# Efficacy of sulfadoxine-pyrimethamine and mefloquine for the treatment of uncomplicated *Plasmodium falciparum* malaria in the Amazon basin of Peru

## Eficácia da sulfadoxina-pirimetamina e mefloquina no tratamento de malária não-complicada por *Plasmodium falciparum* na bacia amazônica peruana

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**ABSTRACT**

*In vivo* antimalarial drug efficacy studies of uncomplicated *Plasmodium falciparum* malaria at an isolated site in the Amazon basin of Peru bordering Brazil and Colombia showed >50% RII/RIII resistance to sulfadoxine-pyrimethamine but no evidence of resistance to mefloquine.

**Key-words**: Antimalarial drug resistance. *Plasmodium falciparum*. Sulfadoxine-pyrimethamine. Mefloquine. Peru.

**RESUMO**

*Testes in vivo foram realizados para avaliar resistência a drogas antimalária, em pessoas com malária não complicada, causada por* Plasmodium falciparum, *numa região isolada da Bacia Amazônica, na fronteira com o Brasil e a Colômbia. Os testes mostraram resistência >50% RII/RIII a sulfadoxina-pirimetamina, mas não evidenciaram resistência a mefloquina.*

**Palavras-chaves**: Resistência a drogas antimalárica. *Plasmodium falciparum*. Sulfadoxina-pirimetamina. Mefloquina. Peru.

Although numerous *in vivo* antimalarial drug efficacy studies have been conducted in the Amazon Basin of South America, the great majority have been carried out in or near large towns or cities, such as Manaus, Porto Velho, and Iquitos, where greater numbers of infected patients are available and the infrastructure necessary to support such studies is more developed. As a result, there are large areas of the Amazon region where little or no information on antimalarial drug resistance is available. We conducted studies of the efficacy of sulfadoxine-pyrimethamine (SP) and mefloquine (MQ) in 1999 and 2000 in the town of Caballococha, population 3,300, in the northeastern Peruvian Amazon region, located less than 30km from the Brazilian and Colombian borders (Figure 1).

We followed World Health Organization/Pan American Health Organization guidelines for *in vivo* antimalarial drug efficacy testing[1-4]. The protocols were approved by the Institutional Review Boards of the U.S. Army, the U.S. Navy, and the Universidad Cayetano Heredia. Patients between 6 months and 60 years of age whose thick blood smears were being examined at the Caballococha Health Center were screened for malaria parasitemia. Those with *P. falciparum* monoinfections between 250 and 50,000 parasites/µl of blood and an axillary temperature ≥37.5°C and/or a history of fever within the previous 72 hours who gave informed consent were enrolled. Subjects were excluded if they had symptoms or signs of severe malaria, had another obvious cause for their fever, had a history of allergy to either of the study drugs, or

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*Figure 1 - Caballococha. Northeastern Peruvian Amazon Region.*

were pregnant or had a positive urine pregnancy test. Sample sizes were calculated based on an expected rate of treatment failure in the population of 25% for SP and 5% for MQ with a precision of 5% and a 5% level of significance.

Subjects were treated under supervision with SP (Fansidar®; Roche SA, Basel, Switzerland), 25mg/kg of the sulfonamide component in a single dose, or MQ (Mephaquin®, Mepha Ltda, Aesch-Basel, Switzerland), 15mg/kg in a single dose. For the SP trial, subjects were asked to return on days 1, 2, 3, 7, and 14; for the MQ trial, additional follow-up visits were made on days 21 and 28. Patients who failed treatment with SP were treated with a 7-day course of quinine plus tetracycline.

Thick blood smears were stained with Giemsa and the parasite density calculated by counting the number of asexual parasites per 300 white blood cells, based on a manual white blood cell count (SP trial) or assuming a mean white blood cell count of 6,000/µl (MQ trial). Each blood smear was independently examined by two microscopists. A total of 200 oil immersion fields were examined before a blood smear was considered negative. The subjects' parasitologic and clinical response to therapy were classified according to WHO/PAHO guidelines[1,4].

Following a major resurgence of malaria in the Amazon region of Peru between 1993 and 1998, malaria incidence fell quite dramatically in 1999 and 2000, making it increasingly difficult to identify subjects for study. As a result, we were unable to attain the intended sample sizes. Of 100 patients with *P. falciparum* malaria whose blood smears were screened over a four-week period in 1999, only 40 could be enrolled in the evaluation of SP. Fifty-four (90%) of the 60 who were not enrolled were not available for initial evaluation or follow-up because only their blood smears had been sent to health center for review. The remaining six patients were not enrolled because they had received prior drug therapy (n = 3), had parasitemia >50,000/µl (n = 2), or had only *P. falciparum* gametocytes on blood smear (n = 1). The following year, a total of 999 febrile patients were screened over a 13-week period, but only 28 had *P. falciparum* malaria. Eighteen of these were enrolled in the evaluation of MQ; the other 10 patients had parasite densities < 250/µl on their initial blood smears.

Overall, 46 (80.7%) of the 58 subjects were males; their median age was 26.4 years. Twenty-three (42.6%) of the subjects had a documented fever on enrollment and 98.1% gave a history of fever in the previous 72 hours. Subjects had a history of 5.3±4.0 days of fever before they were enrolled. Their geometric mean parasite density was 5,856 parasites/µl.

Thirty-four (85%) of the 40 subjects enrolled in the SP trial completed the 14-day follow-up period. Three subjects had parasite densities ≤ 250/µl on re-examination of their day 0 blood smears and three others were lost to follow-up on days 2, 7, and 14. Three of the 18 subjects enrolled in the MQ study did not complete their 28-day follow-up: one was found to have a parasite density ≤ 250/µl on re-examination of her initial blood smears and two were lost to follow-up, one on day 7 and one on day 14 (Table 1).

Table 1 - Parasitologic and therapeutic response of *Plasmodium falciparum* to sulfadoxine-pyrimethamine or mefloquine in Caballococha, Peru, 1999-2000.

| Drug | Parasitologic response (%)* | | | | | | | Therapeutic response (%)* | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Drug | n | RIII | RII | RI | RI/S | S | n | ETF | LTF | ACR |
| Sulfadoxine pyrimethamine | 34 | 24 | 32 | 6 | 38 | - | 34 | 18 | 41 | 41 |
| Mefloquine | 15 | 0 | 0 | 0 | 0 | 100 | 15 | 0 | 0 | 100 |

*RIII, RII, RI = sensitive (S); ETF = early treatment failure; LTF = late treatment failure; and ACR = adequate clinical response are classifications of drug efficacy (1, 4).

Nineteen (55.9%) of the 34 patients in the SP trial had RII/RIII resistance; 15 (44.1%) others were classified as RI or RI/S. Six (18%) of the subjects in the SP trial were classified as early treatment failures (ETF); 14 (41%) others had late treatment failures (LTF). None of the 15 subjects in the MQ study had a recurrence of parasitemia during their 28-day follow-up.

This is the first antimalarial drug efficacy study reported from the northeastern Peruvian Amazon region bordering Colombia and Brazil. To the best of our knowledge, no similar studies have been carried out in the Colombian Amazon region or in the area of Brazil bordering Peru. As in previous studies conducted in and around the city of Iquitos in Peru to the southwest, high levels of resistance to SP were found (RC Navitsky, GM Stennies: personal communication, 1999), and the Peruvian National Malaria Control Program now recommends MQ plus artesunate as its first-line therapy for uncomplicated *P. falciparum* malaria in this area.

Mefloquine alone, or in combination with an artemisinin drug, is commonly used for the treatment of

uncomplicated *P. falciparum* infections in the Brazilian Amazon region[3]. Sporadic *in vitro* resistance to MQ has been reported from this area since the early 1980s[5], but well-documented cases of therapeutic failures with *in vitro* resistant isolates are unusual. RI *in vivo* resistance to a single dose of 15mg/kg has only recently been recorded[2,6]. In the report by Cerutti *et al*[2], only one of 94 patients was classified as a RI failure with recurrence of symptoms and parasitemia on day 27. This patient had a sub-optimal MQ blood level combined with an increased IC50 for mefloquine in the recrudescent isolate. We found no evidence of resistance to MQ at our study site in the northeastern Peruvian Amazon region, but outside the Ministry of Health, this drug can only be found in a few pharmacies, and its cost of approximately $4.70 per tablet makes it too expensive for most patients.

In general, patterns of antimalarial drug resistance tend to be similar in contiguous geographical areas where the epidemiology of malaria is uniform and there are no major topographical barriers. This study, from a relatively isolated area of the Peruvian Amazon region, provides additional evidence for this statement and makes it likely that similar patterns of resistance will be found if studies are done on the Brazilian or Colombian sides of the border, given the frequent movement of residents along the Amazon River between Brazil, Colombia, and northeastern Peru.

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## REFERENCES

1. Bruce-Chwatt LJ, Black RH, Canfield CJ, Clyde DF, Peters W, Wernsdorfer WH. Chemotherapy of Malaria. World Health Organization Monograph Series No 27, World Health Organization, Geneva, 1986.
2. Cerutti C, Durlacher RR, Alencar FEC, Segurado AAC, Pang LW. *In vivo* efficacy of mefloquine for the treatment of falciparum malaria in Brazil. Journal of Infectious Diseases 180: 2077-2080, 1999.
3. Fundação Nacional de Saúde. Manual de Terapêutica de Malária. Ministério da Saúde. Brasilia, 2001.
4. Pan American Health Organization. Evaluation of the Therapeutic Efficacy of Drugs for the Treatment of Uncomplicated *Plasmodium falciparum* Malaria in the Americas. OPS/HCP/HCT/113/98, Washington DC, 1998.
5. Souza JM. A phase II clinical trial of mefloquine in Brazilian male subjects. Bulletin of the World Health Organization 61: 815-820, 1983.
6. Salcedo JMV, Camargo LMA, Braga MFV, Maria PS, Macêdo VO. Avaliação da eficácia e tolerância do artesunato associado á tetraciclina na terapêutica da malária falciparum. Revista da Sociedade Brasileira de Medicina Tropical 30: 251-257, 1997.
7. World Health Organization. Assessment of Therapeutic Efficacy of Antimalarial Drugs for Uncomplicated Falciparum Malaria in Areas of Intense Transmission. WHO Document, WHO/MAL/96.1077, Geneva, 1996.