

COMPARISON OF MEFLOROQUINE, CHLOROQUINE PLUS PYRIMETHAMINE-SULFADOXINE (FANSIDAR^R), AND CHLOROQUINE AS MALARIAL PROPHYLAXIS IN EASTERN THAILAND

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Abstract. From July 1983 to March 1984 a randomized double blind prophylactic trial in Thai gem miners working across the border in Cambodia was conducted to determine the prophylactic efficacy of 3 drug regimens against *P. falciparum* and *P. vivax* malaria along the Thai-Cambodian border. Gem miners have a high incidence of malaria. Maximum duration of individual participation was 14 weeks. Of 334 participants in this study who were seen every 2 weeks, 145 received mefloquine 500 mg fortnightly, 112 received chloroquine 300 mg base weekly plus Fansidar^R (1000 mg sulfadoxine and 50 mg pyrimethamine) fortnightly and 77 received chloroquine as 300 mg base weekly. The significant reduction of vivax malaria in study subjects (compared to background incidence) implied good compliance with self administration of chloroquine in the intervening weeks between scheduled appointments. The attack rate in each prophylactic regimen was 2188 cases/1000/year with mefloquine, 8338 cases/1000/year with chloroquine-Fansidar^R and 10,207 cases/1000/year receiving chloroquine alone. There was a 79% prophylactic efficacy for mefloquine and 18% efficacy for the chloroquine plus Fansidar^R regimen compared to chloroquine. Using life table analysis, 56% of the mefloquine group, 6% of the chloroquine-Fansidar^R group and 4% of the chloroquine group were malaria free at the end of the 14 weeks study. The chloroquine plus sulfadoxine-pyrimethamine regimen prescribed for prophylaxis is no longer effective for multidrug resistant strains of *P. falciparum* in the study area. This study also seriously questions the efficacy of mefloquine prophylaxis.

INTRODUCTION

In eastern Thailand falciparum malaria strains have a long history of chloroquine resistance (Harinasuta *et al*, 1965; Cadigan *et al*, 1966; Bourke *et al*, 1966). Within the past year mefloquine has been recommended for falciparum malaria prophylaxis in drug resistant areas such as Thailand (Bradley and Philips-Howard, 1989; Centers for Disease Control, 1990), replacing previously recommended regimens of either Maloprim^R (pyrimethamine plus dapsone) (Cook, 1988) or doxycycline. Mefloquine is recommended as 250mg weekly for four weeks starting one to two weeks prior to exposure then switching to fortnightly doses thereafter (Centers for Disease Control, 1990). Though this study is somewhat dated we

believe it raises questions about the current efficacy of both mefloquine and pyrimethamine plus sulfonamide regimens and underscores the need to base prophylactic regimens on results of recent trials in areas of rapidly developing parasite resistance.

In 1977 Pearlman performed a large prophylactic trial testing both Fansidar^R (pyrimethamine plus sulfadoxine) and mefloquine as malaria prophylaxis in eastern Thailand (Pearlman *et al*, 1980). He showed that compared to a control group administered placebo, two regimens of Fansidar^R, either 1000 mg sulfadoxine plus 50 mg pyrimethamine fortnightly or 500 mg sulfadoxine plus 25 mg pyrimethamine weekly, had 97-98% efficacy for falciparum malaria prophylaxis. Similarly two regimens of mefloquine, 180 mg weekly or 360 mg fortnightly, each had an efficacy greater than 99%.

This study was prompted by decreasing drug sensitivity patterns in Thailand subsequent to

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Pearlman's study and high attack rates of falciparum malaria in Thai military stationed along the Thai-Cambodian border taking weekly chloroquine plus Fansidar^R or chloroquine plus Maloprim^R. Since the completion of Pearlman's project, we feel our study is the only one with comparable methodology and high enough attack rates to give a valid comparison, examining for increasing resistance of *P. falciparum*. The objectives of this study were to determine the prophylactic efficacy of mefloquine as 500 mg every two weeks, to determine if the addition of Fansidar^R to chloroquine increased its prophylactic efficacy and to determine the tolerance and toxicity of the three prophylactic drug regimens. Because of a new tablet formulation we were forced to administer 500 mg of mefloquine fortnightly instead of 360 mg as used by Pearlman.

METHODS

Location and study population

The town of Bo Rai, located in Trat Province 350 km southeast of Bangkok and areas across the Cambodian border within 10 km east from Bo Rai were the sites for this study. The study was conducted from July 1983 through March 1984. All participants were gem miners working in Cambodia for greater than 3 months duration and had the intention of remaining in the area for 6 months. This group of miners came from distant areas of Thailand, often without prior malaria exposure. All participants were required to have a negative malaria smear (after examination of 200 fields on thick smear) on entry into the study. Patients who desired prophylaxis were referred to the study by the government malaria clinics initially and by other study participants later in the course of the study.

Only males 21 years of age or over were accepted. Each had the study objectives, risks and benefits explained and signed an informed consent form. This study was approved by the Human Subject Review Board of the US Army Surgeon General's Office and the Thai Ministry of Health Ethical Review Committee.

Study design

Assignment to one of three prophylactic groups was randomized and double blinded. Assignment

to mefloquine, chloroquine plus Fansidar^R and chloroquine groups was in a 4:3:2 ratio, so that regimens expected to be more efficacious would have larger groups of patients. All participants were seen every 2 weeks and oral administration of medication was witnessed by one of the investigators. At each 2 week visit malaria films (thick and thin smears) were performed, travel history noted and history of symptoms over the previous fortnight was obtained. Patients were asked about fever, chills, headache, nausea, vomiting, diarrhea, anorexia, rash, myalgia and dysuria or abnormally-colored urine.

Any subject missing one appointment was excluded from the study though each subject's records up to the time of exclusion were entered into the survival analysis. Those patients found positive for malaria on thick smear were placed on treatment consisting of: quinine 650 mg three times daily \times 3 days plus tetracycline 500 mg three times daily \times 7 days for *P. falciparum* malaria and chloroquine phosphate 1500 mg over 48 hours plus primaquine 15 mg per day for 14 days for vivax malaria. After 3 weeks post treatment and a negative malaria smear some patients wishing to continue were reentered under a new study number and were assigned a double blind randomized treatment.

Laboratory studies

On entry each subject had blood drawn for hematocrit, complete blood count, transaminase levels, total and direct bilirubin, alkaline phosphatase, blood urea nitrogen, serum quinine and sulfa levels. A second blood sample was drawn on subjects who remained malaria free after six weeks of medication administration to monitor for any hematologic, hepatic or renal drug toxicity.

These plasma samples were also analyzed for mefloquine levels by the method of Kapetanovic *et al* (1983).

Administration of medications

Every two weeks in a double blind fashion one of the investigators administered five tablets to each subject: One group received two mefloquine tablets (500 mg) plus one chloroquine and two Fansidar^R placebos. A second group received one chloroquine (300 mg base) plus two Fansidar^R tablets and two mefloquine placebos. A third group received one chloroquine tablet plus two

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Fansidar^R and two mefloquine placebos. In addition all subjects were given packets and instructed to self administer 1 tablet weekly between visits. This tablet consisted of chloroquine in the chloroquine or chloroquine-Fansidar^R groups and a matched placebo in the mefloquine group.

Source of medication

The mefloquine used was 250 mg tablets manufactured for the US Army by LaFayette Pharmacal Batch# E 598 (Lot #WRA 140,7153). Mefloquine placebos were Lot #WRA 130413. Chloroquine tablets 'Aralen' 500 mg (Lot 491LD) and chloroquine placebos (Lot #R021CB) were manufactured by Winthrop. Fansidar^R tablets (Lot #B3712U5020) and Fansidar^R placebos (Lot #GET 404 A01U5020) were supplied by the Hoffman-La Roche Corporation.

Analysis of data

Each patient's time contribution to the study was tallied and used in survival analysis of the data. The use of other antimalarials or antibiotics by study-subjects was reason for exclusion from the study. Patients who were removed from the study had their participation counted for compilation by survival analysis (% of group remaining malaria free over time).

Comparability of groups based on the number of previous malaria infections, age and weight was tested by analysis of variance. The biochemical and hematologic parameters before and after 6 weeks of prophylaxis were compared by paired *t* test. Differences in symptoms while on prophylaxis were examined by contingency tables for the 3 groups. Efficacy of prophylaxis was assessed by survival analysis (Breslow, 1970). Estimates of relative prophylactic efficacy was calculated based on comparisons of the number of falciparum malaria breakthroughs per man-fortnightly period of participation in study (proportion of positive smears from fortnightly visits). Taylor series confidence intervals for the relative risks were used to calculate confidence intervals for relative efficacies.

RESULTS

Comparability of drug groups

At the onset, there were no significant differences in age, weight or prior malaria experience among the three drug groups.

Of the 334 total participants over an 8 month duration, the number infected in each prophylactic regimen and the calculated attack rates are displayed below:

	Chloroquine +		
	Chloroquine	Fansidar	Mefloquine
Number in group	77	112	145
Mean time in study (weeks)	3.49	4.21	6.23
Number <i>P. falciparum</i> cases	53	76	38
Attack rate (cases/1000/year)	10,207	8,338	2,188
Man-weeks in study	269	471	903

Calculating prophylactic efficacy, mefloquine reduces the *P. falciparum* attack rate by 79% (95% confidence interval 70%-85%), while chloroquine Fansidar^R produces a 18% (95% confidence interval 0%-38%) reduction. Chloroquine was assumed to be ineffective against *P. falciparum* and was used as a control group.

Participants were defined as individuals who were negative on enrollment and returned for at least one follow-up appointment. Therefore of the original 501 enrollees, 63 were discarded due to positivity at week 0 and 104 were discarded since they never returned beyond week 0.

Of the positive patients during the course of the study only 2 cases of *P. vivax* were detected. The vast majority of cases were *P. falciparum* infections. The background rates of *P. vivax* in the gem mining community during the study ranged from 15-20%. The absence of *P. vivax* in the study patients indicated compliance with the every other week self-administered chloroquine prophylaxis (in the chloroquine and the chloroquine plus Fansidar^R groups). Table 1 displays the distribution of malaria cases throughout the study. Only 194 patients completed the study until positivity or end of the 14 weeks observation period. Adjusting for subjects leaving the study because of falciparum malaria, the attrition rates among the groups were comparable ($p = .19$, survival analysis, Breslow statistic).

A survival curve (% of group malaria free) was graphed from each of the individual time-in-study contributions. At the end of the study mefloquine prophylaxis resulted in 56% malaria free, chloroquine-Fansidar^R had 6% noninfected and chloroquine prophylaxis had 4% noninfected with malaria

Table 1
 Number of subjects developing *P. falciparum* malaria in each week of study.

	Chloroquine n = 77	Chloroquine plus Fansidar ^R n = 112	Mefloquine n = 145
Week 0 *	15	20	28
Week 1	4	1	2
Week 2	25	40	14
Week 3	1	3	0
Week 4	11	11	8
Week 5	2	0	0
Week 6	4	7	3
Week 7	2	0	0
Week 8	2	5	3
Week 9	0	0	0
Week 10	1	5	3
Week 11	1	0	0
Week 12	0	4	3
Week 13	0	0	0
Week 14	0	0	2
Total	53	76	38

* Patients admitted to study whose pre-dose smear was later found to be positive. These patients were excluded from efficacy calculations.

(Fig 1). Survival analysis shows mefloquine's superiority to either chloroquine or chloroquine plus Fansidar^R regimen ($p < .0001$, Breslow statistic) and no benefit of using chloroquine plus Fansidar^R instead of chloroquine alone ($p = .34$, Breslow statistic).

Symptomatology on prophylaxis

Symptoms at each patient visit were compared for those patients with negative malaria smears throughout the study, to avoid confusing malaria symptoms with drug side effects. There were no significant differences in frequency of complaints among the study groups for headache, anorexia, fever, chills, nausea, diarrhea, or vomiting (chi-square or Fisher's exact test). The chloroquine, chloroquine plus Fansidar^R, and mefloquine groups reported myalgias during 49%, 32%, and 24% of their visits, respectively ($p < .005$, chi-square) and rashes during 9%, 14%, and 5%, respectively ($p < .01$, chi-square).

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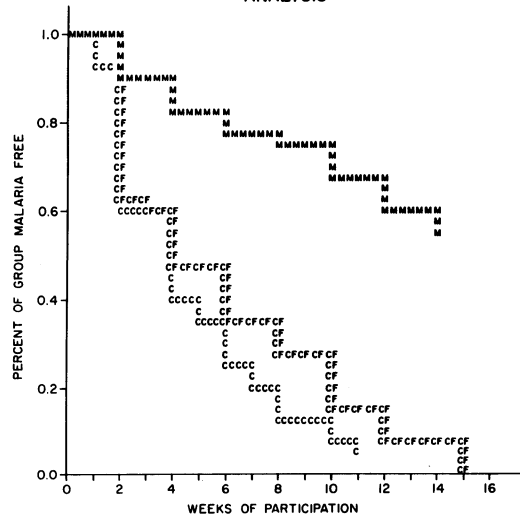


Fig 1—Survival curves of study groups administered mefloquine (M), chloroquine plus Fansidar^R (CF), or chloroquine (C). Curves represent cumulative proportions of each group remaining malaria free as a function of time (units of man-weeks) in study.

Laboratory abnormalities

For subjects not contracting malaria, blood samples were drawn prior to and six weeks following administration of study medications in 21, 10, and 60 subjects administered chloroquine, chloroquine plus Fansidar^R and mefloquine, respectively. For each drug group there were no significant changes seen in total bilirubin, transaminase levels, alkaline phosphatase, blood urea nitrogen, platelet count, or white blood cell count. There was a significant improvement in hematocrit levels in both the chloroquine plus Fansidar^R and the mefloquine groups ($p < .01$, paired *t*-test).

Patients in the mefloquine drug group had an elevated alanine aminotransferase and alkaline phosphatase ($p < .05$ by paired *t*-test) when the six week biochemistry values were compared to the values at zero time. This analysis of toxicity incorporated all changes, including those in the normal range. A rise in liver enzymes was not seen in the chloroquine or chloroquine-Fansidar^R groups although the bilirubin was increased at 6 weeks in the chloroquine patients. Examining only clinically abnormal elevations of liver enzymes and total bilirubin, again the mefloquine group showed increase over baseline levels.

DISCUSSION

A perplexing finding of this study was the large number of prophylactic failures on 500 mg mefloquine administered every two weeks. This finding could be interpreted in one of three ways: (1) Rapidly increasing resistance to mefloquine since 1977 has developed. (2) Poor absorption of the larger 500 mg dose of mefloquine occurred with an inadequate blood level sustained for a 14 day period. (3) Rare parasitemia (less than one parasite per 200 oil immersion fields) is within the scope of drug action and is an observed phenomenon following mefloquine treatment or prophylaxis, which can be detected if subjects are adequately examined and may represent unviable forms. Rare parasitemias which clear spontaneously have been reported in Thailand (World Health Organization, 1984) and Burma (Tin *et al*, 1982) during large scale treatment trials with mefloquine. Ninety-five percent of all the mefloquine failures in this prophylactic trial presented with rare parasitemia

counts compared to 35-50% of breakthroughs on chloroquine or chloroquine-Fansidar^R with rare readings. These latter two study groups had significantly higher parasitemias at the time of prophylactic failure. It is the authors' opinion that these rare parasitemias often cleared without treatment in these patients groups due to the prevalence of partial protection or semi-immunity in these persons living in an area of intense malaria transmission. Non-immune individuals might be unable to contain the parasitemia.

The low prophylactic efficacy seen in this study among gem miners might be explained by the inability to maintain protective drug levels resulting from a fortnightly dosing schedule or a difference in mefloquine sensitivities of *P. falciparum* strains. It should be noted that in Pearlman's study in 1977 where mefloquine efficacy was high, smears were examined by the same group of technicians and under the same method as in this study. Furthermore lower fortnightly doses were used. The subjects in his study probably had lower levels of immunity than our study subjects, arguing against lower immunity as a cause of the poorer efficacy seen in our study. It should be pointed out that Pearlman's study used a control group administered placebo whereas ours was administered chloroquine. It is hypothetically possible that had we used a placebo control group we would have shown a higher efficacy for mefloquine. In this area we believe the efficacy of chloroquine prophylaxis at the time of the study was negligible, certainly much less than the 18% shown for Fansidar^R. Chloroquine was used in the control group so that subjects would not be dropped from the study because of contracting vivax malaria.

Elevations of liver enzymes occurred following every 2 week administration of 500 mg mefloquine for 6 weeks duration. This toxicity was not detected in 250 mg weekly prophylactic doses of mefloquine. Clinical side effects were uncommon in the mefloquine group. The chloroquine group experienced myalgias and along with the chloroquine-Fansidar^R group a greater incidence of rash. There was no difference in gastrointestinal symptomatology.

Mefloquine for multi-drug resistant *P. falciparum* malaria surpasses chloroquine-Fansidar^R or chloroquine. The prophylactic performance of chloroquine and of the combination of chloroquine plus Fansidar^R are both poor, chloroquine protected

4% of the group, and chloroquine plus Fansidar^R protected 6% of the group over a 14 week period in an area of intense transmission. There was no difference in the protection between the chloroquine or chloroquine-Fansidar^R groups.

Fansidar^R contributes little to chloroquine in prophylactic efficacy. Reports of toxic reactions such as Stevens-Johnson Syndrome and toxic epidermal necrolysis, (Centers for Disease Control, 1985; Rombo *et al*, 1985; Vestergaard-Olsen *et al*, 1982; Setia, 1983) with the use of Fansidar^R and chloroquine in combination, together with *in vitro* studies documenting high level Fansidar^R resistance in Thailand to both components of the drug combination, (Childs *et al*, 1986) challenge the rationale of using this drug or similar pyrimethamine plus sulfa regimens. Based on poor efficacy, this study demonstrates that these combinations have no prophylactic indication in Thailand.

Between 1977 and 1984 the prophylactic efficacy of mefloquine has decreased. It should be noted that this study used a higher dose than Pearlman did in 1977 but that our control group was administered chloroquine rather than placebo. It is difficult for us to predict on the basis of this study the efficacy of the currently recommended mefloquine prophylactic regimen (Centers for Disease Control, 1990). One difference is that the current recommendation starts the regimen one or two weeks prior to exposure, ensuring a higher drug level at the onset of exposure (half-life of mefloquine approximately 18 days). However the survival analysis curve for this study shows similar rates of failure over the entire 14 week period for the mefloquine group, indicating that failures were not attributable only to our lack of a "loading dose" but occurred even at higher steady state levels. In fact, examining the attack rates in the other two study groups the best protective effect (largest difference in attack rates) of mefloquine was seen during the first few weeks of subjects entering the study. Another important point is that this study was performed in individuals with some degree of immunity while the prophylactic recommendation is geared towards non-immunes, in whom efficacy may be worse. When designing this study we had hoped to show poor efficacy of Fansidar^R with mefloquine serving as the "gold standard". The decreased efficacy of mefloquine was somewhat alarming. Since mefloquine was not recommended for prophylaxis at that time (1984) we felt our

efforts would be better used looking for more effective alternatives (doxycycline and proguanil plus sulfa). Now that mefloquine has been recommended we regret that we failed to carry out further prophylactic studies to monitor what appears to be increasing resistance to this drug. The increasing number of mefloquine treatment failures recently reported (Thai Ministry of Health, personal communication) indicates that such a study is overdue.

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