Lack of relationship between debrisoquine oxidation phenotype and the pharmacokinetics of quinine

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The relationship between debrisoquine oxidation phenotype and the pharmacokinetics of quinine after a single dose (600 mg) of quinine sulphate was studied in eight extensive metabolizers (EM) and five poor metabolizers (PM). The mean elimination half-life of quinine in the PMs (10.2 ± 1.6 (s.d.) h) was similar to that in the EMs (10.9 ± 1.7 h). The oral clearance of quinine in the PM subjects was 0.092 ± 0.021 l h⁻¹ kg⁻¹ and was not significantly different ($P > 0.05$) from that observed in the EM subjects (0.073 ± 0.019 l h⁻¹ kg⁻¹). This suggests that even though quinine is extensively metabolized by oxidative biotransformation, this is carried out largely by P450 isoenzymes different from P450IID6 which oxidizes debrisoquine.

Keywords quinine debrisoquine metabolism pharmacokinetics genetic polymorphism

Introduction

Quinine is one of the oldest drugs in the pharmacopoeia. While synthetic antimalarial drugs have largely replaced quinine, the emergence of strains of *Plasmodium falciparum* resistant to chloroquine in Southern Asia, South America and East Africa, has necessitated its continued therapeutic use (White, 1985; White *et al.*, 1983). Some 42% (2,117 million) of the world population live in endemic malaria areas (WHO, 1989). Quinine is also widely prescribed for the relief of nocturnal leg cramps. In man, quinine undergoes extensive oxidative metabolism with approximately 20% of an oral dose being excreted unchanged in the urine (Bruce-Chwatt, 1981; White, 1985).

In preparations of human liver microsomes, quinidine and quinine have been shown to be potent competitive inhibitors of debrisoquine (Guengerich *et al.*, 1986a; Inaba *et al.*, 1985; Speirs *et al.*, 1986). Therefore, it is possible that quinine is also oxidized by the same isoenzyme which metabolized debrisoquine *i.e.*, cytochrome P450IID6 (Gonzalez *et al.*, 1988). The elimination of more than 20 clinically used drugs is influenced by the debrisoquine/sparteine oxidation polymorphism (Brosen & Gram, 1989). To determine if the elimination of quinine is related to debrisoquine oxidation phenotype, we have studied its pharmacokinetics after a single oral dose in poor metabolizers (PMs) and extensive metabolizers (EMs) of debrisoquine.

Methods

The study was approved by the Otago Area Health Board Ethics Committee, Dunedin, New Zealand. All subjects gave written informed consent.

Subjects

Ten healthy men and three healthy women, aged 20–35 years, participated in the study. All volunteers were non-smokers, of normal body weight and height (Table 1) and were normotensive for their age. At the beginning of the investigation they had a full clinical examination, together with haematological and biochemical screening tests and urinalysis to confirm that clinically significant abnormalities, particularly relating to hepatic and renal function, were absent. During the 2 preceding weeks and the period of study none of the subjects took alcohol or any other drugs. The volunteers were randomly selected from groups of subjects whose oxidation phenotype had been characterized (Wanwimolruk *et al.*, 1991a) at least 1 month prior to quinine dosing. Five subjects were phenotyped (Wanwimolruk & Ferry, 1990) as debrisoquine PM (metabolic ratio of debrisoquine/4-hydroxydebrisoquine in 0–4 h urine: 13.4–120.7). Eight subjects were identified as EM (metabolic ratio of debrisoquine/4-hydroxydebrisoquine in 0–4 h urine: 0.22–8.29).

Table 1 Subject details and quinine pharmacokinetics in healthy poor metabolizers (PMs) and extensive metabolizers (EMs) of debrisoquine after a single oral dose of 600 mg quinine sulphate

	PMs	EMs	Mann-Whitney test
<i>Subject details</i>			
Number of subjects	5 (1F/4M)	8 (2F/6M)	—
Age (years)	25.8 ± 6.1* (21–35)**	22.8 ± 4.0 (20–35)	NS
Weight (kg)	80 ± 15 (56–96)	75 ± 10 (61–87)	NS
Cigarette smoker	None	None	—
Alcohol drinker	5	8	—
<i>Quinine pharmacokinetic parameters</i>			
$C_{\max}$ (mg l ⁻¹)	4.6 ± 0.8 (3.2–5.4)	6.2 ± 0.9 (4.0–7.4)	NS
$t_{\max}$ (h)	2.7 ± 0.9 (1.5–4)	2.4 ± 0.6 (1.5–3)	NS
$t_{1/2}$ (h)	10.2 ± 1.6 (8.1–12.2)	10.9 ± 1.7 (8.7–12.9)	NS
AUC (mg l ⁻¹ h)	72 ± 17 (53–86)	97 ± 20 (67–122)	NS
Oral clearance (l h ⁻¹ kg ⁻¹)	0.092 ± 0.021 (0.070–0.113)	0.073 ± 0.019 (0.051–0.110)	NS
Protein binding (% unbound)	12.2 ± 2.3 (8.5–14.4)	12.7 ± 1.6 (10.5–14.9)	NS
% Dose excreted as unchanged drug in the urine (0–48 h)	10.3 ± 2.2 (7.5–12.8)	11.8 ± 2.5 (8.4–15.6)	NS
Renal clearance (l h ⁻¹ kg ⁻¹)	0.010 ± 0.003 (0.006–0.011)	0.009 ± 0.003 (0.005–0.016)	NS

* Data given as mean ± s.d.

** Range of the values.

NS: $P > 0.05$.

Protocol

At 09.00 h after an overnight fast subjects took a single dose of 600 mg quinine sulphate (497 mg quinine base). The tablets were swallowed with 200 ml tap water and no food was allowed for the following 3 h. Blood samples (4 ml each) were drawn through a heparinized intravenous catheter (Venflon) or by individual venepuncture before and at 0.25, 0.5, 0.75, 1, 1.5, 2, 2.5, 3, 4, 6, 8, 10, 12, 24, 30, 36 and 48 h after dosing. Blood was collected in heparinized test-tubes and was centrifuged within 2 h. Urine was collected over the 48 h period following the administration of the drug. Plasma was kept frozen (–20° C) until analysis.

Analytical procedures

Quinine concentrations in plasma and urine samples were assayed by a high-performance liquid chromatographic procedure (Zoest *et al.*, 1990). The inter-assay coefficient of variation was less than 8% over the concentration range of 0.02 to 10 mg l⁻¹. Blank plasma collected prior to quinine administration showed no endogenous sources of interference with the assay. Plasma protein binding of quinine was measured by

equilibrium dialysis (Wanwimolruk *et al.*, 1982) using the Dianorm® apparatus. Blank plasma samples were used to determine the protein binding. Plasma was dialysed against 0.1 M phosphate buffer (pH 7.4) at 37° C for 3 h with quinine at an initial concentration of 3 mg l⁻¹. Quinine concentrations were then estimated in the plasma and buffer sides as above. There were no significant changes in volume and pH after 3 h of dialysis.

Pharmacokinetic and statistical analysis

The elimination rate constant (λ_z) was calculated by linear least-squares regression analysis of the terminal phase of the log plasma drug concentration-time profile. The area under the plasma drug concentration-time curve from zero to infinity time (AUC) was calculated by the linear trapezoidal rule from the beginning of drug administration to the last datum point and with extrapolation to infinity. The area from the last datum point (C_t) to infinity was obtained as C_t/λ_z . Oral clearance was calculated from dose/AUC. The elimination half-life ($t_{1/2}$) was calculated from $0.693/\lambda_z$. $C_{\max}$ and $t_{\max}$ values were noted directly from the data. Renal clearance was calculated as (amount excreted unchanged in urine between 0 and 48 h)/AUC (0–48 h).

Results are reported as mean $\pm$ s.d. Statistical differences in pharmacokinetics were analysed by the Mann-Whitney U-test. Values of $P > 0.05$ were regarded as not significant (NS).

Results and discussion

The PM and EM subjects were similar in sex distribution, age and body weight (Table 1). All volunteers were in good physical condition as assessed by medical examination and laboratory tests.

Figure 1 shows mean plasma concentrations of quinine in the PM and EM subjects.

The pharmacokinetic data are summarized in Table 1. There were no significant differences between PMs and EMs in $C_{\max}$ and $t_{\max}$ ($P > 0.05$). The mean AUC in PM subjects was 72 ± 17 (s.d.) $\text{mg l}^{-1} \text{h}$ and was not significantly different ($P > 0.05$) from that observed in the EMs ($97 \pm 20 \text{ mg l}^{-1} \text{h}$). The mean oral clearance of quinine was $0.092 \pm 0.021 \text{ l h}^{-1} \text{ kg}^{-1}$ in PMs and $0.073 \pm 0.019 \text{ l h}^{-1} \text{ kg}^{-1}$ in EMs. The mean elimination half-life of quinine was $10.2 \pm 1.6 \text{ h}$ and $10.9 \pm 1.7 \text{ h}$, respectively. There were no significant differences in either the oral clearance or the half-life of quinine between the two groups. Plasma protein binding of quinine in the PMs was similar to that in the EMs (12.2 ± 2.3 vs $12.7 \pm 1.6\%$ unbound). The mean 48 h recovery (% of dose) of quinine in the PMs and EMs was $10.3 \pm 2.2\%$ and $11.8 \pm 2.5\%$, respectively. There was no difference between either phenotype in this recovery. The mean renal quinine clearance in PM subjects was $0.010 \pm 0.003 \text{ l h}^{-1} \text{ kg}^{-1}$ and this was comparable with the mean value found in the EM subjects ($0.009 \pm 0.003 \text{ l h}^{-1} \text{ kg}^{-1}$, $P > 0.1$). The power to detect a significant difference between parameters in the two groups is low. This cannot rule out the possibility of a type II error due to a small sample size.

The pharmacokinetic parameters found for quinine in the present study are comparable with those reported previously (Wanwimolruk *et al.*, 1991b; White, 1985; White *et al.*, 1983). The absence of significant difference in the plasma drug concentration-time profiles between

Figure 1 Mean plasma quinine concentration following administration of a single oral dose of 600 mg quinine sulphate to five PMs ($\blacktriangle$ ----- $\blacktriangle$) and eight EMs ($\circ$ — $\circ$).

PMs and EMs suggests that debrisoquine oxidation phenotype does not influence the kinetics of quinine. These results also indicate that P450IID6 is unlikely to be involved in the major metabolic pathways of quinine. This study does not definitely rule out a role of P450IID6 in the metabolism of quinine as it is possible for pathways to appear polymorphic with one substrate and yet unimorphic with another. However, our findings are similar to those reported for the diastereoisomer quinidine (Brosen *et al.*, 1990; Mikus *et al.*, 1986) in that P450IID6 was not implicated as making a major contribution to metabolism. Furthermore, *in vitro* studies have shown that quinine is metabolized by P450 as an isoenzyme that oxidizes nifedipine but not debrisoquine (Guengerich *et al.*, 1986a,b).

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