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Treatment of Malaria Parasitaemia in Infants and their Mothers

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Abstract

Malaria is an infection sustained by three parasites namely: Plasmodium falciparum, Plasmodium vivax, and Plasmodium ovale. Plasmodium falciparum is the most common and virulent parasite. These parasites are present in different areas of the sub-Saharan African countries and Asia. In 2010, there were an estimated 219 million cases of malaria resulting in 660,000 deaths and, approximately, two-thirds were children. In sub-Saharan African countries, maternal malaria is associated with up to 200,000 estimated infant deaths yearly. Chloroquine was the world's widely used antimalarial drug, but Plasmodium falciparum is now increasingly resistant. However, Plasmodium ovale and Plasmodium vivax are sensitive to chloroquine. Pregnancy makes women vulnerable to malarial parasites and the risks of anemia, miscarriage, stillbirth and prematurity increase. Resistance to chloroquine is a major concern for treatment of malaria and alternative drugs are needed.

Proguanil is safe, being very rarely associated with severe adverse reactions. Chloroquine, mefloquine, sulfadoxine-pyrimethamine, and amodiaquine have been found to be active against Plasmodium falciparum in-vitro. In the Cameroons, chloroquine was initially replaced by amodiaquine and artemisinin-lumefantrine was gradually introduced in 2004. Tanzania replaced chloroquine with sulphadoxine-pyrimethamine, and in 2006 artemisinin was introduced in the therapy. Pyrimethamine-sulfadoxine should be reserved as a second-line-treatment. Mefloquine may provoke severe neuropsychiatric reactions. In the treatment of Plasmodium malaria, which has a high mortality rate if untreated, a greater risk of adverse reactions to malarial drugs is acceptable. The aim of the present study is to review the published data on the treatment of malaria in infants and their mothers.

Key Words: Antimalarials, Infants, Malaria, Mothers, Pregnant-women, Resistance.

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1-INTRODUCTION

Chloroquine was, for a long time, the world's most widely used antimalarial drug, but the most common and virulent parasite, Plasmodium falciparum, is now increasing resistant. Chloroquine can be used, however, when Plasmodium ovale and Plasmodium vivax are the causative malaria parasite (1). Chloroquine (a 4-aminoquinoline) is well absorbed, widely distributed in the body tissues, slowly metabolized by the liver and only very slowly cleared from the body. There is no evidence that standard-dose treatment during pregnancy is hazardous, and there is good evidence that weekly prophylaxis is safe and advisable where malaria is endemic. Use during lactation exposes the neonate to less than 5% of the weight-adjusted maternal dose, which is probably not enough to protect the neonate from infection (1).

In 2010, there were an estimated 219 million cases of malaria resulting in 660,000 deaths. Most cases (approximately two-thirds) occur in children. Children are more vulnerable than adults to malaria parasites. In sub-Saharan Africa, maternal malaria is associated with up to 200,000 estimated infant deaths yearly. Residents in endemic areas develop considerable immunity over time, but pregnancy makes women more vulnerable, and infection during pregnancy increases the risk of anemia, miscarriage, stillbirth and prematurity. Transplacental spread is oncoming, but infection sometimes occurs during delivery, although florid symptoms (including fever, jaundice, an enlarged liver and spleen and low platelet count) usually only manifest themselves 2-8 weeks later. Infection is considered severe if there is shock, acidosis, hypoglycemia or cerebral symptoms or more than 5% of red cells are involved. In Plasmodium vivax and Plasmodium ovale infections, treatment with chloroquine can leave some organisms dormant in the liver unless primaquine is given as well. There are a few areas where even Plasmodium vivax has now become resistant to chloroquine and where it can still be appropriate to treat overt infection with mefloquine (15 mg/kg by mouth followed, after 12 hours, by a second 10 mg/kg dose (1). Asexual malarial parasites flourish in host erythrocytes by digesting hemoglobin in their acidic digestive vacuoles, a process that generates free radicals and iron-bound heme (ferriprotoporphyrin IX) as highly reactive byproducts (2). Perhaps aided by histidinerich proteins and lipids, heme is sequestrated as an insoluble, chemically inert malarial pigment termed hemozoin. Many theories for the mechanism of action of chloroquine have been advanced (3).

Current evidence suggests that quinolines interfere with heme detoxification. Chloroquine a weak base, concentrates in the highly acidic digestives vacuoles of susceptible Plasmodium, where it binds to heme and disrupts its sequestration. Failure to inactivate heme or even enhanced toxicity of drug-heme complexes is thought to kill the parasites via oxidative damage to membranes, digestive proteases, or other critical biomolecules. Other quinolines such as quinine, amodiaquine, and mefloquine, as well as other amino alcohol analogs (lumefantrine, halofantrine), and Mannich base (pyronaridine), may act by a similar mechanism, although differences in their actions have been proposed (4, 5).

Chloroquine-sensitive Plasmodium parasites concentrate the drug to higher levels than did chloroquine-resistant organisms (6). A parasite-encoded efflux mechanism may account for the reduced levels of chloroquine in the digestive vacuoles of chloroquine-resistant parasites (7, 8). In the World Health Organization (WHO) African regions, malaria still remains a public health problem that constitutes a serious obstacle to development. This is due to the high

intensity of transmission and lack of sufficient resources to apply effective methods of control (9). However, the recent adoption of the primary health care delivery system offers a new basis for antimalarial strategies, and this has led to renewed hope of (1) reducing mortality and morbidity, (2) minimizing harmful effects on socioeconomic development, (3) achieving ultimate eradication, and (4) protecting the status of areas freed from malaria.

Primary health care systems make it possible to ensure the ready availability of antimalarial drugs for treatment and for chemoprophylactic schemes to protect young children and pregnant women. However, with the presence of chloroquine resistance in several countries, a critical review of the use of drugs is required. In countries of Southern-Africa where malaria is unstable and liable to be endemic, control is achieved mainly by the seasonal use of residual insecticides and chemotherapy. So far, eradication has been achieved only in Réunion, but it is also likely to be achieved soon in the islands of Sao Tome and Principe. Considerable reduction of transmission has been achieved in Ethiopia.

2- MATERIALS AND METHODS

2-1. Literature Search

The following databases were searched for relevant papers and reports: MEDLINE via (PubMed), CINAHL, EMBASE, Google scholar as search engines; December 2017 was the cutoff point. Key references from extracted papers were also hand-searched.

2-2. Search Terms

The following key words "chloroquine dosing neonates", "chloroquine effects neonates", and "chloroquine resistance neonates" were used. In addition, the book Neonatal Formulary (1) was consulted.

3-RESULTS

3-1. Prevention in visitors

Give 5 mg/kg of chloroquine-base to children by mouth once a week in areas where sensitive parasites are endemic. Start 1 week before entering the area and stop 4 weeks after leaving. Consider giving proguanil as well (1).

3-2. Cure

Give a 10 mg/kg loading dose of chloroquine-base intravenously or by mouth and then three 5 mg/kg doses (given at 24 hours intervals) starting 6 hours after loading dose should be given. In Plasmodium vivax or Plasmodium ovale infections, there may be a cause for giving primaquine after this to eliminate any dormant parasites (1).

3-3. Eradicating liver organisms

Giving 300 μg/kg of primaquine once a day by mouth for 2 weeks will usually kill residual organisms. A higher dose may occasionally be necessary but increases the risk of haemolysis in those with the Mediterranean and Asian variants of Glucose-6-phosphate dehydrogenase (G6PD) deficiency. Use in young children has not been studied (1).

3-4. Toxicity

Excess chloroquine is toxic to the heart and the central nervous system. Prompt high-dose diazepam (2 mg/kg daily), and ventilation seem beneficial. Gastric lavage may be appropriate once the airway has been protected, and activated charcoal may reduce gut absorption. Intravenously adrenaline helps control hypotension. Correct any acidosis. Phenytoin or a beta-blocker can be used to treat arrhythmia (1).

3-5. Effects of antimalarial-drugs in infants and children living in malaria endemic areas

Chronic use of chloroquine and hydroxychloroquine in the treatment of rheumatic disease carries a small risk of

sight-threatening pigmentary retinopathy. To obtain safety data for its use in pregnancy, Klinger et al. (10) did ophthalmic examinations in 21 children born to women who took these drugs during pregnancy. Average daily maternal doses of the two drugs were 371 mg hydroxychloroquine and 332 mg chloroquine. The mean duration of gestational exposure was 7.2 months. No ophthalmic abnormality was detected in their children. Therapeutic use of these drugs during pregnancy may not pose a significant risk of ocular toxicity to offspring.

In malaria-endemic regions, the impact of malaria upon pregnancy and the value of chemoprophylaxis for malaria for pregnant women remain controversial. Nyirjesy et al. (11) prospectively studied 302 pregnant women who presented in labor to Central Médical Evangélique, Nyankunde, Zaire. Nyirjesy et al. (11) evaluated the incidence of malarial infection in mothers, placentas, and neonates and examined the effect of infection on birth-weight and perinatal mortality. These authors analyzed the outcome of pregnancy in relation to prophylaxis with chloroquine, controlling for parity and prenatal clinic attendance. Peripartum smears of maternal blood (21%), placentas (33%), cord blood (9%), and neonatal malaria increased the risk of perinatal death (relative risk = 12.4), and low-birth-weight (relative risk = 3.7). Neonatal malaria increased the risk of perinatal death (relative risk = 0.38). Peripartum malaria increases the risk of perinatal death and low-birth-weight. Chemoprophylaxis with chloroquine during pregnancy may have a protective effect, even in certain areas where chloroquine-resistant Plasmodium falciparum is endemic.

The use of the 4-aminoquinoline is controversial. The current practice of discontinuing these medications because of pregnancy makes little sense as the half-life of these drugs is so long. Patients with Systemic lupus erythematosus (SLE) have increased fetal wastage and one of the factors known to contribute to this fetal wastage is disease activity. It is also known that discontinuing the 4-aminoquinoline antimalarial-drugs can precipitate flares of disease in lupus patients. Mothers and their potential offspring are therefore at risk for flares of disease and pregnancy failure if these medications are discontinued because of pregnancy. Unlike most centers in North-America, Parke and Rothfield (12) continue their patients on these medications throughout pregnancy and to date have documented 16 lupus patients who have taken these drugs throughout pregnancy. There were no congenital abnormalities in infants and follow-up to date has revealed no evidence of ocular or oral deficits in any children. One patient experienced a flare of disease when her antimalarial therapy was temporarily discontinued.

Etuk et al. (13) examined the diagnosis of malaria and pattern of prescription of antimalarial-drug in the most vulnerable age group (children were aged < 5 years) in the study environment in order to identify the possible shortcomings and suggest solutions so as to improve the treatment outcome in future. Analysis of the data revealed that more male (213) than female (169) children were admitted for malaria treatment. Fever with convulsion (55.8%) was the commonest presenting symptoms, and anemia was the most frequent complication of malaria recorded. Chloroquine was found to be the most prescribed antimalarial agent and overall an artemisinin-based drug was prescribed either as first- or second-line treatment in only 18.2% of the cases. The death rate recorded was 16%. The pattern of antimalarial-drugs prescription in the study center in most cases did not meet the recommended guidelines.

The prescriptions were predominantly chloroquine, instead of artemisinin based. The death rate was comparatively high. Measures to raise the level of awareness among the practitioners on the Current National Policy on malaria treatment through seminars and workshops were suggested. Following the WHO protocol for in-vivo tests in areas with intense transmission of uncomplicated falciparum malaria, a randomized comparison of the in-vivo efficacy of chloroquine alone, sulfadoxine-pyrimethamine alone, and their combined administration was carried out in the third quarter of 2001 in Kaberamaido District, Northeastem Uganda (14). Malaria in the study area is hyper-endemic, with a high prevalence of Plasmodium falciparum. The patients had a median age of 15 months. Of the 117 originally enrolled patients, 104 had a complete follow-up with presentation at all scheduled examinations.

In the chloroquine group (n = 42), 55% were classified as adequate clinical response, 26% as early treatment failure, and 19% as late clinical failure. In the sulfadoxine-pyrimethamine group (n = 30), the respective figures were 83%, 13%, and 6%. In terms of clinical cure rate, speed of clinical relief and parasite clearance the combined treatment proved to be the most effective of the three drug regimens. In the patients with adequate clinical response, a significant post-therapeutic increase of the hematocrit was observed, which was particularly marked in patients who had also cleared their parasitaemia. Increase of the efficacy of chloroquine with age indicates the early development of semi-immunity in the study area, with conserved efficacy of chloroquine in semi-immune response.

In spite of increasing resistance, chloroquine remains the primary drug for treatment of malaria in most sub-Saharan African countries. Zucker et al. (15) evaluated the effects of drug treatment policy on the case-fatality rates of children, adjusting for differing distributions of malaria and severe anemia. In 1991, 63% of children were treated with chloroquine while the remaining 37% were treated with a regimen that would eliminate and clear parasitaemia. Case-fatality rates were 13% and 4.1%, respectively ($p < 0.001$); the proportion of deaths attributed to chloroquine treatment was 69%. The trend in case-fatality rates for malaria decreased as an increasing proportion of children received an effective treatment regimen; adjusted malaria case-fatality rates were 5.1%, 3.6%, and 3.3% in 1992, 1993, and 1994, respectively, when 85% of children in 1992 and 97% of children in 1993-1994 received effective therapy. These 4 years of data provide strong evidence that continued use of chloroquine in areas with resistance is contributing to excess Plasmodium falciparum-related deaths.

The role of chloroquine as the first-line drug to treat Plasmodium falciparum is almost universally becoming questionable. Kebede et al. (16) conducted a study in one of the country's unstable malaria endemic area, North Shoa with the objective of assessing the in-vivo treatment efficacy of chloroquine to Plasmodium falciparum malaria using the standard WHO 14 days of treatment response monitoring guideline. A total of 427 patients were followed among which 87.8% showed treatment failure. This was more pronounced in children than in adults (Chi-squared for trend = 8.16; $p < 0.01$). Clinical presentation with high grade fever on day 0 was found to be more predictive of treatment failure in children (odds ratio [OR] = 2.06; 95% confidence interval [CI] = 1.26, 3.36; $p < 0.005$). Tendency to remain febrile on subsequent follow-up days was also more observed in children compared to adults. Treatment failure was further associated with high Parasite Density index on day 0 in all age groups.

Supplemented with large scale sensitivity studies, it is high time that a switch to alternative drugs needs due consideration by policy makers. Since 1993 sulphadoxine-pyrimethanine has been used as the first-line drug for uncomplicated Plasmodium falciparum malaria in Malawi. To investigate the current efficacy of sulphadoxine-pyrimethanine and other antimalarial drug resistance, Takechi et al. (17) studied in-vivo and in-vitro response to sulfadoxine-pyrimethamine, chloroquine (CQ), mefloquine (MF), quinine (QN), and halofantrine (HF) in Salima, central Malawi. In a follow-up of 14 days, nine (13.8%) of 65 children aged less than 5 years showed moderate (RII) and high (RIII) RII/RIII level parasitological resistance, and in in-vitro microtests 18 (62.1) of 29 isolated showed < 90% inhibition of schizont maturation at pyrimethamine 75 μmol/l blood medium mixture explained by acquired immunity in this holoendemic area. The discrepancy between in-vivo an in-vitro results might be partially explained by acquired immunity in this holoendemic area. Compared with an in-vitro study conducted in 1988 in another region of Malawi using the same cut-off point, the proportion of resistant isolates had decreased significantly ($p < 0.01$).

Although 31% of isolates were borderline, showing schizont maturation at 0.8 μmol/l blood but no schizont at 1.6 μmol/l in the study by Takechi et al. (17), these results suggest possibly recovery of chloroquine sensitivity after long-term absence of drug pressure. Resistance remains a major problem in malaria control. Monitoring resistance patterns in-vitro provides early signs of impending loss of therapeutic efficacy of the standard treatment, and may detect changing patterns in alternative drug resistance. While there is a broad evidence for the adverse effects of Plasmodium falciparum infection in pregnancy, and the WHO recommends preventive strategies, there is markedly reduced efficacy in Sub-Saharan Africa on the most widely available affordable and used antimalarial drug for chemoprophylaxis-chloroquine. During 1987-1990, Steketee et al. (18) studied pregnant women in an area of high malaria endemic in rural Malawi to compare the efficacy of chloroquine (the drug recommended by national policy) with mefloquine (a relatively new and high effective antimalarial) in preventing low-level-birth-weight due to prematurity and intrauterine growth retardation. Among 1,766 women monitored during at least six weeks of pregnancy with observed ingestion of their regimen monitored and facility delivery of a live born singleton, their neonates had a birth-weight of 2,905±461 grams and 18.6% had a low-body-weight. In a multivariate, analysis factors significantly associated: first birth (OR= 4.27), female infant (OR= 2.92), and placental blood Plasmodium falciparum infection (OR= 1.71).

Factors significantly associated with intrauterine grown retardation low-birth-weight included first birth, female infant, low maternal weight, and placental malaria. Factors significantly associated with preterm low-birth-weight included maternal syphilis infection, umbilical cord blood malaria, first birth, low maternal weight, and female infants. Use of an effective antimalarial mefloquine was protective against low-birth-weight through its effect on reducing placental and umbilical cord blood malaria infection. The proportion of low-birth-weight neonates born to women on mefloquine (12.5% [parity-adjusted for population of delivering women]) was significantly lower than the proportion born to women on chloroquine (15.5%; $p = 0.05$). Effective prevention of malaria in pregnant women in malaria-endemic-settings may reduce the likelihood of low-birth-weight by 5 to 14%, and may reduce the amount

of preventable low-birth-weight more than 30%. When evaluating antenatal care programs, health policy makers must consider providing an effective drug (either mefloquine or other drugs identified in an additional studies e.g., sulfa-pyrimethamine compounds) as a means to prevent low-birth-weight and its consequences. Barsalou et al. (19) assessed if maternal intake of antimalarials throughout pregnancy lowered the risk of cardiac and non-cardiac neonatal lupus. A total of 315 individuals were screened and 268 participants were included. Outcomes were cardiac and non-cardiac lupus. These authors hypothesized that prenatal antimalarials exposure would decrease the risk of cardiac neonatal lupus but could not be categorized as unaffected since their full non-cardiac neonatal lupus.

A total of 268 pregnancies were included; 73 were exposed to antimalarials throughout pregnancy. Ninety-nine children developed neonatal lupus, 117 remained unaffected and 52 children did not develop cardiac neonatal lupus. Logistic regression suggested a protective of antimalarial on cardiac neonatal lupus, but results were not statistically significant. The effect of antimalarials on non-cardiac neonatal lupus was not significant ($p = 0.21$). In this large single-centre cohort study, exposure to antimalarials on non-cardiac neonatal lupus was associated with a decreased probability of developing cardiac but not non-cardiac neonatal lupus.

A randomized, double-blind, placebo-controlled trial, which compared the effects of three interventions (weekly chloroquine prophylaxis, dial iron and weekly folic acid supplementation, and case management of malaria) on congenital malaria, maternal hemoglobin and fetal outcome, was conducted among primigravidae residents in Hoima District, Uganda (20). Among 473 neonates examined at birth or within 7 days of birth, 198 (42%) were parasitaemic, the level of parasitaemia in an infant being strongly correlated with those of placental ($p < 0.001$). However, 33 (17%) of the parasitaemic neonates were born to mothers who had placental but not peripheral parasitaemia, 22 (11%) to mothers who had peripheral but not placental parasitaemia, and 12 (6%) to mothers with neither peripheral nor placental parasitaemia. Overall, 163 neonates were each examined for malarial parasites at birth and 1 month later. Of the 76 (47%) found to have parasitaemia at birth, 37 (23%) appeared parasitaemic at 1-month follow-up but 28 (17%) were still parasitaemic at that time. Among the infants born to mothers who only received case managements of malaria during pregnancy, parasitaemia at birth was associated with infant anemia at birth (i.e. < 140 grams hemoglobin/l; $p = 0.03$). Infants found to be parasitaemic at the 1-month follow-up had lower mean concentration of hemoglobin at that time than their stone counterparts ($p = 0.03$). Parasitaemia at birth was not significantly associated with low-birth-weight, in any of three intervention groups. The intervention given to the mother had no significant effect on the parasitaemia of her neonate, either at birth or at the age of 1 month. Congenital malaria per se may have little influence on birth-weight but may have an impact on anemia.

In conclusion, congenital parasitaemia was not associated with birth-weight, but was related to anemia at birth in infants born to mothers who had only received active case management during their pregnancies.

The parasitological, clinical, and hematological response to chloroquine treatment were studied in children during a 28-day follow-up in an area of Côte d'Ivoire with intermediate chloroquine resistance (21). The parasitological, clinical, and hematological responses to Fansidar were also investigated in patients

who returned to the health centre within 28 days with symptoms of malaria. Of 82 children aged 0 to 9 years who completed the study, only 67% were parasite-negative on thick blood film on day 7, which decreased to 21% by day 28. While chloroquine treatment still produced clinical remission at day 7 in 95% of the children, 35% had recurrent fever with concomitant parasitaemia before day 28. All fever cases subsequently with Fansidar remained parasite-negative over a period of 28 days. On day 28 the hematocrit levels were higher in those children who responded successfully to treatment with either chloroquine or Fansidar than in the children who were still parasite-positive but without fever (two-tailed t-test, $p = 0.02$). The rate of resistance to chloroquine was most pronounced among the younger children (aged less than 5 years), 8% of whom showed clinical failure by day 14. The present findings underline the importance of monitoring the durability of response to chloroquine treatment for at least 14 days in young children in Côte d'Ivoire.

Anti-malarial drug resistance to chloroquine and sulfadoxine-pyrimethamine has spread from Southeast Asia to Africa. Furthermore, the recent emergence of resistance to artemisinin-based combination therapy in Southeast Asia highlights the need to identify new anti-malaria drugs (22). Doxycycline is recommended for malaria chemoprophylaxis for travel in endemic areas, or in combination with the use of quinine for malaria treatment when artemisinin-based combination therapy is unavailable or when the treatment of severe malaria with artesunate fails. However, doxycycline is not used in young children under 8 years of age due to its contraindication due to the risk of yellow tooth discoloration and dental enamel hypoplasia. Doxycycline was developed after tetracycline and was labeled with the same side-effects as the earlier tetracyclines. However, recent studies report little or no effects of doxycycline on tooth staining or dental enamel hypoplasia in children under 8 years of age. In the United States, the Centers for Disease Control and Prevention have recommended the use of doxycycline for the treatment of acute and chronic chloroquine fever and tick-borne rickettsial diseases in young children. It is time to rehabilitates doxycycline and to recommend it for malaria treatment in children under 8 years of age.

In two maternity clinics in Ouidah, an observational study was conducted between April 2004 and April 2005. Among 1,090 singleton live births, prevalence of placental malaria and low-birth-weight were 16% and 17%, respectively (23). After adjustment, there was non-significant association between placental malaria and low-birth-weight ($p = 0.10$). Multiple linear regression showed a positive association between placental malaria and decreased birth-weight in primigravidae. More than 98% of the women reported regular chemoprophylaxis and chloroquine was detectable in 99% of urine samples. Protection from low-birth-weight was high in women reporting regular chloroquine prophylaxis, with a strong duration-effect-relationship (test for linear trend; $p < 0.001$), Despite high parasite resistance and limited effect on placental malaria, a chloroquine chemoprophylaxis taken at adequate doses proved to be still effective in reducing low-birth-weight in Benin.

3-6. Drugs for preventing malaria in pregnant women living in malaria endemic areas

Malaria contributes to maternal illness and anemia in pregnancy, especially in first-time-mothers, and can harm the mother and neonate. Drugs given routinely to prevent or mitigate the effects of malaria during pregnancy are often recommended.

Garner and Gulmezoglu (24) assessed drugs given to prevent malarial infection and its consequences in pregnant women living in malarial areas. This includes prophylaxis and intermittent preventive treatment. Sixteen trials (12,638 participants were enrolled; two used adequate methods to conceal allocation were used). Antimalarials reduced antenatal parasitaemia when given to all pregnant women (relative risk 0.53, 95% confidence limits = 0.33 to 0.86; 328 participants, 2 trials), placental malaria (relative risk 0.34, 95% confidence limits 0.26 to 0.45; 1,236 participants, 3 trials), but no effect was detected with perinatal deaths (2,890 participants, 4 trials). In women in their first or second pregnancy, antimalarial-drugs reduced severe antenatal anemia (relative risk 0.62, 95%CI = 0.50 to 0.78; 2,809 participants, 1 prophylaxis and 2 intermittent preventive treatments), antenatal parasitaemia (relative risk 0.27, 95% CI = 0.17 to 0.44, random-effects model; 2,906 participants, 6 trials), and perinatal deaths (relative risk 0.73, 95%CI 0.53 to 0.99; 1,986 participants, 2 prophylaxis and 1 intermittent preventive treatment trial; mean birth-weight was higher (weighted mean difference 126.70 grams, 95% confidence limits 88.64 to 164.75 grams; 2,648 participants, 8 trials), and low-birth-weight less frequent (relative risk 0.57, 95%CI 0.46 to 0.72; 2,350 participants, 6 trials). Proguanil performed better than chloroquine in one trial of women of all parties in relation to maternal fever episodes. Sulfadoxine-pyrimethamine performed better than chloroquine in two trials of low-parity-women. Chemoprophylaxis or intermittent preventive treatment reduces antenatal parasite prevalence and placental malaria when given to women in all parity groups. They also have positive effects on birth-weight and possibly on perinatal death in low-parity-women.

Radeva-Petrova et al. (25) assessed the effects of malaria chemoprevention given to pregnant women living in malaria endemic areas on substantive maternal and infant health outcomes. Seventeen trials enrolling 14,481 pregnant women were carried out between 1957 and 2008, in Nigeria (3 trials), the Gambian (3 trials), Kenya (3 trials), Mozambique (2 trials), Uganda (2 trials), Cameroon (1 trial), Burkina Faso (1 trial), and Thailand (2 trials). Six different antimalarials were evaluated against placebo or no intervention; chloroquine (given weekly), pyrimethamine (given weekly or monthly), proguanil (given daily), pyrimethamine-dapsone (given weekly or fortnightly), and mefloquine (giving weekly). Trials recruited women in their first or second pregnancy (8 trials); only multigravid women (1 trial); or all women (8 trials).

Only six trials had adequate allocation concealment. For women in their first or second pregnancy, malaria chemoprevention reduces the risk of moderate to severe anemia by around 40%, and the risk of any anemia by around 17%. Malarial chemoprevention reduces the risk of antenatal parasitaemia by around 61%, and two trials reported a reduction in febrile illness. There were 16 maternal deaths. For infants of women in their first and second pregnancies, malaria chemoprevention probably increases mean-birth-weight by around 93 grams. Fever trails evaluated spontaneous abortions, still births, perinatal deaths, or neonatal deaths, and these analyzes were underpowered to detect clinically antenatal parasitaemia. In multigravid women, chemoprevention has similar effects on antenatal parasitaemia, but there are too few trials to evaluate effects on other outcomes. In trials giving chemoprevention to all pregnant women irrespective of parity, the average effects of chemoprevention measured in all women indicated it may prevent severe

anemia, but consistent benefits have not been shown for other outcomes. A single trial of prophylaxis against Plasmodium vivax showed chloroquine prevented vivax infection. Routine chemoprevention to prevent malaria and its consequences has been extensively tested with clinically important benefits on anemia and parasitaemia in the mother, and on birth-weight in infants. A total of 231 parturient mothers who gave birth to 232 neonates were included in the study. Ninety-five of them (41.1%) took antimalarial prophylaxis (chloroquine) in the index pregnancy, and 136 (58.9%) did not. Both groups were similar with respect to demographic characteristics except for the educational level of the mother, which was significantly higher in the group on prophylaxis (Chi-square = 8.05, p = 0.02) (26). The overall prevalence of maternal parasitaemia was 37.2%. The group on chloroquine experienced a lesser parasitaemia (26.3%) than the non-prophylactic group (44.9%, odds ratio (OR = 2.28), 95%CI = 1.24 - 4.19).

The proportion of women with severe parasitaemia (> 4000 parasites/μl) was also lower in the group of chloroquine than non-prophylactic group (17.6 versus 7.3%; OR = 2.69, 95%CI = 1.04-7.63). A modest reduction in low-birth-weight was found in the chloroquine group which was not significant (p = 0.16). In conclusion, chloroquine given to prevent malaria in pregnancy was found to be effective in reducing peripheral malaria parasitaemia, but improvement in birth-weight could not be demonstrated. Among other factors, impaired biological activity of the drug at the level of the placenta, where parasite sequestration frequently occurs, might be the explanation. Salihu et al. (26) recommend that further investigation be carried out in the study area to evaluate this finding, and if confirmed, institute appropriate changes in the present policy of chloroquine prophylaxis in pregnancy.

Few studies have documented the effectiveness in west-Africa of intermittent preventive treatment of malaria with sulfadoxine-pyrimethamine in pregnancy. Pregnant Nigerian women were assigned to receive either sulfadoxine-pyrimethamine given twice or presumptive chloroquine treatment followed by weekly pyrimethamine; 250 women were enrolled in each group (27). Of those completing follow-up, 4 (1.8%) in the sulfadoxine-pyrimethamine, and 22 (9.8%) in the chloroquine plus pyrimethamine groups had febrile illness (p = 0.005). None in the sulfadoxine-pyrimethamine group but 11 (4.9%) in the chloroquine plus pyrimethamine had peripheral parasitaemia prior or during delivery (p = 0.002). There were 6 low-birth-weight infants in the sulfadoxine-pyrimethamine group and 8 in the chloroquine plus Pyrimethamine group (p = 0.21). Intermittent preventive treatment with sulfadoxine-pyrimethamine is superior to chloroquine plus pyrimethamine for prevention of malaria and anemia in pregnant women in Nigeria.

Malaria contributes to maternal illness and anemia in pregnancy. Randomized and quasi-randomized controlled trials compared antimalarial-drugs given regularly with no antimalarial-drugs for preventing malaria in pregnant women living in malaria-endemic areas. Antimalarials reduced antenatal parasitaemia when given to all pregnant women, but no effect was detected with perinatal deaths (2,890 participants, 4 trials). In women in their first or second pregnancy, antimalarial-drugs reduced severe antenatal anemia. Mean-birth-weight was higher (weight mean difference 126.70 grams), and low-birth-weight was less frequent in 2,350 participants (8 trials). Proguanil performed better than chloroquine in 1 trial of women of all parties in relation to maternal fever episodes. Sulfadoxine-pyrimethamine

performed better than chloroquine in 2 trials of low-parity-women. Chemoprophylaxis or intermittent preventive treatment reduces antenatal parasite prevalence and placental malaria parasitaemia when given to women in all parity groups. They also have positive effects on birth-weight and possibly on perinatal death in low-parity-women.

In West-Africa, administration of chloroquine chemoprophylaxis during pregnancy is common, but little is known about its impact on Plasmodium falciparum infection during pregnancy. Therefore, cross-sectional studies in antenatal care clinics and during delivery units were conducted in Koupela District, Burkina Faso (28). Chloroquine chemoprophylaxis was reported by 69% of 597 pregnant women at antenatal care clinics and by 93% of 853 women in delivery units. Plasmodium falciparum peripheral parasitaemia was identified in 22% of delivering women and was strongly associated with low-birth-weight. Use of chemoprophylaxis was not associated with a reduction in the prevalence of placental parasitaemia, low-birth-weight, or prematurity. Despite the high reported chloroquine chemoprophylaxis coverage, peripheral and placental malaria rates remain high and are associated with known adverse outcomes during pregnancy, including maternal anemia, prematurity, and low-birth-weight. Alternative prevention strategies, such as use of insecticide-treated mosquito nets and intermittent preventive treatment with more-effective antimalarials, are needed. The WHO recommends intermittent preventive treatment in pregnancy with sulfadoxine-pyrimethamine in African regions with moderate to high malarial transmission. However, growing resistance to sulfadoxine-pyrimethamine threatens the effectiveness of intermittent preventive treatment in pregnancy where sulfadoxine-pyrimethamine, and alternative drugs are needed. Kimani et al. (29) tested the efficacy, tolerability, and safety of a fixed-dose combination of azithromycin-chloroquine. These authors administered 250 mg azithromycin and 155 mg chloroquine-base for intermittent preventive treatment in pregnancy relative to intermittent preventive treatment in pregnancy sulfadoxine-pyrimethamine. A randomized, phase 3, open-label, multi-center study was conducted in sub-Saharan Africa (Begin, Kenya, Malawi, Tanzania, and Uganda) between October 2010 and November 2013. Pregnant women received 3 intermittent preventive treatment courses with azithromycin-chloroquine (each course: 1,000/620 mg azithromycin/chloroquine for 3 days) or sulfadoxine-pyrimethamine (each course 1,500/75 mg sulfadoxine-pyrimethamine/chloroquine for 1 day) at 4- to 8 week intervals during the second and third trimester. Long-lasting insecticide-treated bed nets were also provided at enrollment. Study participants were followed-up until day 28 post-delivery (time window: day 28-42).

The primary endpoint was the proportion of participants with sub-optimal-pregnancy outcomes (a composite endpoint comprising live-born neonates with low-birth-weight < 2,500 grams, premature birth < 37 weeks, still birth > 28 weeks, abortion ≤ 28 weeks, lost to follow-up prior to observation of pregnancy outcome, or missing birth-weight). The study was terminated early after recruitment of 2,891 of the planned 5,044 participants, due to futility observed in a pre-specified 35% interim analysis. In the final intent-to-treat dataset, 378/1,445 (26.2%) participants in the azithromycin-chloroquine and 342/1,445 (23.7%) in the sulfadoxine-pyrimethamine group had sub-optimal pregnancy outcomes, with an estimated risk ratio of 1.11 (95%CI: 0.97, 125; p = 0.12). There was no significant difference

in the incidence of low-birth-weight between treatment groups (57/1,138 [5%] in the azithromycin-chloroquine group, 68/1188 [5.7%] in the sulfadoxine-pyrimethamine group, relative risk 0.87 (95% confidence limit: 0.62, 1.23; p = 0.44). Intermittent preventive treatment in pregnancy-azithromycin-chloroquine was less well-tolerated in mothers than intermittent preventive in pregnancy sulfadoxine-pyrimethamine. Occurrences of congenital anomalies, deaths, and serious adverse events were comparable in neonates for both groups. Limitations included the open-label-design and early study termination. Intermittent preventive treatment in the pregnancy azithromycin-chloroquine group was not superior to intermittent preventive treatment in the pregnancy sulfadoxine-pyrimethamine group in the present study. The proportions of sub-optimal-pregnancy outcomes and low-birth-weight were lower than expected, which may be linked to insecticide-treated bed net use throughout the study. Reduced incidence of symptomatic malaria infection and peripheral parasitaemia in the azithromycin-chloroquine group relative to the sulfadoxine-pyrimethamine group suggests that azithromycin-chloroquine warrants further investigation as an alternative treatment of uncomplicated malaria.

3-7. Effects of antimalarial-drugs on Plasmodium parasites

Saliba et al. (30) identified independent risk factors of severe falciparum malaria among travelers to endemic regions. A retrospective study was conduct on imported malaria into metropolitan France. The WHO severity criteria were used to classify malarial episodes. Nine hundred and twenty-one malarial cases were studied; 81 were severe. Independent risk factors of severe malaria were; aged above 40 years, high level of parasitized erythrocytes (more than 4%), parasite acquisition in the south-eastern Asian region, infection with chloroquine resistant Plasmodium falciparum phenotype and a self-administered antimalarial treatment. The present findings point out two particularly interesting results: severe malaria is significantly associated with infection by a chloroquine resistant Plasmodium falciparum phenotype and with the parasite's acquisition in the south-eastern Asian region.

3-8. Alternative antimalarial-drugs to chloroquine

Resistance to antimalarial chemotherapy is a major concern for malaria control in Vietnam. In a study undertaken in 1998, 65 patients with uncomplicated Plasmodium falciparum malaria were monitored for 28 days after completion of a 5-day treatment course with artemisinin. Overall 36.9% (24/65) of patients had recurrent parasitaemia during the surveillance period. Plasmodium falciparum isolates were tested for sensitivity in-vitro to chloroquine, mefloquine quinine, sulfadoxine-pyrimethamine and results were compared to those from a similar study in 1995. Increased parasite sensitivity to sulfadoxine-pyrimethamine, chloroquine, and quinine was demonstrated, with significantly lower mean EC_{50} and EC_{99} values in 1988 compared to 1995 (31).

Parasite sensitivity to mefloquine did not differ significantly in the 2 surveys. Isolates were also tested for sensitivity in-vitro to artemisinin in 1988 survey. The mean half maximal effective concentration (EC50) EC50 was 0.03 μmol/l and the EC99 was 0.94 μmol/l. Parasite sensitivity to artemisinin will need be monitored in view of its increasing use in Viet Nam.

The information needed to perform accurate analyses on antimalarial is not available. In the prophylaxis of malaria, chloroquine and proguanil have an excellent safety record, being very rarely

associated with severe adverse reactions in the recommended dosages. However, in many parts of the world they are no longer effective prophylactic agents (22). Pyrimethamine-dapsone is associated with agranulocytosis, especially if the recommended dose is exceeded, and should be reserved as a second-line-agent for travelers to high risk areas. Pyrimethamine-sulfadoxine and amodiaquine are associated with a relatively high incidence of potentially fatal reactions, and are no longer recommended for prophylaxis. Mefloquine, a relative newcomer, may provoke severe neuropsychiatric reactions with a frequency of 1 in 15,000 to 20,000 users at the prophylactic dosages. In the treatment of Plasmodium malaria, which has a high mortality rate if untreated, a greater risk of adverse reactions to malarial drugs is acceptable. As chloroquine resistance has become widespread, alternative agents including quinine, mefloquine-pyrimethamine-sulfadoxine, tetracycline, halofantrine and artemisinin and its derivatives may be used in treatment regimens. The therapeutic ratios for chloroquine, quinine and mefloquine are narrow and toxicity is frequent when recommended dosages are exceeded; parenteral administration above the recommended dose range is especially associated with cardiac and neurologic toxicity. Regular monitoring of the levels of anti-malarial resistance of Plasmodium falciparum is an essential policy to adapt therapy and improve malarial control (33).

This monitoring can be facilitated by using molecular tools, which are easier to implement than the classical determination of the resistance phenotype. In Cameroon, chloroquine, previously the first-line-therapy for uncomplicated malaria was officially withdrawn in 2002 and replaced initially by amodiaquine monotherapy. Then, artemisinin-lumefantrine was gradually introduced in 2004. This situation raised the question of the evolution of Plasmodium falciparum resistance molecular markers in Yaoundé, a highly urbanized Cameroonian city. The genotype of pfcrt 72 and 76 and pfmdr1 86 alleles and pfmdr1 copy number were determined using real-time PCR in 447 Plasmodium falciparum samples collected between 2005 and 2009. The present findings showed a high prevalence of parasites with mutant pfcrt 76 (83%) and pfmdr1 86 (93%) codons. On the contrary, no mutations in the pfcrt 72 codon and no samples with duplication of the pfmdr1 gene were observed. The high prevalence of mutant pfcrt 76T and pfmdr1 gene 86Y alleles might be due to the use of alternative drugs (amodiaquine and artesunate-amodiaquine) known to select such genotypes. Mutant pfcrt 72 codon was not detected despite the prolonged use of amodiaquine either as monotherapy or combined with artesunate. The absence of pfmdr1 multicopies suggests that artemether-lumefantrine would still remain efficient. The limited use of mefloquine or the predominance of mutant pfmdr1 86Y codon could explain the lack of pfmdr1 amplification. Indeed, this mutant codon is rarely associated with duplication of pfmdr1. In Cameroon, the changes of therapeutic strategies and the simultaneous use of several formulations of artemisinin-therapy or other anti-malarias that are not officially recommended result in a complex selective pressure, rendering the prediction of the evolution of Plasmodium falciparum resistance difficult. This public health problem should lead to increased vigilance and regular monitoring.

A distinct side-effect of synthetic quinolinic antimalarial-drugs, still widely used for the treatment and prophylaxis of malaria, is the introduction of psoriasis in predisposed or susceptible individuals (34). To describe two patients that had induction of psoriasis due to the administration of hydroxychloroquine, to

adapt pertinent literature on the pathophysiologic of this side-effect, to review psoriasis-triggered cases by newer, these patients were treated with hydroxychloroquine for a newly diagnosed lichen planopilaris and for an exacerbation of psoriatic arthritis, respectively. Psoriasis was controlled in both patients. The primigravidae gave birth to a healthy child at 39 weeks of gestation. The literature review returned no articles that linked the newer antimalarials with psoriasis. Despite the increased awareness, antimalarial-triggered psoriasis is still diagnosed. Fortunately, the current artemisinin-based antimalarial treatment can be safely offered to susceptible individuals. Additionally, prophylaxis with doxycycline or the combination atovaquone-proguanil could be a safe suggestion for malaria prophylaxis in psoriatic patients.

In 2001, Tanzania replaced chloroquine with sulfadoxine-pyrimethamine as first-line-drug, which in turn was replaced by artemisinin combination therapy in 2006. Sulphadoxine-pyrimethamine has however, continued to be used in intermittent preventive treatment of malaria in pregnancy despite reports of high levels of resistance to sulfadoxine-pyrimethamine due to the lack of an alternative to sulfadoxine-pyrimethamine for treatment of malaria in pregnancy (35). Recent reports have indicated recovery of chloroquine-susceptibility in Malawi, Kenya, Mozambique, and Tanzania based on the prevalence of wild types at codon 76 of the Pfcrt gene in indigenous Plasmodium falciparum populations. The current prevalence of this Pfcrt gene, particularly on codon 76 chloroquine resistance marker from 5 regions of Tanzania mainland is hereby reported. Seven hundred and forty one samples were genotyped. The current frequency of the chloroquine-susceptible Pfcrt-K76 was above 92% and did not differ between 85.7%, and 93% in regions. The chloroquine resistance recovery trends showed regional variability that may be caused by differences in malaria transmission intensity, but overall the trends converge as the susceptibility levels in all regions approach > 90%. Chloroquine withdrawal in Tanzania has resulted into > 90% recovery of susceptibility in ten years of withdrawal. These findings are in support of the search for chloroquine-based combination drugs as a possible future alternative to sulfadoxine-pyrimethamine for intermittent preventive treatment of malaria in pregnancy in places where full recovery of chloroquine-susceptibility will be evident.

Stratification of the type of anti-malarial drugs taken revealed that 37.0% used sulfadoxine-pyrimethamine, 32.0% artemisinin-based combined therapy, 11% antipyretics, 7.3% chloroquine, 7.1% quinine, 2.5% amodiaquine, while 3.0% used other drugs which were perceived as antimalarials (cough syrups and antibiotics) (36). In a regression model, it was demonstrated that age (p-level = 0.05), household size (p = 0.047), household head (p = 0.049), household source of income (p = 0.015), monthly income (p = 0.020), duration of use (p = 0.029), dosage of drugs taken (p = 0.036), and source of drugs (p = 0.005) significantly influenced anti-malarial drug use. Overall, 38.8% of responders used drugs as recommended by the Ministry of Health. These findings demonstrate that consumers require access to correct and comprehensible information associated with use of drugs, including self-prescription. There is a potential need by the Kenyan government to improve malaria care and decrease malaria-related morbidity and mortality by increasing drug affordability, ensuring that the recommended antimalarial drugs are easily available in all government approved drug

outlets and to educate the local shopkeepers on the symptoms and appropriate treatment of malaria.

3-9. Resistance of malaria Plasmodium parasites to antimalarial-drugs

The study included 45 cases of neonatal malaria. Thirty cases of malaria, admitted during first 10 days of life, diagnosed as congenital malaria, were kept in group A, while 15 cases admitted in the ward from 11 to 28 days, labeled as acquired malaria, were named group B (37). The clinical features at the time of presentation were noted in each group from the charts having positive malaria parasite on thick and thin slides. The diagnosed cases were treated with the standard dose of chloroquine sulphate. Those neonates who improved clinically as well as revealing no parasite on follow-up were labeled as chloroquine sensitive. On the other hand, neonates showing poor clinical response with persistence of the parasites in the blood or initially disappearing but later again having a clinical disease with positive malaria parasite on follow-up, were labeled as chloroquine resistant. They were treated with quinine sulphate.

Outcome was compared in both the groups regarding the pattern of chloroquine resistance and death/survival. Data were collected on Fischer's exact test, $p < 0.05$ was taken as statistically significant. Low-birth-weight, severe hemolytic anemia with history of fever in the mother were main features in group A while in group B fever, anemia and blood transfusion were marked features. In group A 76% were caused by Plasmodium falciparum, while in group B 60% were caused by Plasmodium vivax. In group A 26% were chloroquine resistant while 33.65% were chloroquine resistant in group B. The mortality rate was 16.66% in group A and 13.33% in group B. Intrauterine growth retardation, hemolytic jaundice and history of fever in the mother in the last trimester of pregnancy is the congenital while fever, history of blood transfusion in neonates in acquired malaria but pallor in both groups, were important clinical features. Pattern of chloroquine resistance and mortality in both the groups was not statistically different. Yusuf et al. (38) aimed to find out factors associated with anti-malarial-drug resistance in some selected areas in Ibadan. One thousand one hundred and two subjects were interviewed using a semi structured questionnaire. Responses were put into two subjects (high and low resistant areas). The results revealed a high level of drug use for treating malaria particularly chloroquine and sulfadoxine-pyrimethamine. The results also showed that the two groups were significantly different with respect to clearance of infection, but there was a significant difference between clearance of infection and whether or not the respondent completed the course of treatment in each group ($p < 0.05$).

When both groups were combined, the Mantel-Haenszel test showed that the response difference between the two groups was significant i.e. those that completed the treatment were 3 times more likely to have their infection cleared than those that did not complete the treatment. A significant finding was that non-compliance with treatment was a major factor associated with treatment failure. The prevalence of drug resistance was a little higher in the high resistant group compared to the low resistant group; but this difference was not statistically significant. These results underscore the need for adequate health education about the treatment of malaria and the importance of compliance in this community. Ethiopia is one of the few African countries where Plasmodium vivax is co-endemic with Plasmodium falciparum. Malaria transmission intensity varies mainly by landscape and climate. Although the recent emergence of drug resistant parasites presents a major issue to

malaria control in Ethiopia, little is known about the transmission pathways of parasite species and prevalence of resistant markers. Lo et al. (39) used microsatellites to determine population diversity and gene flow patterns of Plasmodium falciparum (n=226) and Plasmodium vivax (n= 205), as well as prevalence of drug resistant markers to infer the impact of gene flow and existing malaria treatment regimes. Plasmodium falciparum indicated a higher rate of polyclonal infections than Plasmodium vivax. Both species revealed moderate genetic diversity and similar population structure. Populations in the northern highlands were closely related to the eastern Rift Valley, but slightly distinct from the southern basin area. Gene flow via human migrations between the northern and eastern populations were frequent and mostly bidirectional. Landscape genetic analyses indicated that environmental heterogeneity and geographical distance did not constrain parasite gene flow.

This may partly explain similar patterns of resistant marker prevalence. In Plasmodium falciparum, a high prevalence of mutant alleles was detected in codons related to chloroquine (pfdhps and pfmdr1) resistance. Over 60% of the samples showed pfmdr1 duplications. Nevertheless, no mutation was detected in pfk13 that relates to artemisinin resistance. In Plasmodium vivax, while sequences of pvcrt-o were highly conserved and less than 5% of the samples showed pfmdr1 duplications, over 50% of the samples had pvmdr1 976F mutation. It remains to be tested if this mutation relates to chloroquine resistance. Monitoring the extent of malaria spread and markers of drug resistance is imperative to inform policy for evidence-based antimalarial choice and interventions. To effectively reduce malarial burden in Ethiopia, control efforts should focus on seasonal migrant populations. Asexual forms of Plasmodium falciparum which were present in the blood films of all patients before commencing treatment disappeared rapidly from the blood so that by the third day no parasites were seen in the blood film. The blood films remained negative for the rest of the seven-day observation period. Plasma chloroquine determination in eight of the patients showed high blood levels during the first three days. These results do not confirm the suspicion of chloroquine-resistant Plasmodium falciparum in the area studied although the level of resistance by WHO criteria was not excluded.

Malaria in pregnancy is one of the most common preventable causes of maternal and neonatal morbidity and mortality in sub-Saharan Africa. To prevent its adverse effects, such as maternal anemia, placental parasitaemia and low-birth-weight neonates, the WHO recommends effective malaria case management, use of insecticide-treated bed nets and intermittent preventive therapy in pregnancy (40). Sulphadoxine-pyrimethamine has been the standard for intermittent preventive therapy in pregnancy in several countries, but parasite resistance to sulfadoxine-pyrimethamine is growing. Therefore, new intermittent preventive therapy in pregnancy therapies are urgently needed. One candidate being evaluated for intermittent preventive therapy in pregnancy is a fixed-dose combination of azithromycin and chloroquine.

The azithromycin-chloroquine for intermittent preventive therapy in pregnancy pivotal trials is a multicentre, multicounty, phase III, open-label, randomized superiority study of sulphadoxine-pyrimethamine-chloroquine intermittent preventive therapy versus sulphadoxine-pyrimethamine-intermittent preventive therapy in pregnant women of sub-Saharan Africa. The trial was designed to meet stringent regulatory agency

scientific advice and intermittent preventive therapy in pregnancy policy makers' recommendations, and incorporates an innovative adaptive design to manage programmed risk, maintain the operating characteristics of the study and optimize resources. The trial's novel composite primary endpoint is the proportion of participants with a suboptimal pregnancy outcome (abortion [≤ 28 weeks], stillbirths [> 28 weeks], and premature [< 37 weeks] deliveries, low-birth-weight [< 2,500 grams] live neonates, missing neonatal-birth-weight data or loss to follow-up). The study employs a prospective group sequential design with three unblended analyses when 50%, 70%, and 100% of participants achieve the primary endpoint; the study team will remain blinded to the analyses until after the completion of the study.

The number of participants randomized will be adaptive, based on the blinded review of the observed pooled primary endpoint data across the two treatment arms, when approximately 1,000 participants complete the primary endpoint assessments. The present findings describe the unique challenges and innovative solutions implemented in designing this azithromycin-chloroquine trial, which may serve as a prototype for future intermittent preventive therapy in pregnancy trial, which may serve as a prototype for other studies involving similar conditions.

A case-control study was conducted to evaluate the efficacy of the combination of chloroquine plus proguanil as malaria prophylaxis in a non-immune population living in the Central-African-Republic (41). Cases were patients presenting with a malaria attack confirmed by a positive blood film and/or a histidine-rich protein 2 (HRP2) positive antigen test at the Pasteur Institute of Bangui. Two control subjects were included per case: one was a relative or close friend and the other was matched to the patient with respect to the length of stay. A questionnaire assessing malaria prophylaxis habits and malarial risk factors over the 2-months period prior to inclusion in the study was given to 48 cases and 96 controls. A conditional logistic regression was used to identify risk factors. The efficacy of the chloroquine plus proguanil regimen was found to be (95.5% and 95%, respectively, the confidence interval was 74.0 to 99.2%) in this country known for high chloroquine resistance. The present data lend some support to the use of chloroquine plus proguanil in Bangui, and the protective efficacy of chloroquine plus proguanil should now be studied prospectively as part of a randomized controlled trial of various prophylactic drugs.

Resistance of Plasmodium falciparum to chloroquine is widespread in Papua New Guinea. At a meeting in Port Moresby in October 1997, it was decided to explore a possible change of the current first-line-treatment of uncomplicated malaria with chloroquine alone (amodiaquine for children under 5 years) to chloroquine or amodiaquine in combination with sulfadoxine-pyrimethamine (42). To assess the therapeutic efficacy of the new drug combination in Papua New Guinea, a study was carried out in 1998-1999 at five hospital outpatient departments. From the 513 patients enrolled for the study, 95 defaulted from follow-up.

Of the remaining 418, 399 (95.5%) had an adequate clinical response. Out of the 19 patients who did not have an adequate clinical response, 3 (0.7% of the total) developed severe signs in the 24 hours and were treated in hospital; they were regarded as early treatment failures. The remaining 16 patients did not complete the study on the basis of various exclusion criteria but were not excluded from the analysis. From these results it was concluded that the combination was effective and a decision was taken in May 2000 to introduce combination regimens as

the standard first-line-treatment of uncomplicated malaria, including falciparum malaria, in Papua New Guinea.

One hundred and sixty-six children from Equatorial Guinea, all under 10 years of age and with acute uncomplicated falciparum malaria, were randomly allocated to four groups and treated with one of the following regimens: chloroquine or amodiaquine (25 mg/kg base over 3 days) quinine (8 mg/kg every 8 hours for 3 or 5 days), and sulphadoxine-pyrimethamine (25-1.25 mg/kg, in one dose) [43]. The parasite clearance rates up to 14 days were 28% with chloroquine, 74% with amodiaquine, and 95% with quinine or sulphadoxine-pyrimethamine. The times required to clear asexual blood forms of Plasmodium falciparum in sensitive cases were 64, 70, 73, and 65 hours, respectively. Although quinine and sulphadoxine-pyrimethamine are equally effective, quinine is recommended for treatment of multidrug-resistant malaria in pediatric patients, essentially because of the risk of serious reactions to sulpha drugs. Health providers are, however, encouraged to keep supplies of sulphadoxine-pyrimethamine as an option and to refer patients quickly, if required.

Over 12 years, from 1984 to 1995, Trape et al. (44) conducted a prospective study of overall malaria specific mortality among three rural populations in the Sahel, savanna and forest areas of Senegal. The emergence of chloroquine resistance has been associated with a dramatic increase in malaria mortality in each of the studied populations. After the emergence of chloroquine resistance, the risk of malaria death among children 0 to 9 years old in the three populations was multiplied by 2.1, 2.5, and 5.5, respectively. This is the first study to document malaria mortality at the community level in Africa before and after the emergence of chloroquine resistance. These findings suggest that the spread of chloroquine resistance has had a dramatic impact on the level of malaria mortality in most epidemiological contexts in tropical Africa. Multidrug-resistant Plasmodium vivax is widespread in eastern Indonesia, and has emerged elsewhere in Asia-Pacific and south-America, but is generally regarded as a benign disease. Tjitra et al. (45) reviewed the spectrum of disease associated with malaria due to Plasmodium vivax and Plasmodium falciparum in patients presenting to a hospital in Timika, Southern Papua, and Indonesia. Between January 2004 and December 2007, clinical malaria was present in 16% of hospital out-patients and 32% of in-patients. Among patients admitted with confirmed malaria, 64% of patients had plasmodium falciparum, 24% had Plasmodium vivax, and 10.5% had mixed infections.

The proportion of malaria admissions attributable to Plasmodium vivax rose to 47% in children under 1 year of age. Severe disease was present in 22% in patients with malaria, with the risk greater among Plasmodium vivax (23%) infections compared to Plasmodium falciparum (20%, $p= 0.001$), and greatest in patients with mixed infections (31%); (overall $p < 0.0001$). Severe anemia (hemoglobin < 5 grams/dl) was the major complication associated with Plasmodium vivax, accounting for 87% of severe disease compared to 73% of severe manifestations with Plasmodium falciparum ($p < 0.001$). Pure Plasmodium vivax infection was also present in 78 patients with respiratory distress and 42 patients with coma. In total, 2.0% patients with malaria died during admission: 2.2% with Plasmodium falciparum, 1.6% with Plasmodium vivax, and 2.3% with mixed infections ($p = 0.126$). In this region with established high-grade chloroquine resistance to both Plasmodium vivax and Plasmodium falciparum, Plasmodium vivax is associated with severe and fatal malaria particularly in young children. The

epidemiology of Plasmodium vivax needs to be re-examined elsewhere where chloroquine resistance is increasing. Frosch et al. (46) estimated drug use on a nation level, 2006-2007 Demographic Health Survey and Multiple Indicator Cluster Survey data from 21 African countries were analyzed. Resistance data were compiled by systematic review of the published literature on the prevalence of the Plasmodium chloroquine resistance transporter polymorphism at codon 76, which causes chloroquine resistance. Chloroquine was the most common anti-malarial used according to surveys from 14 of 21 countries analyzed, predominantly in west-Africa. Sulphadoxine-pyrimethamine was most commonly reported in two of 21 countries. Among eight countries with longitudinal molecular resistance data, the four countries where the highest proportion of children treated for fever received chloroquine (Uganda, Burkina Faso, Guinea Bissau, and Mali), also showed no significant declines in the prevalence of chloroquine-resistant infections.

The three countries with low or decreasing chloroquine use among children who reported fever treatment (Malawi, Kenya, and Tanzania) had statistically significant declines in the prevalence of chloroquine resistance. The present findings demonstrate that in 2006-2007, chloroquine and sulphadoxine-pyrimethamine continued to be used at high rates in many African countries. In countries reporting sustained chloroquine use, chloroquine-resistant malaria persists. In contrast, a low level of estimated chloroquine use is associated with a declining prevalence of chloroquine resistance. Infections with the malaria parasite Plasmodium falciparum typically comprise multiple strains, especially in high-transmission areas where infectious mosquito bites occur frequently. However, little is known about the dynamics of mixed-strain infections, particularly whether strains sharing a host complete or grow independently. Competition between drug-sensitive and drug-resistant strains could be a crucial determinant of the spread of resistance. Bushman et al. (47) analyzed 1,341 Plasmodium falciparum infections in children from Angola, Ghana, and Tanzania and found compelling evidence for competition in mixed-strain infections: overall parasite density did not increase with additional strains, and densities of individual chloroquine-sensitive and chloroquine-resistant strains were reduced in the presence of competitors. Bushman et al. (47) also found that chloroquine-resistant strains exhibited low densities compared with chloroquine-sensitive strains (in the absence of chloroquine), which may underlie observed declines of chloroquine resistance in many countries following the retirement of chloroquine as a first-line therapy. The present findings support a key role for within-host competition in the evolution of drug-resistant malaria. Malaria control and resistance-management efforts in high-transmission regions may be significantly aided or hindered by the effects of competition in mixed-strains infections. Consideration of within-host dynamics may spur development of novel strategies to minimize resistance while maximizing the benefits of control measures.

Plasmodium falciparum infection is an important cause of the high childhood mortality rate in sub-Saharan Africa. Increasingly, the contribution of Plasmodium falciparum-associated to severe anemia to pediatric mortality is being recognized while the impact of chloroquine resistance on mortality has not been evaluated. To address the issue of pediatric mortality, causes of death among hospitalized children less than 5 years of age in western Kenya were identified using standardized clinical examinations and

laboratory evaluations [48]. Follow-up examinations were conducted to determine the child's clinical status post-hospitalization. Of 1,223 children admitted to Siaya District Hospital from March to September 1991, 293 (24%) were severely anemic (hemoglobin level <5.0 grams/dl). There were 265 (22%) deaths; 121 (10%) occurred in-hospitalized and 144 (13%) occurred out-of-hospital within 8 weeks after admission; 32% of all deaths were associated with malaria. Treatment for malaria with chloroquine was associated with a 33% case fatality rate compared with 11% for children treated with more effective regimens (pyrimethamine/sulfa, quinine, or trimethoprim-sulfamethoxazole for 5 days). The risk of dying was associated with younger age ($p < 0.0001$) and severe anemia (relative risk = 1.52, confidence interval = 1.22, 1.90), and was decreased by treatment with an effective antimalarial drug (relative risk = 033, 95% confidence interval = 0.19, 0.65). Effective drug therapy for Plasmodium falciparum with regimens that are parastocidal in areas with a high prevalence of severe anemia and chloroquine resistance can significantly improve the survival of children in Africa.

Plasmodium falciparum infection is an important cause of the high childhood mortality rates in sub-Saharan Africa. Causes of death among hospitalized children less than 5 year old in western Kenya were identified using standardized clinical examinations and laboratory evaluations. Follow-up examinations were then conducted to determine the child's clinical status post-hospitalization. Of a total of 1,223 children, 293 admitted to Siaya District Hospital during March-September 1991 were severely anemic. A total of 265 children died; 32% of the deaths were associated with malaria. Deaths of 121 children occurred in-hospital and 144 out-of-hospital within 8 weeks after admission. The treatment of malaria with chloroquine was associated with a 33% case fatality rate compared with 11% in children less than 5 years old treated with more effective regimens such as pyrimethamine/sulfa, quinine or trimethoprim-sulfamethoxazole. The risk of dying was associated with younger age and severe anemia, and was decreased by treatment with an effective antimalarial drug. Al-Mekhlfi et al. (49) carried out a study to determine the prevalence of chloroquine resistance of Plasmodium falciparum isolates from Yemen based on the pfcrt T76 mutation. The prevalence of pfcrt T76 mutation was 81.5% (66 of 81 isolates). Coastal areas/foothills had a higher prevalence of pfcrt T76 mutation compared to highland areas (90.5% versus 71.8%, $p = 0.031$). Univariate analysis shows a significant association of pfcrt T76 mutation with people aged > 10 years ($p = 0.027$), no insecticide spray ($p = 0.025$), and not sleeping under insecticide treated nets ($p = 0.025$). Logistic regression models confirmed age > 10 years and low household income as predictors of pfcrt T76 mutation in Yemen Plasmodium falciparum isolates. The high prevalence of pfcrt T76 mutation in Yemen could be a predictive marker for the prevalence of Plasmodium falciparum chloroquine resistance and should be addressed in the national strategy to control malaria.

Although chloroquine monotherapy is now generally inadequate for the treatment of Plasmodium malaria in northern Ghana, (recently 58% of 225 children failed treatment by day 14) use of this drug continues because of its low cost and wide availability (50). The risk factors associated with chloroquine treatment failure in this region of Africa, including the T76 mutation in the chloroquine resistant transporter (pfcrt) gene and the Y86 mutation in the multidrug resistance (pfmdr1) gene of Plasmodium falciparum, have now been investigated, and genotype-

failure indices have been calculated. Treatment failure was found to be associated with young age, poor nutritional status, pfcrt T76 and pfmdr1 Y86, and early treatment failure was also associated with high parasitaemia. The pfcrt T76 genotype-failure indices for clinical and all treatment failures were 2.8 and 1.4, respectively. These indices were relatively low in the younger children, those with malnutrition, and those with high parasitaemia when treated. Residual chloroquine did not affect the genotype-failure indices substantially. Both pfcrt T76 and, to a lesser extent, pfmdr1 Y86 would be useful tools for the surveillance of chloroquine resistance in northern Ghana. In the current transition phase to the alternative first-line treatment for Plasmodium falciparum malaria, it should be possible to provide estimates of the level of chloroquine resistance by monitoring the prevalence of these mutations.

Plasmodium falciparum has developed resistance to almost all routinely used antimalarial drugs. Sulfadoxine-pyrimethamine has replaced chloroquine as first-line treatment of uncomplicated malaria infection in Kenya but resistance to sulfadoxine-pyrimethamine is already reported. The addition of artemisinin derivates to sulfadoxine-pyrimethamine may delay the development of drug resistance, improve cure rates, and reduce trans- mission of sulfadoxine-pyrimethamine (51). The efficacy and safety of artesunate plus sulfadoxine-pyrimethamine in the treatment of uncomplicated Plasmodium falciparum malaria was evaluated in a randomized trial of 600 children at Siaya District Hospital, western-Kenya between October 1999 and March 2000. Children aged < 5 years were randomly assigned to receive sulfadoxine-pyrimethamine alone (1.25 mg/kg based on pyrimethamine), or in combination with artesunate (4 mg/kg per day) for either 1 or 3 days. Treatment failure rates by day 14 were 25.5% in the sulfadoxine-pyrimethamine alone group, 16.2% (p = 0.027) in the 1-dose artesunate group, and 9.4% in the 3 dose of artesunate group (p < 0.001). Corresponding rates by day 28 were 46.0% in the sulfadoxine-pyrimethamine alone, 38.2% in 1-dose artesunate group (p-level = 0.16), and 26.0% in the 3-dose artesunate group (p < 0.001). The artesunate and sulfadoxine-pyrimethamine combination was well tolerated. There were no serious drug-related adverse events. Parasite clearance and gametocyte carriage were reduced significantly in both combination groups compared with sulfadoxine-pyrimethamine alone. Three days of artesunate were required to reduce significantly the risk of treatment failure by day 28. However, the high background rate of parasitological failure with sulfadoxine-pyrimethamine may make this combination unsuitable for widespread use in Kenya.

3-10. The need to find an international survey for establishing a therapy to combat the malaria infection

In Gambia, 760 children less than 10 years of age with Plasmodium falciparum malaria were treated with chloroquine (25 mg/kg), and followed-up 2 and 9 days after the start of treatment (52). A total of 700 children (92.1%) completed the study. The level of in-vivo resistance to chloroquine varied by area from 0.4% to 16.4%. Of the 28 children found to have chloroquine resistant malaria, none was ill when seen at 9 days follow-up and only 3 (10.3%) required further treatment with alternative malarial drugs because of persistent high levels of parasitaemia. The blood film at day 2 did not usefully predict resistance. A total of 67 isolates were tested in-vitro for chloroquine sensitivity. The mean EC_{50} was 15.5 nmol/l, an eight-fold decrease in sensitivity from that of isolates tested in 1982. A percentage of 11 (n = 8) of the isolates had EC_{50}s above the WHO

reference value for sensitive isolates of 18.3 nmol/l, with values ranging from 22 to 65 nmol/l of culture medium. Gambian isolates were sensitive to quinine (mean EC_{50} was 49.6 nmol/l). Response of Plasmodium falciparum to chloroquine treatment was assessed in-vivo in 219 malaria cases from 8 villages in a formerly hyperendemic area of Zimbabwe experiencing a malaria outbreak (53).

Seven (3%) of the cases were fully sensitive to chloroquine while 182 cases (85%) exhibited chloroquine-resistant response. Of the 182 chloroquine-resistant cases 74 (41%) showed RI resistance while 108 (59%) exhibited RII-RIII resistance. In-vivo follow-up was not completed to-day 28 in the remaining 30 (14%) of the malaria cases pyrexia and increasing parasitaemia occurred between day 3 and day 7 following treatment. Mean parasite clearance time was 5.8±2.89 days, in patients who were cleared of asexual parasitaemia. The present findings show an acute problem of chloroquine resistance in an area of Zimbabwe. It is recommended that the drug policy be modified to allow distribution of limited stocks of sulfadoxine-pyrimethamine to the local clinics for restricted use on documented chloroquine treatment failures within 7 days. Falciparum malaria is a significant problem for Afghanistan refugees in Pakistan. Refugee treatment guidelines recommended standard three-day chloroquine treatment (25 mg/kg) for first episodes and extended five-day treatment (40 mg/kg) for recrudescent infections, based on the assumption that a five-day course would more likely achieve a cure (54). An in-vivo randomized controlled trial was conducted among refugees with uncomplicated falciparum malaria to determine whether 5-day treatment of chloroquine 40 mg/kg was more effective than standard treatment of chloroquine of 25 mg/kg for 3 days. A total of 142 falciparum patients were recruited into 25 mg/kg or 40 mg/kg chloroquine treatment arms and followed-up to 60 days with regular blood smears. The primary outcome was parasitological cure without recrudescence. Treatment failures were retreated with 40 mg/kg chloroquine. Polymerase chain reaction (PCR) genotyping of 270 samples, from the same and nearby sites, was used to support interpretation of outcomes. A percentage of 84 of 25 mg/kg chloroquine versus 51% of chloroquine 40 mg/day patients experienced parasite recrudescence during follow-up. Cure rates were significantly improved with 40 mg/kg chloroquine, particularly among adults. Fever clearance time, parasite clearance time, and proportions gametocytaemic post-treatment were similar between treatment groups. Second-line-chloroquine 40 mg/kg treatment resulted in higher failure rates than first-line 40 mg/kg chloroquine treatment. Chloroquine-resistance marker pfcrt 76T was found in all isolates analyzed, while pfmdr1 86Y and 184Y were found in 18% and 37% of isolates, respectively. Chloroquine is not suitable for first-line-falciparum treatment in Afghan refugee communities. The extended-dose chloroquine regimen can overcome 39% of resistant infections that would recrudesce under the standard regimen, but the high failure rate of directly observed treatment demonstrates its use is inappropriate.

Chloroquine resistance was first documented in Uganda in 1988. Subsequent surveillance of antimalarial drug resistance, conducted by the Uganda Ministry of Health and several research organizations, suggested that resistance to chloroquine is now widespread, reaching critical levels in many areas of the country. In June 2000, the Ministry of Health held a National Consensus Meeting to evaluate the available drug efficacy data and review the national antimalarial drug policy. After extensive debate, the combination of

chloroquine plus sulfadoxine-pyrimethamine was chosen to replace chloroquine as the first-line treatment of uncomplicated malaria as an interim policy (55). This review evaluated the in-vivo drug efficacy studies conducted in Uganda since 1988 and issues confronted in revision of the drug policy. The Uganda experience illustrates the challenges faced by sub-Saharan African countries confronted with rising chloroquine resistance but limited data on potential alternative options. The choice of chloroquine plus sulfadoxine-pyrimethamine as a provisional policy in the absence of prerequisite efficacy, safety and cost-effectiveness data reflects the urgency of the malaria treatment problem, and growing pressure to adopt combination therapies. Surveillance of chloroquine plus sulfadoxine-pyrimethamine treatment efficacy, collection of additional data on alternative regimens and active consequences building among key partners in the malaria community will be necessary to develop a rational long-term antimalarial treatment policy in Uganda. There is the need to carry out an international survey to establish the prevalence of antimalarial drugs in malaria endemic areas.

4-DISCUSSION

Malaria is an infectious disease caused by 3 parasites namely: Plasmodium falciparum, Plasmodium vivax, and Plasmodium ovale. Plasmodium falciparum is the most common and virulent of malarial parasites. These parasites are present in different areas of sub-Saharan Africa countries and Asia. In 2010, there were an estimated 219 million cases of malaria resulting in 660,000 deaths, and about two-thirds were children. The children are more vulnerable than adults to malaria parasites. In sub-Saharan African countries, about 200,000 infants die yearly. Residents in endemic areas develop considerable immunity over time, but pregnancy increases the risk of anemia, miscarriage, stillbirth and prematurity (1).

Many theories for the mechanisms of the action of chloroquine have been advanced. Quinolines interfere with heme detoxification (3). Chloroquine concentrates in the highly acidic digestive vacuoles of Plasmodium parasites, where it binds to heme and disrupts its sequestration. Chloroquine-sensitive Plasmodium parasites concentrate the drug more than chloroquine-resistant organisms (6). Nyirjesy et al. (11) evaluated the incidence of malarial infection in mothers, placentas, and neonates and examined the effects of infection on birth-weight and perinatal mortality. Peripartum smears of maternal blood (21%), placentas (33%), and cord blood (9%) are effected by malaria. Neonatal and maternal malaria increase the risk of perinatal death, and low-birth-weight. Neonatal malaria increases the risk of perinatal death. Chemoprophylaxis with chloroquine during pregnancy may have a protective effect, even in certain areas where chloroquine-resistant Plasmodium falciparum is endemic.

More male (213) than female (169) children were admitted to the hospital for malaria treatment. Fever with convulsion (55.8%) was the commonest presenting symptoms, and anemia was the commonest presenting complication recorded. Chloroquine was found to be the most prescribed antimalarial drug and an overall artemisinin-based drug was prescribed either as first- or second-line treatment in only 18.2% of the cases. Chloroquine remains the primary drug for treatment of malaria in most sub-Saharan African countries (15). Zucker et al. (15) evaluated the effects of drug treatment policy on the case-fatality-rates of children for malaria and severe anemia. In 1991, 63% of children were treated with chloroquine while the remaining 37% were treated with a regimen that would

eliminate and clear parasitaemia. Case-fatality rates were 13% and 4.1%, respectively; the proportion of deaths attributed to chloroquine was 69%. The trend in case-fatality rates for malaria decreased as an increasing proportion of children received an effective treatment regimen. There is broad evidence for the adverse effects of Plasmodium falciparum in pregnancy, and the WHO recommends preventive strategies. There is a markedly reduced efficacy in sub-Saharan Africa in on the most widely available affordable and used antimalarial drug for chemoprophylaxis-chloroquine. Steketee et al. (18) studied pregnant women in an area of high malaria endemic in rural Malawi to compare the efficacy of chloroquine (the drug recommended by national policy of WHO) with mefloquine (a relatively new and high effective antimalarial drug); 1,766 women monitored during at least six weeks of their pregnancy were treated with chloroquine, and their neonates had a birth-weight of 2,905±461 grams and 18.6% had a lower-body-weight. Factors significantly associated with intrauterine grown retardation and low-birth-weight included maternal syphilis infection and umbilical cord blood malaria. Use of the effective antimalarial mefloquine was protective against low-birth-weight through its effect on reducing placental and umbilical cord blood malaria infection.

The proportion of low-birth-weight neonates born to women on mefloquine was significantly lower than the proportion born to women on chloroquine. Effective prevention of malaria in pregnant women in malaria-endemic-setting may reduce the likelihood of low-birth-weight by 5 to 14% and may reduce the amount of preventable low-birth-weight more than 30%. Barsalou et al. (19) assessed if maternal intake of antimalarials throughout pregnancy lowers the risk of cardiac and non-cardiac neonatal lupus. A total of 268 pregnancies were included; 73 were exposed to antimalarials throughout pregnancy. Ninety-nine children developed neonatal lupus, 117 remained unaffected and 52 children did not develop cardiac neonatal lupus but could not be categorized as unaffected since their full non-cardiac lupus was unknown. A randomized, double-blind, placebo-controlled trial, which compared the effects of three interventions (weekly chloroquine prophylaxis, dial iron and weekly folic acid supplementation, and case management of malaria) on congenital malaria, maternal hemoglobin and fetal outcome, was conducted among primigravidae resident in Uganda (20). Among 473 neonates examined at birth or within 7 days of birth, 198 (42%) were parasitaemic. However, 33 (17%) of the parasitaemic neonates were born to mothers who had placental but not peripheral parasitaemia. A total of 22 (11%) were born to mothers who had peripheral but not placental parasitaemia, and 12 (6%) were born to mothers with neither peripheral nor placental parasitaemia. Parasitaemia at birth was not significantly associated with low-birth-weight, in any of three intervention groups. Congenital malaria per se have little influence on birth-weight but may have an impact on anemia.

Henry et al. (21) studied the parasitological, clinical, and hematological response to chloroquine treatment in children during a 28-day follow-up in an intermediate chloroquine resistance. While chloroquine treatment still produced clinical remission at day 7 in 95% of the children, 35% had recurred fever with concomitant parasitaemia before day 28. All fever cases subsequently with Fansidar remained parasite-negative over a period of 28 days. The rate of resistance to chloroquine was most pronounced among the children aged less than 5 years, 8% of whom showed clinical failure by day 14.

The durability of response to chloroquine treatment should be at least 14 days in young children living in intermediate chloroquine resistance. Anti-malarial drug resistance to chloroquine and sulfadoxine-pyrimethamine has spread from Southeast Asia to Africa. The recent emergence of resistance to artemisinin-based-combination-therapy in Southeast Asia highlights the need to identify new antimalarial-drug (22). Doxycycline is recommended for malaria chemoprophylaxis for travel in endemic areas, or in combination with the use of quinine when artemisinin-base-combination therapy is unavailable. However, doxycycline is not used in young children under 8 years of age. In United States, the centers for Disease Control and Prevention have recommended the use of doxycycline for the treatment of acute and chronic chloroquine fever and tick-borne rickettsial diseases in young children.

Among 1,090 singleton live births, prevalence of placental malaria and low-birth-weight were 16% and 17%, respectively (23). There was non-significant-association between placental malaria. Protection from low-birth-weight was high in women reporting regular chloroquine prophylaxis, with a strong duration-effect relationship ($p < 0.001$). Despite high parasite resistance and limited effect on placental malaria, a chloroquine chemoprophylaxis at adequate doses showed to be still effective in reducing low-birth-weight in Benin. Malaria contributes to maternal illness and anemia in pregnancy, especially in first-time-mothers, and can harm the mother and neonate. Drugs given routinely to prevent or mitigate the effects of malaria during pregnancy are often recommended. Garner and Gulmezoglu (24) assessed drugs given to prevent malaria infection and its consequences in pregnant women living in malarial areas. In women in their first- or-second pregnancy, antimalarial-drugs reduced severe antenatal anemia. Proguanil performed better than chloroquine in one trial of women of all parties in relation to maternal fever episodes. Sulfadoxine-pyrimethamine performed better than chloroquine in two trials of low-parity-women. Chemoprophylaxis or intermittent-preventive-treatment reduces antenatal parasite prevalence and placental malaria when given to women in all parity-groups. They also have positive effects on birth-weight and possibly on perinatal-death in low-parity-women.

Radeva-Petrova et al. (25) assessed the effects of malaria chemoprevention given to pregnant women living in malaria endemic areas on substantive maternal and infant outcomes. Six different antimalarials were evaluated against placebo or no intervention; chloroquine (given weekly), pyrimethamine (giving weekly or monthly), proguanil (giving daily) pyrimethamine-dapsone (giving weekly or fortnightly), and mefloquine (giving weekly). Trials recruited women in their first- or-second pregnancy. Malaria chemoprevention reduces the risk of moderate to severe anemia by around 40%, and the risk of any anemia by around 17%. Malarial chemoprevention reduces the risk of antenatal parasitaemia by around 61%. There were 16 maternal deaths.

Fever trails evaluated spontaneous abortions, still births, perinatal deaths, or neonatal deaths, and these analyzes were underpowered to detect clinically antenatal parasitaemia. In trials giving chemoprevention to all pregnant women irrespective of parity, the average effects of chemoprevention measured in all women indicated it may prevent severe anemia, but consistent benefits have not been shown for other outcomes. A single trial of prophylaxis against Plasmodium vivax showed that chloroquine prevented vivax infection. Routine chemoprevention

to prevent malaria and its consequences has been extensively tested with clinically important benefits on anemia and parasitaemia in the mother, and on birth-weight in infants. A total of 231 parturient mothers who gave birth to 232 neonates were included in the study (26). A percentage of 41.1 took chloroquine and 58.9% did not. The group on chloroquine experienced a lesser parasitaemia (26.3%) than the non-prophylactic-group (44.9%). The proportion of women with severe parasitemia (> 4,000 parasites/μl) was also lower in the group of chloroquine than non-prophylactic-group. Chloroquine given to prevent malaria in pregnancy was found to be effective in reducing peripheral malaria parasitaemia, but improvement in birth-weight could not be demonstrated.

Pregnant Nigerian women were assigned to receive either sulfadoxine-pyrimethamine given twice or presumptive chloroquine treatment followed by weekly pyrimethamine (27). A perceptual of 1.8 in the sulfadoxine-pyrimethamine and 9.8% in the chloroquine plus pyrimethamine groups had febrile illness ($p = 0.005$). None in the sulfadoxine-pyrimethamine group but 11 (4.9%) in the chloroquine plus pyrimethamine had peripheral parasitaemia prior or during delivery ($p = 0.002$). Intermittent preventive treatment with sulfadoxine-pyrimethamine is superior to chloroquine plus pyrimethamine for prevention of malaria and anemia in Nigerian pregnant women.

Malaria contributes to maternal illness and anemia in pregnancy. A total of 12,638 pregnant women were enrolled. Randomized and quasi-randomized controlled trials compared antimalarial-drug given regularly with no antimalarial drugs for preventing malaria in pregnant women living in malarial-endemic areas (24). Antimalarial reduced antenatal parasitaemia when given to all pregnant women, but no effect was detected with perinatal deaths. In women in their first- or second-pregnancy, antimalarial-drugs reduced severe antenatal anemia. Chemoprophylaxis or intermittent preventive treatment reduces the antenatal parasite prevalence and placental malaria parasitaemia when given to women in all parity-groups. They also have positive effects on birth-weight and possibly on perinatal death in low-parity-women. Cross-sectional studies in antenatal care clinics and during delivery units were conduct in Koupela District, Burkina Faso (28). Chloroquine chemoprophylaxis was reported by 69% of 597 pregnant women at antenatal care clinics and by 93% of 853 women in delivery units. Plasmodium falciparum peripheral parasitaemia was identified in 22% of delivering women and was strongly associated with low-birth-weight, or prematurity. Despite the high reported chloroquine chemoprophylaxis coverage, peripheral and placental malaria rates remain high and are associated with known adverse outcomes during pregnancy, including maternal anemia, prematurity, and low-birth-weight.

The WHO recommends intermittent preventive treatment in pregnancy with sulfadoxine-pyrimethamine in African regions with moderate to high malarial transmission. However, growing resistance to sulfadoxine-pyrimethamine threatens the effectiveness of intermittent preventive treatment in pregnancy with sulfadoxine-pyrimethamine, alternative drugs are needed. Kimani et al. (29) tested the efficacy, tolerability, and safety of a fixed-dose combination of azithromycin-chloroquine. The doses of these drugs were 250 mg and 155 mg, respectively. A randomized, phase 3, open-label, multi-center study was conducted in Begin, Kenya, Malawi, Tanzania, and Uganda. Pregnant women received 3 intermittent preventive courses with azithromycin-chloroquine for 3 days, or sulfadoxine-pyrimethamine for 1 day at 4-8 week

intervals during the second and third trimester. Long-lasting insecticide-treated bed nets were also provided at enrollment. The primary endpoint was a composite endpoint comprising live-born neonates with low birth-weight < 2,500 grams, premature birth < 37 weeks, still birth > 28 weeks, abortion ≤ 28 weeks, lost to follow-up prior to observation of pregnancy outcome, or missing birth-weight. In the final intent-to-treat-dataset, participants in the azithromycin-chloroquine were 26.2% and the participants in the sulfadoxine-pyrimethamine group were 23.7%. Intermittent preventive treatment in pregnant azithromycin-chloroquine was less well-tolerated in mothers than intermittent preventive in pregnancy-sulfadoxine-pyrimethamine.

The information needed to perform accurate analyses on antimicrobial is not available. In the prophylaxis of malaria, chloroquine and proguanil have an excellent safety record, being very rarely associated with severe adverse reactions in the recommended dosages. However, in many parts of the world they are no longer effective prophylactic agents (32). Pyrimethamine-dapsone is associated with agranulocytosis, especially if the recommended dose is exceeded, and should be reserved as a second-line-agent for travelers to high risk areas. Pyrimethamine-sulfadoxine and amodiaquine are associated with a relatively high incidence of potentially fatal reactions. Mefloquine may provoke severe neuropsychiatric reaction with a frequency of 1 in 15,000 to 20,000 users. In the treatment of Plasmodium malaria, which has a high mortality rate if untreated, a greater risk of adverse reactions to malarial drugs is acceptable. As chloroquine resistance has become widespread, alternative drugs including quinine, mefloquine, pyrimethamine-sulfadoxine, tetracycline, halofantrine, and artemisinin may be used in treatment regimens. Regular monitoring of the levels of antimalarial resistance of Plasmodium falciparum is an essential policy to adapt therapy and improve malarial control (33). In Cameroon, chloroquine, previously the first-line-therapy for uncomplicated malaria was officially withdrawn in 2002 and replaced initially by amodiaquine monotherapy. Then, artemisinin-lumefantrine was gradually introduced in 2004. In 2001, Tanzania replaced chloroquine with sulfadoxine-pyrimethamine as the first-line-drug, which in turn was replaced by an artemisinin combination in 2006. Sulphadoxine-pyrimethamine has however, continued to be used in intermittent preventive treatment of malaria in pregnancy, despite reports of high levels of resistance to sulphadoxine-pyrimethamine (35).

Recent reports have indicated recovery of chloroquine-susceptibility in Malawi, Kenya, Mozambique, and Tanzania based on prevalence of wild types at codon 76 of the Pfcrt gene in indigenous Plasmodium falciparum populations. Chloroquine withdrawal in Tanzania has resulted > 90% recovery of susceptibility in ten years of withdrawal. These findings are in support of the search for chloroquine-based combination drugs as a possible future alternative to sulphadoxine-pyrimethamine for intermittent preventive treatment of malaria in pregnancy in places where full recovery of chloroquine-susceptibility will be evident.

Stratification of the type of anti-malarial drugs taken revealed that 37.0% used sulphadoxine- pyrimethamine, 32.0% artemisinin-based combined therapy, 11% antipyretics, 7.3% chloroquine, 7.1% quinine, 2.5% amodiaquine, while 3.0% used drugs which were perceived as anti-malarial (cough syrups and antibiotics) (36). There is the need to carry out an international survey to establish the prevalence of antimalarial drugs. Malaria

in pregnancy is one of the most common preventable causes of maternal and neonatal morbidity and mortality in sub-Saharan Africa. To prevent its adverse effects, such as maternal anemia, placental parasitaemia and low birth-weight-neonates, the WHO recommends effective malaria case management, use of insecticide-treated bed-nets and intermittent preventive therapy in pregnancy (40). Sulphadoxine-pyrimethamine is the standard for intermittent preventive therapy in pregnancy in several countries, but parasite resistance to sulphadoxine-pyrimethamine is growing. Therefore, new intermittent preventive therapy in pregnancy therapies are urgently needed. One candidate being evaluated for intermittent preventive therapy in pregnancy is a fixed-dose-combination of azithromycin and chloroquine.

The azithromycin-chloroquine for intermittent preventive therapy in a pregnancy pivotal trial is a multi-country, phase III, open-label, randomized superiority study of a sulphadoxine-pyrimethamine-chloroquine intermittent preventive therapy versus sulphadoxine-pyrimethamine-intermittent preventive therapy in pregnant women of sub-Saharan Africa. The trial novel composite primary endpoint is the proportion of participants with a suboptimal pregnancy outcome (abortion [≤ 28 weeks], stillbirths [> 28 weeks], and premature [< 37 weeks] deliveries, low-birth-weight [< 2,500 grams] live neonates, missing neonatal birth-weight data or loss to follow-up). The study employs a prospective group sequential design with three unblended analyses when 50%, 70%, and 100% of participants achieve the primary endpoint. The study team will remain blinded to the analyses until after the completion of the study. The present findings describe the unique challenges and innovative solutions implemented in designing this azithromycin-chloroquine trial, which may serve as a prototype for future intermittent preventive therapy in pregnancy trials and other studies involving similar conditions. Resistance of plasmodium falciparum to chloroquine is widespread in Papua New Guinea. In 1997, it was decided to explore a possible change of the current first-line-treatment of uncomplicated malaria with chloroquine alone (amodiaquine for children under 5 years of age) to chloroquine or amodiaquine in combination with sulfadoxine-pyrimethamine (42). To assess the therapeutic efficacy of the new drug combination in Papua New Guinea, a study was carried out in 1998-1999 at five hospital outpatient departments. A total of 399 had an adequate clinical response. Out of the 19 patients who did not have an adequate clinical, 3 (0.7% of the total) developed severe signs in the 24 hours and were treated in hospital; they were regarded as early treatment failures. From these results it was concluded that the combination was effective and in 2000 it was decided to introduce combination regimens as the standard first-line-treatment.

A total of 166 children aged under 10 years with acute uncomplicated falciparum malaria living in Equatorial Guinea, were allocated to four groups and treated with one of the following regimens: chloroquine or amodiaquine (25 mg/kg over 3 days) quinine (8 mg/kg every 8 hours for 3 or 5 days, and sulphadoxine-pyrimethamine (25-1.25 mg/kg, in one dose [43]. The parasite clearance rates up to 14 days were 28% with chloroquine, 74% with amodiaquine, and 95% with quinine or sulphdoxine-pyrimethamine. The times required to clear asexual blood forms of Plasmodium falciparum in sensitive cases were 64, 70, 73, and 65 hours, respectively. Quinine is recommended for treatment of multidrug-resistant malaria in pediatric patients.

Trape et al. (44) conducted a prospective study of overall malaria mortality among rural population in the Sahel, savanna and forest areas of Senegal. The emergence of chloroquine resistance has been associated with a dramatic increase in malarial mortality in the populations studied. These findings suggest that the spread of chloroquine resistance has a dramatic impact on the level of malaria in low-birth-weights in most epidemiological contexts in tropical Africa. Multidrug-resistant Plasmodium vivax is widespread in Eastern Indonesia, and has emerged elsewhere in Asia-Pacific and south-America. Tjitra et al. (45) reviewed the spectrum of disease associated with malaria due to Plasmodium vivax and Plasmodium falciparum in patients presenting to a hospital in Timika, southern Papua, Indonesia. Among patients admitted with confirmed malaria, 64% of patients had plasmodium falciparum, 24% had Plasmodium vivax, and 10.5% had mixed infections. Severe anemia (hemoglobin < 5 grams/dl) was the major complication associated with Plasmodium vivax, accounting for 87% of severe diseases compared to 73% of severe manifestations with Plasmodium falciparum ($p < 0.001$). In this region with established high-grade chloroquine resistance to both Plasmodium vivax and Plasmodium falciparum, Plasmodium vivax is associated with severe and fatal malaria particularly in young children.

Chloroquine was the most common anti-malarial used according to surveys from 14 of 21 countries analyzed, predominantly in west-Africa. Sulphadoxine-pyrimethamine was most commonly reported in two of 21 countries. Uganda, Burkina Faso, Guinea Bissau, and Mali showed no significant declines in the prevalence of chloroquine-resistant infection. Malawi, Kenya, and Tanzania had statistically significant declines in the prevalence of chloroquine resistance. Chloroquine and sulphadoxine-pyrimethamine continued to be used at high rates in many African-countries. Competition between drug-sensitive and drug-resistant strains could be a crucial determinant of the spread of resistance. Bushman et al. (47) analyzed 1,341 children with Plasmodium falciparum living in Angola, Ghana, and Tanzania. These authors found compelling evidence for competition in mixed-strain infections. Overall parasite density did not increase with additional strains, and densities of individual chloroquine-sensitive and chloroquine-resistant were reduced in the presence of competitors. These authors also found that chloroquine-sensitive strains (in the absence of chloroquine), underline observed declines of chloroquine resistance in many countries following the retirement of chloroquine as a first-line-therapy. Malaria control and resistance-management efforts in high-transmission regions may be significantly aided or hindered by the effects of competition in mixed-strains infections.

Plasmodium falciparum is an important cause of childhood mortality in sub-Saharan Africa. Zucker et al. (48) investigated the cause of death among hospitalized children less than 5 years of age in western Kenya. Of the 1,223 children admitted to Siaya District Hospital were severely anemic (hemoglobin level < 0.5 grams/dl), 265 (22%) death and 32% of all deaths were associated with malaria.

Treatment with chloroquine caused a 33% case-fatality rate compared with 11% for children treated with pyrimethamine/sulfa, quinine, or trimethoprim-sulfamethoxazole for younger children ($p < 0.001$). The follow-up examinations were conducted to determine the child's clinical status post-hospitalized. A total of 293 admitted to the hospital were severely anemic and 265 children died; 32% of the deaths were associated with malaria. The risk of dying increased with a younger age and severe

anemia, and decreased with treatment using effective antimalarial drug.

5- CONCLUSION

In conclusion, malaria is an infection caused by three parasites namely: Plasmodium falciparum, Plasmodium vivax, and Plasmodium oval. Plasmodium falciparum is the most common and virulent malaria parasite. These parasites are present in different areas of the sub-Saharan Africa countries and Asia. In 2010, there were an estimated 219 million cases of malaria resulting in 660,000 deaths and, approximately, two-thirds were children. Children are more vulnerable than adults to malaria infection. In sub-Saharan African countries, maternal malaria is associated with up 200,000 estimated infant deaths yearly. Malaria contributes to maternal illness and anemia in pregnancy, especially in first-time-mothers, and chemoprevention reduces the risk of antenatal parasitaemia by 61%.

Malaria in pregnancy is one of the most common preventable causes of maternal and neonatal morbidity and mortality. Fever trials provoked spontaneous abortions, still births, perinatal deaths, or neonatal deaths. Malaria contributes to maternal illness and anemia in pregnancy and the average effects of chemoprevention measured in all women may prevent severe anemia. Chloroquine was the world's widely used antimalarial drug, but Plasmodium falciparum is now increasingly resistant. However, Plasmodium vivax and Plasmodium ovale are sensitive to chloroquine. Chloroquine-sensitive-Plasmodium parasites concentrate the drugs to higher level than did chloroquine-resistant parasites. There is the need to find an alternative antimalarial drug to chloroquine, and proguanil, mefloquine, sulfadoxine-pyrimethamine, amodiaquine, and artemisinin-lumefantrine are found active against Plasmodium falciparum. In Cameroon, chloroquine, previously the first-line-therapy for malaria, was officially withdrawn in 2002, and was replaced initially by amodiaquine monotherapy, then, artemisinin-lumefantrine was gradually introduced in 2004. In 2001, Tanzania replaced chloroquine with sulphadoxine-pyrimethamine as the first-line-drug, which in turn was replaced by an artemisinin combination therapy in 2006. There is to carry find out an international survey regarding for the prevalence of antimalarial-drugs in areas where malaria is endemic.

6- CONFLICT OF INTERESTS: None.

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