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The pharmacokinetics of three multiple dose regimens of chloroquine: implications for malaria chemoprophylaxis

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The pharmacokinetics of chloroquine were studied in healthy volunteers who received one of three different multiple-dose regimens for 3 weeks: once weekly 300 mg, twice weekly 200 mg and once daily 50 mg chloroquine. Plasma concentrations of chloroquine and metabolites were determined by h.p.l.c. with fluorescence detection. The concentration-time course was fitted to a multiple-dose pharmacokinetic model. Volume of distribution, elimination half-life and clearance were not different for the three regimens, ranging from 250–302 l kg^{-1}, 374–479 h and 0.44–0.58 l h^{-1} kg^{-1} respectively. After the first week of all dosage regimens, peak and trough concentrations of chloroquine were above 16 μg l^{-1}, sufficiently suppressive for chloroquine-sensitive *P. falciparum* strains. These data suggest that once daily chloroquine could be combined with proguanil in a single tablet and should improve compliance when given for malaria chemoprophylaxis.

Keywords chloroquine multiple-dose chemoprophylaxis malaria

Introduction

For more than three decades chloroquine has been the drug of choice for the treatment and prevention of malaria. The first signal of emerging chloroquine-resistance of *Plasmodium falciparum* was the failure of chloroquine chemoprophylaxis in non-immune travellers to East Africa [1, 2].

For travellers to chloroquine-resistant areas, sulfadoxine-pyrimethamine (SP) was at one time recommended but then discarded because of serious side-effects. In several European countries SP was replaced by the combined use of chloroquine and proguanil (C+P). More recently, mefloquine has become first choice for chloroquine-resistant malarious areas; however, several contraindications and emerging resistance leave a place for C+P-prophylaxis. Ideally, malaria chemoprophylactic regimens are designed in such a way that drug concentrations are quickly raised above the suppressive concentration and remain sufficiently high without reaching toxic levels.

To distinguish prophylaxis-failure from non-compliance with the recommended prophylaxis, measurement of chloroquine concentrations in whole-blood, plasma or serum is essential [3–6].

Compliance with malaria chemoprophylaxis is notoriously poor [7, 8] and decreases with complexity of the prescription [9].

To investigate whether daily chloroquine would result in adequate suppressive plasma concentrations, the pharmacokinetic parameters of chloroquine were determined in healthy volunteers during three different multiple dose prophylactic regimens.

The follow up of the subjects was about 6 weeks.

Methods

Subjects and sampling

The study protocol was reviewed and approved by the Institutional Review Board of the hospital of the University of Amsterdam. Group 1 consisted of five subjects (three male/two female) with a mean age of 41 years and a mean (± s.d.) bodyweight of 64 (± 10) kg. Group 2 consisted of four male subjects with a mean age of 31 years and a mean (± s.d.) weight of 76 (± 7) kg. Group 3 consisted of five male subjects with a mean age of 30 years and a mean (± s.d.) weight of 73 (± 8) kg.

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Informed consent was obtained from all subjects. Group 1 received three weekly dosages of 300 mg chloroquine base (Nivaquine®, Specia, Alkmaar, The Netherlands) (total dose: 900 mg). Group 2 received twice weekly 200 mg chloroquine base during 3 weeks (total dose: 1200 mg). Group 3 received once daily 50 mg chloroquine for 3 weeks (total dose: 1050 mg). In all groups, blood samples were drawn before the first dose and at 0.5, 1, 2, 3, 4, 6, 10, 24, 32, 48, 72 and 96 h, at days 6, 9, 14 and from then on once weekly.

Analysis of chloroquine and metabolites

Blood was centrifuged and plasma collected and stored at −20° C until analysis. Plasma samples were analysed for chloroquine, deethylchloroquine and *bis*-deethylchloroquine using a previously published h.p.l.c. method [6].

With this assay plasma from several pools did not show any interfering peaks. Calibration plots using four concentrations of 25, 50, 100 and 200 μg l^{-1}, chloroquine and its metabolites were linear, with correlation coefficients greater than 0.998.

Duplicate measurements revealed coefficients of variation of 6.5% for the range 0–100 μg l^{-1} and 7.1% for 100–400 μg l^{-1} chloroquine. The detection limit was 1 μg l^{-1}. All results are given as the concentration of chloroquine base.

Pharmacokinetic calculations

Plasma chloroquine and deethylchloroquine concentration-time curves were fitted to a three-compartment linear model with first order absorption, using the computer program NONLIN [10], with a special subroutine for the curve fitting of multiple dose concentration-time profiles.

The following equation describes the model used:

$$C_t = C_1e^{-\lambda_1 t} + C_2e^{-\lambda_2 t} + C_3e^{-\lambda_3 t} - (C_1 + C_2 + C_3)e^{-k_a t}$$

The terminal elimination rate constant λ_3 was derived from the terminal series of the plasma concentrations of chloroquine and deethylchloroquine by log-linear regression. All observations received a weighting of one.

The area under the curve (AUC) was calculated according to the following expression:

$$\text{AUC} = \frac{C_1}{\lambda_1} + \frac{C_2}{\lambda_2} + \frac{C_3}{\lambda_3} - \frac{(C_1 + C_2 + C_3)}{k_a}$$

and also with the trapezoidal rule with extrapolation of the terminal part of the curve as last concentration/λ_3. The volume of distribution V_{area}/F was calculated as CL/λ_3 and clearance (CL/F) as dose/AUC.

Mean plasma chloroquine concentrations were calculated for each group of subjects receiving the same dosing regimen and the mean data curves subjected to the curve fitting procedure using the NONLIN program.

Statistical analysis

The estimated parameters of the three regimens were compared using ANOVA and calculation of the 95% confidence intervals of the difference between two groups.

Results

Total sampling times were 53, 40 and 42 days for groups 1, 2 and 3 respectively. The mean number (min-max) of plasma-samples was 15.8 (15–18), 14.8 (12–16) and 14.8 (14–16) respectively.

Curve fitting was successful, applying a three compartment model for all subjects. Mean r^2 (min-max) = 0.979 (0.96–0.988) for group 1, 0.896 (0.851–0.936) for group 2 and 0.951 (0.93–0.964) for group 3. Attempts to apply a two-compartment model did not result in better fittings [11].

The chloroquine plasma concentration-time curves of the mean data of the three groups are shown in Figure 1.

The estimated pharmacokinetic parameters of the three regimens are shown in Table 1. The area under

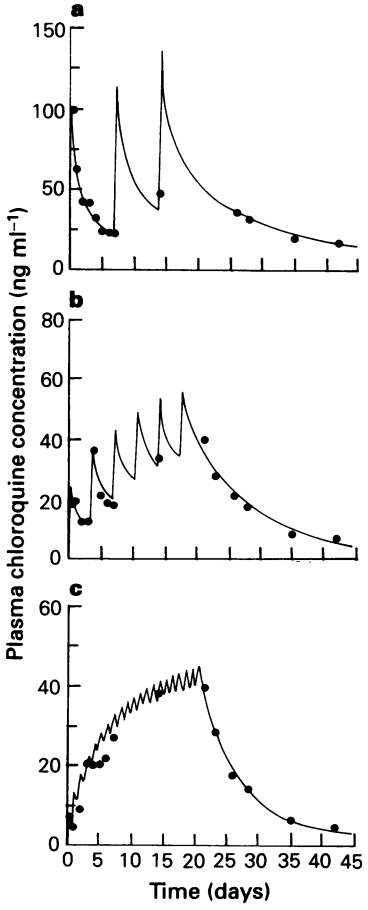

Figure 1 Mean plasma chloroquine concentration-time curves during three different chemoprophylactic regimens for 3 weeks: a) chloroquine 300 mg once weekly for 3 weeks; b) chloroquine 200 mg twice weekly; c) chloroquine 50 mg daily.

Table 1 Chloroquine pharmacokinetic parameters of three multiple dose prophylactic regimens

	Dosing regimens					
	1: 1 × 300 mg week^{-1} (n = 5)	*2: 2 × 200 mg week^{-1}* (n = 4)	*3: 7 × 50 mg week^{-1}* (n = 5)	*95% confidence intervals of the difference (CI) between two regimens*		
				CI 1–2	*CI 1–3*	*CI 2–3*
V/F (l kg^{-1})	250 (116)	302 (102)	283 (112)	−91 to 195	−108 to 174	−121 to 159
$t_{1/2}$ (h)	386 (108)	374 (144)	479 (323)	−160 to 182	−206 to 392	−211 to 421
CL/*F* (l h^{-1} kg^{-1})	0.441 (0.143)	0.579 (0.117)	0.571 (0.114)	−0.032 to 0.038	−0.031 to 0.290	−0.144 to 0.161
AUC (μg l^{-1} h)*	24751	13977	13257	−154 to 21703	−246 to 23236	−4482 to 5922

V = volume of distribution.
$t_{1/2}$ = elimination half-life.
CL = clearance.
*AUC: corrected to a single 600 mg dose.
Data are expressed as the mean (s.d.) value.

the curve was corrected to correspond to a single dose of 600 mg in order to permit comparison. No significant differences in parameters were detected. In all subjects *bis*-deethylchloroquine was detectable. The mean (min-max) AUC-ratios chloroquine/ deethylchloroquine for the three regimens were 0.36 (0.19–0.48), 0.40 (0.22–0.52) and 0.79 (0.46–0.99) respectively. The mean (min-max) AUC-ratios deethylchloroquine/*bis* deethylchloroquine were: 1.10 (0.88–1.42), 2.19 (1.48–3.24) and 1.53 (1.14–1.92) respectively.

In chemoprophylaxis no parasitaemia should be present at a chloroquine concentration in plasma of ≥ 16 μg l^{-1} [3]. In group 1 the chloroquine concentration in the first week decreased below 16 μg l^{-1} in all subjects. In one subject the concentration did not reach 16 μg l^{-1} at all, in the four other subjects the last sample with a higher concentration was taken at 24, 72, 96 and 120 h. In the second week, i.e. after the second dose, plasma chloroquine concentration was always above 16 μg l^{-1}.

In group 2 the plasma chloroquine concentration dropped below 16 μg l^{-1} in three subjects in the first dosing interval. In the second interval this occurred in two subjects. In the subjects of group 3 plasma chloroquine concentration reached 16 μg l^{-1} after 48, 72, 120 and 144 h.

Discussion

Our study shows that all three prophylactic regimens provide adequate chloroquine concentrations to suppress infection with a chloroquine-sensitive *P. falciparum* strain.

In all regimens the peak and trough levels were above the target concentration of 16 μg l^{-1} [3] after 1 week. As the erythrocytic phase of the malarial infection is not reached within 7–10 days after the bite of an infecting mosquito in the malarious area, sensitive parasites will encounter suppressive chloroquine levels.

Our multiple prophylactic dose experiments revealed kinetic parameters for chloroquine comparable with the results from the therapeutic regimen previously reported by us [6]. The reported volume of distribution from studies in man ranges from 130–880 l kg^{-1} and half-life from 8 to 58 days [4–6], comparable with our data (Table 1). The concentration-time curves could adequately be fitted using a three compartment model. With dose dependent kinetics this would not have been possible. Moreover, the derived kinetic parameters were comparable with those found in a single dose study [12]. The AUC-ratios of the metabolites were not significantly different when comparing the three regimens. This does not point to a saturable metabolic process.

There is much disagreement concerning the preferred prophylactic regimen for areas with high transmission of chloroquine-resistant *Plasmodium falciparum*, such as subSaharan Africa. In the Netherlands the combined use of weekly chloroquine and daily proguanil (C+P) has been recommended for these areas since 1983. After emergence of chloroquine-resistance in a certain area, the resistance appears to stabilise at a certain level, as noted by several authors [13–15]. A comparison of three regimens for malaria prophylaxis in Dutch travellers [7] performed between 1987–1989 showed a risk of a prophylaxis failure of 5.4 per 1000 person-months for chloroquine 300 mg weekly with once daily 100 mg proguanil and 2.8 per 1000 person-months for chloroquine with 200 mg proguanil daily. This was not statistically different. Gozal *et al.* showed that C+P prophylaxis in a non-immune resident population in Cameroon is still effective [16].

The conclusion of this study is that a daily tablet of chloroquine (50 mg) results in adequate prophylactic concentration. As shorter intervals between low dosages reduce peak levels, a daily dose could improve safety [17].

As compliance with prophylaxis is an even greater problem than drug resistance, one daily tablet of chloroquine (50 mg) and proguanil (100–200 mg) is feasible and should help to improve compliance.

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