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Pharmacokinetics of Amodiaquine and Desethylamodiaquine in Pregnant and Postpartum Women with *Plasmodium vivax* Malaria▽†

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In order to study the pharmacokinetic properties of amodiaquine and desethylamodiaquine during pregnancy, 24 pregnant women in the second and third trimesters of pregnancy and with *Plasmodium vivax* malaria were treated with amodiaquine (10 mg/kg of body weight/day) for 3 days. The same women were studied again at 3 months postpartum. Plasma was analyzed for amodiaquine and desethylamodiaquine by use of a liquid chromatography-tandem mass spectrometry method. Individual concentration-time data were evaluated using noncompartmental analysis. There were no clinically relevant differences in the pharmacokinetics of amodiaquine and desethylamodiaquine between pregnant ($n = 24$) and postpartum ($n = 18$) women. The results suggest that the current amodiaquine dosing regimen is adequate for the treatment of *P. vivax* infections during pregnancy.

Amodiaquine (AQ) is a 4-aminoquinoline with a structure similar to that of chloroquine (CHQ). Both AQ and its principal biologically active metabolite, desethylamodiaquine (DEAQ), have antimalarial properties, but DEAQ is eliminated much more slowly than AQ and is therefore the main agent responsible for treatment efficacy. AQ is generally more effective than CHQ against chloroquine-resistant *Plasmodium falciparum* and *P. vivax* infections, and it has been used both as treatment for symptomatic malaria and intermittent preventive treatment (IPT) during pregnancy (3, 4, 11, 12, 20, 22). In Southeast Asia, amodiaquine is no longer effective for falciparum malaria but may be used increasingly if chloroquine resistance in *P. vivax* spreads (1). According to the World Health Organization, there is no evidence to contraindicate the use of amodiaquine during pregnancy, although data are limited and additional safety data are needed (2, 23, 25). The pharmacokinetic properties of AQ and DEAQ have been described for children and adults (5, 7, 13, 16, 19, 21, 29) but not for pregnancy. The pharmacokinetic properties of many antimalarials are altered during pregnancy (27). Ideally, drug regimens for pregnant women should be recommended on the basis of pharmacokinetic and pharmacodynamic studies to maximize efficacy (28). We report the pharmacokinetics of AQ and its principal biologically active metabolite, DEAQ, in the treatment of *P. vivax* infections in 24 pregnant women. The pharmacokinetic parameters during 42 days posttreatment are compared with those measured in the same women 3 months after delivery.

MATERIALS AND METHODS

Antenatal clinics. The study was carried out in two antenatal clinics of the Shoklo Malaria Research Unit (SMRU). These clinics are located on the northwestern border of Thailand, an area of malaria endemicity where transmission is low and seasonal for *P. falciparum* and *P. vivax*. The majority of the people in this region belong to the Karen ethnic group. All women have a dating ultrasound scan at their first antenatal clinic attendance (18) and are invited to attend weekly consultations providing early detection and treatment of all malaria episodes (14). Women routinely receive ferrous sulfate and folic acid supplementation from the first consultation until delivery. Pregnant women with a viable pregnancy in the second or third trimester and with *P. vivax* monoinfection (minimum parasitemia of >80/μl), a field sample hematocrit of >25%, and willingness to return for sampling at 3 months postpartum were eligible for inclusion. Before enrolment, the purpose of the study was explained in the patient's own language, and written consent was obtained (by thumbprint if she was unable to read or write).

Ethics. Approval of the study was obtained from the ethics committee of the Faculty of Tropical Medicine, Mahidol University, Bangkok, Thailand (MUTM 2007-112), and from the Oxford Tropical Research Ethics Committee (OxTREC 024-06).

Amodiaquine dosing regimen. The patients were treated with amodiaquine tablets (10 mg/kg of body weight/day) (Flavoquine; Aventis, France) by directly observed dosing with water at exactly 24-h intervals for 3 doses, i.e., hour zero (H0), H24, and H48. The number of tablets was calculated from the actual weight of the (pregnant) woman, and the tablets were divided (to the nearest quarter) if necessary.

Sampling regimen. Blood samples (2 ml) were obtained by venous puncture and taken into lithium heparin tubes at baseline (H0; before the first dose), H4, H24 (before the second dose), H28, and H48 (before the third dose). A catheter was then inserted into a vein, from which blood was drawn at H48.5, H49, H50, H51, H52, H54, H56, H58, and H72. The catheter was removed, and additional samples were taken at day 4 (D4), D5, D7, D14, D21, D28, D35, and D42. Blood samples were centrifuged at 1,500 to 2,000 × *g* at room temperature for 10 min to obtain plasmas. Immediately after centrifugation, the plasmas were transferred to screw-cap cryovials and frozen at −20°C in a laboratory freezer. The

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sampling and freezer times were recorded and a note made in case of visible hemolysis. Within 2 months, the frozen plasma samples were transferred to a −80°C freezer before analysis. The samples were shipped to the Service de Pharmacologie Clinique, Hôpital St. Vincent de Paul, Paris, France, on dry ice.

Drug analysis. AQ and DEAQ were analyzed using protein precipitation with acetonitrile and quantification by liquid chromatography-tandem mass spectrometry (LC-MS/MS) (6). Hydroxychloroquine was used as an internal standard. AQ and DEAQ were quantified using a TSQ Discovery Max triple-quadrupole mass spectrometer (Thermo Finnigan) operated in the positive ion mode. Quantification was performed using selected reaction monitoring (SRM) for the transitions *m/z* 356 to 283 and 328 to 283 for AQ and DEAQ, respectively, and *m/z* 336 to 247 for the internal standard hydroxychloroquine. Total assay coefficients of variation (CV) for AQ and DEAQ during analysis were <15% at all quality control levels. The lower limits of quantification were 1 and 2 ng/ml for AQ and DEAQ, respectively.

Pharmacokinetic analysis. Individual concentration-time data were evaluated using a noncompartmental analysis (NCA) approach (WinNonlin, version 5.0; Pharsight Corporation, CA). Total exposure up to the last measured concentration (area under the concentration-time curve from time zero to the time of the last measured concentration [AUC_{0-LAST}]) was calculated using the linear trapezoidal method for ascending concentrations and the logarithmic trapezoidal method for descending concentrations. Drug exposure was extrapolated from the last observed concentration to time infinity by the relationship C_{LAST}/λ_Z for each individual subject to compute total drug exposure ($AUC_{0-\infty}$). The terminal elimination half-life ($t_{1/2}$) was estimated by log-linear regression of 2 to 8 observed concentrations in the terminal elimination phase after the last dose. The maximum concentration after the last dose (C_{max}) and the time to maximum concentration after the last dose (T_{max}) were taken directly from the observed data. The apparent volume of distribution (V_Z/F) and oral clearance (CL/*F*), where *F* is the fraction of drug absorbed, were computed individually according to standard procedures.

Complete *in vivo* conversion of amodiaquine into desethylamodiaquine was assumed (17), and the administered dose of desethylamodiaquine was calculated based on molecular weight (dose of DEAQ = dose of AQ × 327.81 g/mol [molecular weight of DEAQ]/355.86 g/mol [molecular weight of AQ]). Individual ratios (pregnant/postpartum) of parameter estimates for each woman were calculated and summarized to illustrate potential trends.

Statistical analysis. Data were described using the statistical program SPSS for Windows v. 15 (SPSS, Benelux, Inc., Gorinchem, Netherlands) and EpiInfo v. 6 (Centers for Disease Control and Prevention). Continuous normally distributed data were compared by Student's *t* test, and nominal data were compared by the Mann-Whitney U test. Survival analysis on day 63 was studied by use of Kaplan-Meier plots: the endpoint was the first reappearance of *P. vivax*. Patients who delivered, were lost to follow-up, or had *P. falciparum* and were treated with another drug before day 42 were included only until that date. The Wilcoxon matched-pair signed-rank test was used to test the null hypothesis for individual pharmacokinetic parameters during pregnancy and after delivery (STATA v.10).

Efficacy and safety assessment. At enrolment, a full medical history and physical and obstetric examination were carried out. Gestational age of the pregnancy and fetal viability were confirmed by ultrasound. The complete blood count (CBC), parasite count, and biochemistry parameters were measured on admission. On day 14, the CBC and biochemistry studies were repeated.

On each of the first 5 days of the study, patients had a clinical and parasitological evaluation. Adverse effects were evaluated daily until day 5 and weekly thereafter for 6 weeks. Parasitological follow-up was continued for 9 weeks (D63) in total or until delivery. At 3 months postpartum, all women were invited for the second part of the study and followed up for 6 weeks (D42). All cases of reappearing *P. vivax* parasites were treated with chloroquine (total dose of 25 mg base/kg; Government Pharmaceutical Organization, Thailand), and cases of *P. falciparum* parasites were treated with artesunate (2 mg/kg/day for 7 days; Guilin, People's Republic of China) and clindamycin (300 mg three times daily for 7 days; Siam Pharmaceutical Company Ltd., Bangkok, Thailand). All women were encouraged to deliver in the SMRU obstetric facility. Each baby was examined by a trained physician for the presence of congenital abnormalities. Mothers and their babies were invited to monthly follow-up visits.

Definitions. A case of *P. vivax* or *P. falciparum* malaria was defined by the presence in the peripheral blood of asexual-stage parasites of *P. vivax* or *P. falciparum*, respectively. Blood smears (thin and thick films) were prepared using Giemsa coloration and were read for 200 fields before being declared negative. All stages of the parasites were recorded (asexual stage and gametocytes). CBCs were determined in the SMRU laboratory, using a quality-checked Sysmex pocH-100i automated hematology analyzer, and serum samples for biochemistry analysis were sent to the Mae Sot hospital on a daily basis. Anemia was defined by a hematocrit of <30%, with severe anemia defined by a hematocrit of <20%.

TABLE 1. Demographic characteristics on admission for 24 pregnant Karen women with uncomplicated *P. vivax* malaria and the same women at 3 months postpartum

Characteristic	Value[a]	
	Pregnant women (n = 24)	Postpartum women (n = 18)
Median (range) body wt (kg)	49.0 (37.0–68.0)	44.5 (38.0–57.0)
Median (range) age (yr)	23 (18–39)	26 (18–35)
Median (range) ht (cm)	151.5 (137–164)	151.5 (137–164)
Proportion of primigravidae (%)	42 (10/24)	NA
Median (range) gravidity	2 (1–6)	NA
Median (range) parity	1 (0–5)	NA
Proportion of patients who smoked (%)	29 (7/24)	28 (5/18)
Median (range) temp (°C)	36.6 (35.8–40.2)	36.8 (36.0–37.3)
Median (range) duration of fever (days)	1 (0–6)	0
Proportion of women with fever (%)	12.5 (3/24)	0
Median (range) no. of vivax episodes during pregnancy	2 (1–8)	NA
Median (range) hematocrit (%)[b]	32.9 (22.8–39.8)	36.9 (22.3–42.1)
Proportion of anemic women (%)	25 (6/24)	11 (2/18)
Geometric mean (range) parasitemia (no. of parasites/μl)	1,220 (96–50,119)	143 (16–1,023)[c]

[a] NA, not applicable.
[b] From CBCs. For inclusion into the study, we used field hematocrit readings.
[c] Parasitemia in the 6 postpartum women with infection.

RESULTS

From October 2007 to May 2008, 28 pregnant women with acute *P. vivax* malaria consented to participate in the study. Twenty-four women completed the pharmacokinetic sampling during pregnancy. There was one protocol violation (quality control repeat malaria smear parasitemia of <80/μl) and three premature terminations from the study (one woman vomited the study drug and two withdrew consent). These four women continued to follow antenatal care and delivered congenitally normal live singletons. No further analysis of these women was done.

The median (range) gestational age of pregnant women at recruitment was 26.8 (13.2 to 35.5) weeks, and there was no significant difference in baseline characteristics (except for weight) for women enrolled in the second and third trimesters (Table 1).

Postpartum sampling. Nineteen women were sampled at a median (range) of 99 (84 to 173) days postpartum from March 2008 to January 2009 (Table 1). One woman experienced a serious adverse event (SAE) and was excluded from further analysis. Of the remaining 18 women, 6 were diagnosed with an asymptomatic *P. vivax* infection, and the others were smear negative for infection (Table 1). The six women with *P. vivax* infection postpartum were not different in baseline characteristics from the women without *P. vivax* infection (data not shown).

Pharmacokinetics. The results of the noncompartmental analysis of AQ and DEAQ pharmacokinetics in pregnant and postpartum Karen women are shown in Table 2. There was no difference in the pharmacokinetic parameters between preg-

TABLE 2. Pharmacokinetic properties of amodiaquine and desethylamodiaquine in pregnant and postpartum Karen women

Parameter[a]	Median (range) value		*P* value[b] ($n = 18$)
	Pregnant women ($n = 24$)	Postpartum women ($n = 18$)	
Amodiaquine parameters			
Total dose (mg/kg)	29.9 (28.0–32.0)	30.2 (28.0–32.1)	0.407
No. of points for lambda	3.00 (2.00–5.00)	3.00 (3.00–4.00)	0.513
C_{max} (ng/ml)	30.5 (15.7–53.1)	29.7 (15.3–59.3)	0.913
T_{max} (h)	1.46 (0.500–4.00)	2.00 (1.00–3.00)	0.493
CL/*F* (liter/h)	2,380 (1,480–4,470)	2,140 (1,580–4,210)	0.711
CL/*F* (liter/h/kg)	48.8 (29.0–97.1)	52.8 (35.8–82.6)	0.472
V_Z/F (liters)	46,200 (22,300–106,000)	41,200 (21,800–73,200)	0.811
V_Z/F (liters/kg)	1,010 (474–2,070)	886 (504–1,630)	0.711
$t_{1/2}$ (h)	12.4 (6.43–49.5)	11.9 (8.53–23.6)	0.679
$AUC_{0–LAST}$ (h-ng/ml)	576 (293–975)	533 (344–832)	0.349
$AUC_{0–\infty}$ (h-ng/ml)	602 (308–1,090)	573 (382–880)	0.557
$AUC_{0–\infty}$/dose [h-ng/ml/(mg/kg)]	20.5 (10.3–34.5)	18.9 (12.1–28.0)	0.396
Extrapolated AUC (%)	5.21 (2.44–29.3)	6.31 (1.91–13.3)	0.122
Desethylamodiaquine parameters			
Total dose (mg/kg)	28.0 (26.2–35.0)	28.3 (26.2–30.1)	0.896
No. of points for lambda	5.50 (3.00–8.00)	6.00 (5.00–7.00)	0.315
C_{max} (ng/ml)	396 (262–675)	462 (329–828)	0.019
T_{max} (h)	3.00 (2.00–4.00)	3.00 (2.00–4.00)	0.265
CL/*F* (liters/h)	34.1 (17.7–78.6)	29.9 (19.3–64.3)	0.071
CL/*F* (liters/h/kg)	0.736 (0.260–1.51)	0.707 (0.429–1.26)	0.557
V_Z/F (liters)	12,400 (6,630–40,400)	10,900 (7,030–18,700)	0.064
V_Z/F (liters/kg)	252 (109–986)	249 (156–361)	0.286
$t_{1/2}$ (days)	10.0 (5.18–52.7)	10.1 (5.89–19.1)	0.396
$AUC_{0–LAST}$ (h-μg/ml)	36.5 (17.7–69.8)	37.8 (21.8–63.3)	0.711
$AUC_{0–\infty}$ (h-μg/ml)	38.4 (19.1–109)	39.5 (22.5–66.7)	0.648
$AUC_{0–\infty}$/dose [h-μg/ml/(mg/kg)]	1.36 (0.662–3.85)	1.41 (0.793–2.33)	0.811
Extrapolated AUC (%)	3.67 (1.68–36.1)	4.34 (1.97–12.2)	0.711
Day 7 concn (ng/ml)	62.7 (25.8–125)	55.8 (33.5–125)	0.744

[a] No. of points for lambda, number of observations taken in the terminal elimination phase for use in log-linear regression; C_{max}, maximum observed plasma concentration after the last oral administration; T_{max}, observed time to reach C_{max} after last dose; CL, clearance; V_Z, volume of distribution; $t_{1/2}$, terminal elimination half-life; $AUC_{0–LAST}$, predicted area under the plasma concentration-time curve from time zero to the last sampling time; $AUC_{0–\infty}$, predicted area under the plasma concentration-time curve from time zero to infinity; extrapolated AUC, percentage of $AUC_{0–\infty}$ extrapolated from the last observation to infinity; day 7 concentration, measured concentration on day 7.

[b] *P* values were calculated using the paired Wilcoxon signed-rank test for pregnant patients and patients after delivery ($n = 18$).

nant and postpartum women, except for the maximal concentration of DEAQ being higher in nonpregnant women ($P = 0.019$), but the total drug exposures were comparable between both groups (Table 2 and Fig. 1). No significant difference could be seen in pharmacokinetic parameters for AQ or DEAQ in patients in the 2nd trimester compared to those in the 3rd trimester ($P > 0.05$). Pregnant women who displayed recurrent *P. vivax* malaria during follow-up ($n = 10$) showed a significantly lower ($P = 0.036$) dose-normalized amodiaquine exposure [$AUC_{0–\infty}$/dose, 17.6 h-ng/ml/(mg/kg)] than pregnant women with no recurrent malaria ($n = 10$) [$AUC_{0–\infty}$/dose, 26.4 h-ng/ml/(mg/kg)]. The same trend could be seen for desethylamodiaquine [$AUC_{0–\infty}$/dose, 1.26 versus 1.50 h-μg/ml/(mg/kg)], but this difference did not reach statistical significance ($P = 0.089$). There were no significant differences in any pharmacokinetic parameters between postpartum women with *P. vivax* malaria ($n = 6$) and those without *P. vivax* malaria ($n = 12$): for AQ, $AUC_{0–\infty}$/dose $P = 0.482$ and C_{max} $P = 0.426$; and for DEAQ, $AUC_{0–\infty}$/dose $P = 0.282$ and C_{max} $P = 0.242$.

Clinical findings. The median (range) total dose of amodiaquine over 3 days was 30.1 (28.0 to 32.1) mg/kg. Three pregnant women had fever on admission, which cleared within 48 h in all cases. Parasite clearance time was a median (range) of 2 (1 to 4) days. By day 63, four women had *P. falciparum* parasitemia (on days 21, 41, 42, and 63), five women had delivered, and three had moved from the study area. The proportions of women who had a recurrence of *P. vivax* infection at days 28, 42, and 63 were 0 of 20 (0%), 2 of 18 (11.1%), and 6 of 13 (46.2%) women, respectively. The median (range) time to reappearance of *P. vivax* was 52 (35 to 63) days. The overall reappearance rate of *P. vivax* by day 63, using Kaplan-Meier survival analysis, was 74.4% (95% confidence interval [95% CI], 51.5 to 97.3%). None of the postpartum women diagnosed with *P. vivax* parasitemia had fever or a history of fever; parasite clearance time was a median (range) of 1 (1 to 2) day. None of these 6 women had any parasite reappearance in 42 days following treatment with amodiaquine.

Early vomiting occurred in 2.2% (1/46 women) of women, and the repeat dose was tolerated. In pregnant women with malaria, the most commonly reported symptoms not present on enrolment were dizziness (15/24 [63%] women), vomiting (12/24 [50%] women), nausea (12/24 [50%] women), tiredness/weakness (11/24 [46%] women), and anorexia and headache (7/24 [29%] women for each). Itching was reported in only 17% of women (4/24 women). All side effects were reported less often in the postpartum women, including dizziness (4/18 [22%] women), tiredness/weakness (1/18 [6%] women), anorexia (2/18 [11%] women), and headache (1/18 [6%] women),

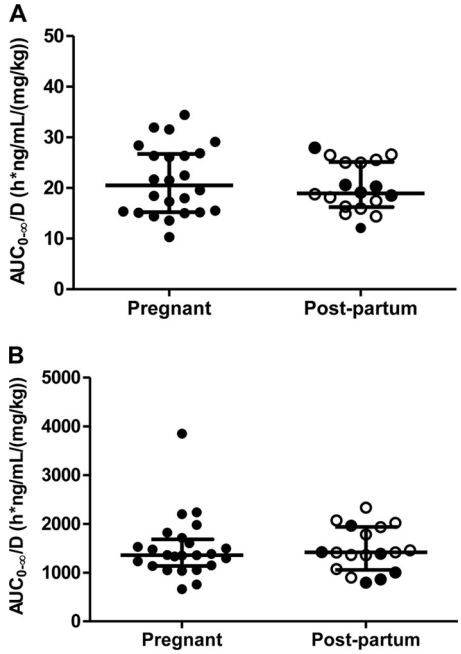

FIG. 1. Total amodiaquine (A) and desethylamodiaquine (B) drug exposures ($AUC_{0-\infty}$/dose) in pregnant ($n = 24$) and postpartum ($n = 18$) women. The black circles indicate predicted values, and the black bars indicate medians and interquartile ranges. Open circles indicate postpartum women without vivax parasites.

with no nausea or vomiting reported. There was one serious adverse event of doubtful relationship to the drug in a postpartum woman with *P. vivax* infection and prolonged fever. Hypotension developed 4 h after the second dose, and electrocardiography showed a sinus bradycardia (pulse, 50 beats/min), a normal axis, and a QTc interval of 442 ms (normal QTc interval is <450 ms). She was already smear negative but remained febrile. The AQ treatment was supplemented with artesunate (4 mg/kg for 3 days), and she was treated for suspected typhoid fever and made a full recovery.

Platelet and white blood cell counts of pregnant women improved significantly from day 0 to day 14, with medians (ranges) ($\times 10^3/\mu l$) of 162 (59 to 255) versus 267 (131 to 447) and 8.9 (3.6 to 11.3) versus 10.7 (6.8 to 17.3), respectively ($P < 0.01$ for both), as expected during malaria. Among biochemical parameters, the median (range) total bilirubin (mg%) decreased significantly from baseline: 0.45 (0.22 to 1.46) versus 0.34 (0.21 to 0.71) ($P = 0.01$). For postpartum women, no significant difference in CBC or biochemical parameters was observed between day 0 and day 14.

Pregnancy outcomes. Of the 24 women, 3 left the study area before the pregnancy outcome was available, 1 had a miscarriage (estimated gestational age, 23.1 weeks; day 61 posttreatment), and the remaining 20 women had live, congenitally normal singleton babies (11 males). The median (range) gestational age was 39.0 (36.4 to 41.2) weeks, based on ultrasounds (before 24 weeks of gestation) performed on all women. There was 1 premature birth and 2 low-birth-weight babies. The placentas of these women were all malaria smear negative. There were no congenital abnormalities. Two infants went back to Burma after delivery; the remaining 18 were followed up until they were 6 months of age. Three infants were infected with *P. vivax* malaria within this period (days 83, 147, and 166) and were treated successfully with CHQ. All infants had achieved normal developmental milestones at their last assessment.

DISCUSSION

Amodiaquine was safe and reasonably well tolerated in this small group of pregnant and postpartum women. Women experienced similar side effects to those reported for chloroquine in the same study area (26). Nausea was reported more commonly, but only 0.7% (1/138 doses) of all doses was vomited. Comparable high rates of mild side effects (dizziness, vomiting, nausea, and tiredness/weakness) were reported for pregnant women treated with AQ in Ghana (22). Only pruritus was less common in our study. There were no signs of agranulocytosis and/or hepatitis (12, 15).

Unlike lumefantrine (10, 24), proguanil, and atovaquone (9), for which dose adjustments in pregnancy are recommended, amodiaquine did not show a change in pharmacokinetic properties during pregnancy. The significantly lower C_{max} observed for DEAQ in pregnant women in this study is likely to have limited clinical impact considering that the actual difference was below 10% and that pregnancy had no effect on total drug exposure or any other pharmacokinetic parameter. It is unlikely that a disease effect masked a difference between pregnancy and postpartum results in this study, since there was no significant pharmacokinetic difference between postpartum women with *P. vivax* infection and those without it. Massele et al. reported an increased blood clearance for chloroquine, a drug structurally similar to amodiaquine, during the second and third trimesters in pregnant Tanzanian women ($n = 49$) who received weekly chloroquine prophylaxis (8). This was not seen in the present study in a paired comparison between total drug exposure of AQ and DEAQ in postpartum women and that during the second (for AQ, $P = 0.767$; and for DEAQ, $P = 0.314$) or third (for AQ, $P = 0.374$; and for DEAQ, $P = 0.374$) trimester of pregnancy (data not shown).

However, caution is needed in interpreting pharmacokinetic results derived from NCA of drugs with high interindividual variability, especially for small sample sizes. Model-independent results are also highly dependent on the sampling strategy, and the total exposure of AQ and DEAQ could be underestimated slightly, since dense sampling was not performed during the first two doses. However, an accurate curve stripping cannot be performed for drugs with multiphasic distribution profiles and long half-lives. Furthermore, the derived pharmacokinetic parameters obtained using NCA were almost identical to estimates derived from a population pharmacokinetic approach (our unpublished data). The exact same sampling schedules were also applied to both groups, which should minimize the risk of a biased comparison between pregnant and postpartum women. Interindividual variability should not be a major confounder, since a crossover approach was used where subjects functioned as their own controls in the paired comparison.

The dose-normalized total drug exposures reported in this study for pregnant women with *P. vivax* malaria [for AQ, 20.5 h-ng/ml/(mg/kg); and for DEAQ, 1.36 h-μg/ml/(mg/kg)] are

comparable to those published previously for amodiaquine treatment of nonpregnant healthy volunteers [for AQ, 16.2 h-ng/ml/(mg/kg); and for DEAQ, 1.20 h-μg/ml/(mg/kg)] (16). Dose-normalized total amodiaquine exposure was significantly lower in pregnant women who displayed recurrent malaria during follow-up than in pregnant women without malaria, suggesting that high drug exposure suppresses recurrent malaria. Desethylamodiaquine has a longer half-life than amodiaquine, and the main protective effect is more likely to result from a high desethylamodiaquine exposure. However, the desethylamodiaquine exposure in pregnant women with recurrent malaria during follow-up was not significantly different from that in women without recurrent malaria. This might be on account of the small number of subjects studied here.

In conclusion, this preliminary analysis using a noncompartmental approach does not indicate important differences in the pharmacokinetics of AQ or the main metabolite DEAQ between pregnant and postpartum women.

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REFERENCES

1. **Baird, J. K.** 2009. Resistance to therapies for infection by Plasmodium vivax. Clin. Microbiol. Rev. **22:**508–534.
2. **Bhattarai, A., M. Maini-Thapar, A. S. Ali, and A. Bjorkman.** 2004. Amodiaquine during pregnancy. Lancet Infect. Dis. **4:**721–722.
3. **Clerk, C. A., et al.** 2008. A randomized, controlled trial of intermittent preventive treatment with sulfadoxine-pyrimethamine, amodiaquine, or the combination in pregnant women in Ghana. J. Infect. Dis. **198:**1202–1211.
4. **D'Alessandro, U., and F. O. ter Kuile.** 2006. Amodiaquine, malaria, pregnancy: the old new drug. Lancet **368:**1306–1307.
5. **Hietala, S. F., et al.** 2007. Population pharmacokinetics of amodiaquine and desethylamodiaquine in pediatric patients with uncomplicated falciparum malaria. J. Pharmacokinet. Pharmacodyn. **34:**669–686.
6. **Hodel, E. M., et al.** 2009. A single LC-tandem mass spectrometry method for the simultaneous determination of 14 antimalarial drugs and their metabolites in human plasma. J. Chromatogr. B Anal. Technol. Biomed. Life Sci. **877:**867–886.
7. **Jullien, V., et al.** 2010. Population pharmacokinetics and pharmacodynamic considerations of amodiaquine and desethylamodiaquine in Kenyan adults with uncomplicated malaria receiving artesunate-amodiaquine combination therapy. Antimicrob. Agents Chemother. **54:**2611–2617.
8. **Massele, A. Y., et al.** 1997. Chloroquine blood concentrations and malaria prophylaxis in Tanzanian women during the second and third trimesters of pregnancy. Eur. J. Clin. Pharmacol. **52:**299–305.
9. **McGready, R., et al.** 2003. The pharmacokinetics of atovaquone and proguanil in pregnant women with acute falciparum malaria. Eur. J. Clin. Pharmacol. **59:**545–552.
10. **McGready, R., et al.** 2006. The pharmacokinetics of artemether and lumefantrine in pregnant women with uncomplicated falciparum malaria. Eur. J. Clin. Pharmacol. **62:**1021–1031.
11. **Mutabingwa, T. K., et al.** 2009. Randomized trial of artesunate+amodiaquine, sulfadoxine-pyrimethamine+amodiaquine, chlorproguanal-dapsone and SP for malaria in pregnancy in Tanzania. PLoS One **4:**e5138.
12. **Naing, T., H. Win, and Y. Y. Nwe.** 1988. Falciparum malaria and pregnancy: relationship and treatment response. Southeast Asian J. Trop. Med. Public Health **19:**253–258.
13. **Navaratnam, V., et al.** 2009. Tolerability and pharmacokinetics of non-fixed and fixed combinations of artesunate and amodiaquine in Malaysian healthy normal volunteers. Eur. J. Clin. Pharmacol. **65:**809–821.
14. **Nosten, F., F. ter Kuile, L. Maelankirri, B. Decludt, and N. J. White.** 1991. Malaria during pregnancy in an area of unstable endemicity. Trans. R. Soc. Trop. Med. Hyg. **85:**424–429.
15. **Olliaro, P., et al.** 1996. Systematic review of amodiaquine treatment in uncomplicated malaria. Lancet **348:**1196–1201.
16. **Orrell, C., et al.** 2008. Pharmacokinetics and tolerability of artesunate and amodiaquine alone and in combination in healthy volunteers. Eur. J. Clin. Pharmacol. **64:**683–690.
17. **Parikh, S., J. B. Ouedraogo, J. A. Goldstein, P. J. Rosenthal, and D. L. Kroetz.** 2007. Amodiaquine metabolism is impaired by common polymorphisms in CYP2C8: implications for malaria treatment in Africa. Clin. Pharmacol. Ther. **82:**197–203.
18. **Rijken, M. J., et al.** 2009. Obstetric ultrasound scanning by local health workers in a refugee camp on the Thai-Burmese border. Ultrasound Obstet. Gynecol. **34:**395–403.
19. **Sinou, V., et al.** 2009. Pharmacokinetics and pharmacodynamics of a new ACT formulation: artesunate/amodiaquine (TRIMALACT) following oral administration in African malaria patients. Eur. J. Drug Metab. Pharmacokinet. **34:**133–142.
20. **Steketee, R. W., A. D. Brandling-Bennett, D. C. Kaseje, I. K. Schwartz, and F. C. Churchill.** 1987. In vivo response of Plasmodium falciparum to chloroquine in pregnant and non-pregnant women in Siaya District, Kenya. Bull. World Health Organ. **65:**885–890.
21. **Stepniewska, K., et al.** 2009. Population pharmacokinetics of artesunate and amodiaquine in African children. Malar. J. **8:**200.
22. **Tagbor, H., et al.** 2006. Efficacy, safety, and tolerability of amodiaquine plus sulphadoxine-pyrimethamine used alone or in combination for malaria treatment in pregnancy: a randomised trial. Lancet **368:**1349–1356.
23. **Tagbor, H. K., D. Chandramohan, and B. Greenwood.** 2007. The safety of amodiaquine use in pregnant women. Expert Opin. Drug Saf. **6:**631–635.
24. **Tarning, J., et al.** 2009. Population pharmacokinetics of lumefantrine in pregnant women treated with artemether-lumefantrine for uncomplicated Plasmodium falciparum malaria. Antimicrob. Agents Chemother. **53:**3837–3846.
25. **Thomas, F., A. Erhart, and U. D'Alessandro.** 2004. Can amodiaquine be used safely during pregnancy? Lancet Infect. Dis. **4:**235–239.
26. **Villegas, L., et al.** 2007. Chloroquine prophylaxis against vivax malaria in pregnancy: a randomized, double-blind, placebo-controlled trial. Trop. Med. Int. Health **12:**209–218.
27. **Ward, S. A., E. J. Sevene, I. M. Hastings, F. Nosten, and R. McGready.** 2007. Antimalarial drugs and pregnancy: safety, pharmacokinetics, and pharmacovigilance. Lancet Infect. Dis. **7:**136–144.
28. **White, N. J., R. M. McGready, and F. H. Nosten.** 2008. New medicines for tropical diseases in pregnancy: catch-22. PLoS Med. **5:**e133.
29. **Winstanley, P. A., et al.** 1990. The disposition of amodiaquine in Zambians and Nigerians with malaria. Br. J. Clin. Pharmacol. **29:**695–701.