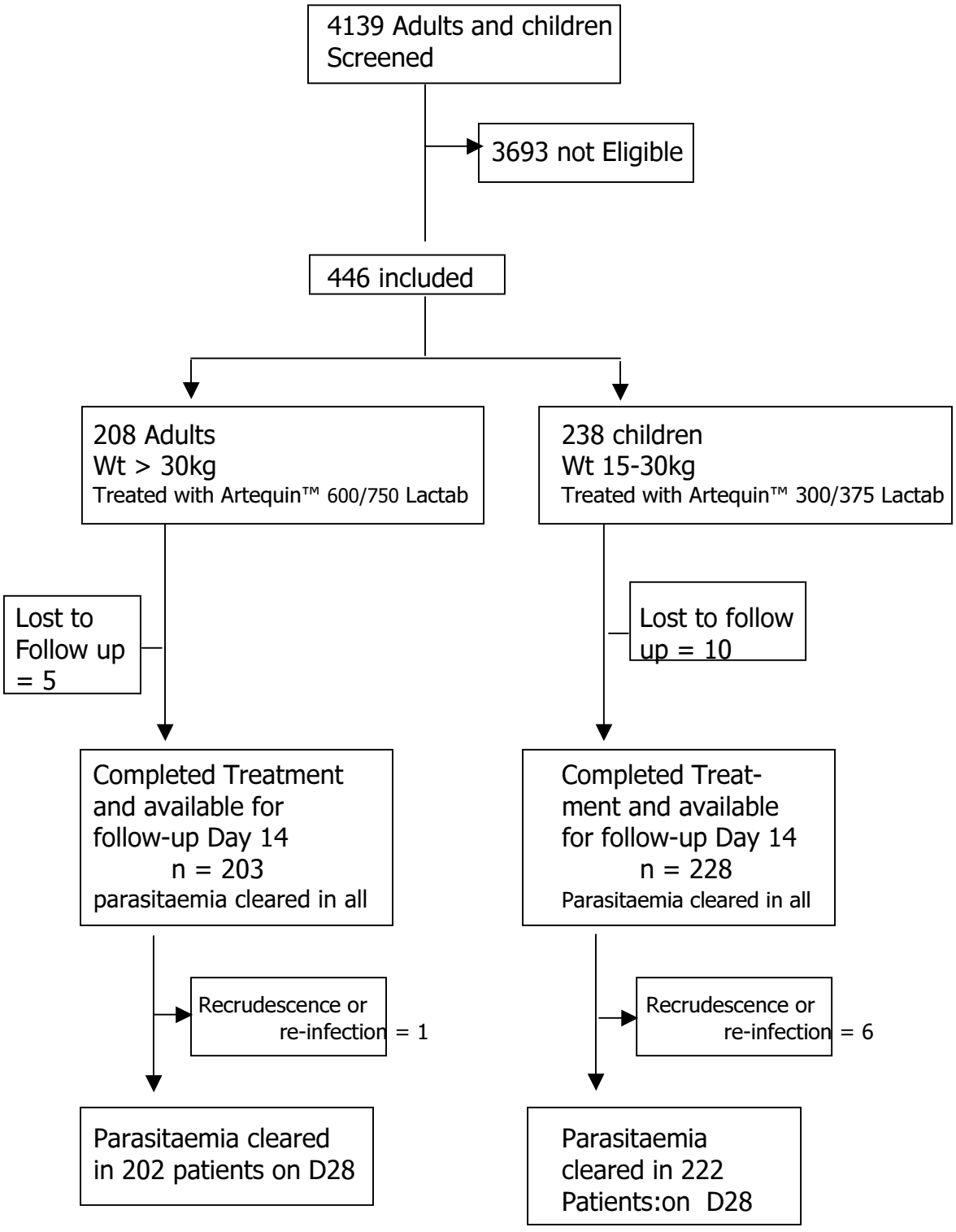

Figure 1

ARTEQUIN™ TRIAL PROFILE IN 4 GEOGRAPHIC ZONES OF NIGERIA

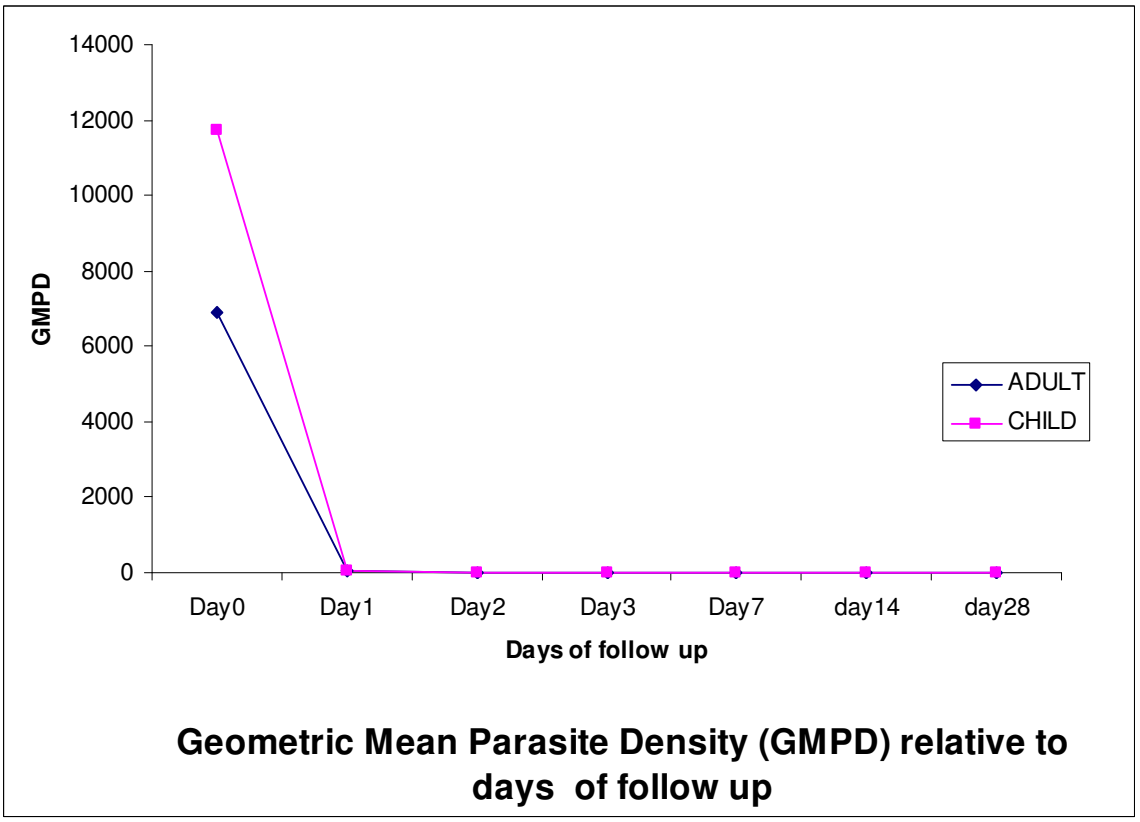
14000
12000
10000
8000
6000
4000
2000
0
GMPD
Day0
Day1
Day2
Day3
Day7
day14
day28
Days of follow up
ADULT
CHILD
Geometric Mean Parasite Density (GMPD) relative to days of follow up

Figure 2

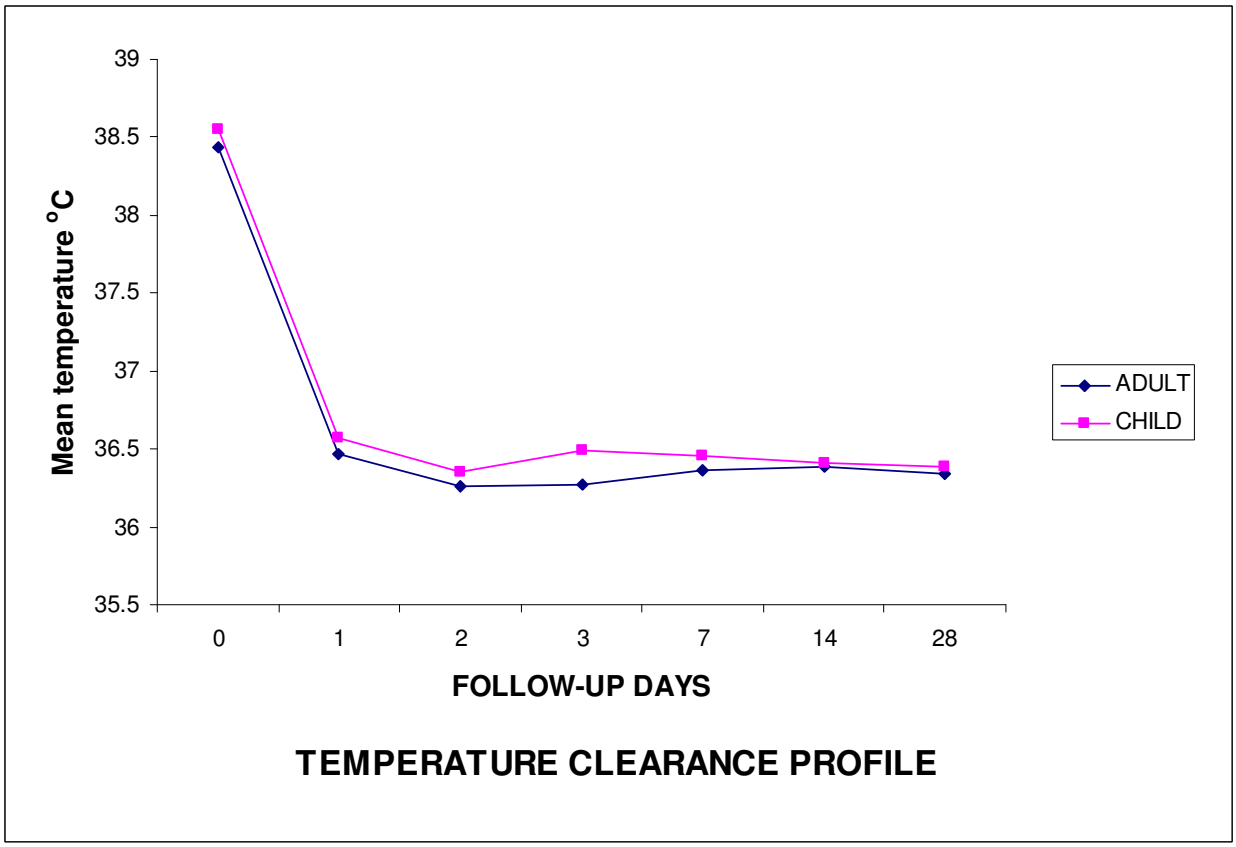
39
38.5
38
37.5
37
36.5
36
35.5
Mean temperature °C
0
1
2
3
7
14
28
FOLLOW-UP DAYS
ADULT
CHILD
TEMPERATURE CLEARANCE PROFILE

Figure 3

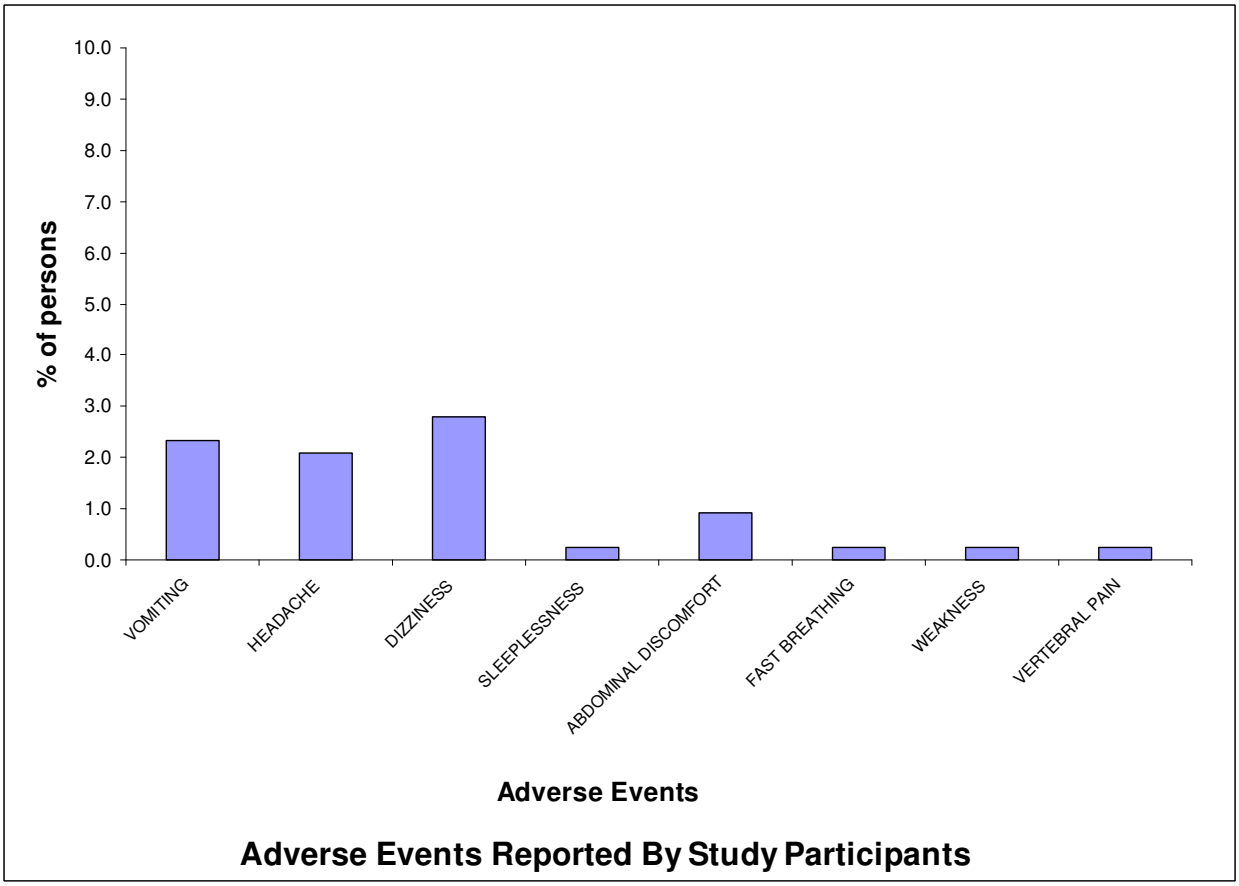
10.0
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
% of persons
VOMITING
HEADACHE
DIZZINESS
SLEEPLESSNESS
ABDOMINAL DISCOMFORT
FAST BREATHING
WEAKNESS
VERTEBRAL PAIN
Adverse Events
Adverse Events Reported By Study Participants

Figure 4