A COMPARATIVE STUDY OF ARTESUNATE AND ARTEMETHER IN COMBINATION WITH MEFLOQUINE ON MULTIDRUG RESISTANT FALCIPARUM MALARIA IN EASTERN THAILAND

Krongthong Thimasarn[1], Jeeraphat Sirichaisinthop[2], Pramuan Chanyakhun[3], Chatchai Palananth[4] and William Rooney[1]

[1]The Malaria Division, Department of Communicable Disease Control, Ministry of Public Health, Bangkok; [2]Center for Malaria Region 1, Saraburi Province; [3]Malaria Unit 56, Trat Province; [4] Malaria Unit 57, Chanthaburi Province, Thailand

Abstract. *Plasmodium falciparum* in Thailand is highly resistant to chloroquine, sulfadoxine-pyrimethamine and there is increasing resistance to quinine and mefloquine. The use of qinghaosu derivatives alone or in combination with mefloquine has been shown successfully effective against multi-drug resistant *P. falciparum* in many clinical trials. However their applications with ambulatory treatment should be assessed. 394 uncomplicated falciparum malaria cases studied at Trat and Chanthaburi malaria clinics, eastern Thailand, were allocated at random to receive either one of the seven following regimens: A) artesunate 600 mg over 2 days and mefloquine 1,250 mg in divided doses. B) artemether 640 mg over 2 days and mefloquine 1,250 mg in divided doses. C) artesunate alone 700 mg over 5 days period. D) artemether alone 800 mg over 5 days period. E) quinine plus tetracycline for 7 days. F) mefloquine 1,250 mg in divided doses and G) artesunate 600 mg over 2 days period and mefloquine 750 mg. The follow-up was on Days 1, 2, 7, 14, 21 and 28. Patients tolerated all regimens very well and there was no serious side effects. The adverse effects did not differ among the seven regimens. The cure rates were 98.7, 97.1, 97.9, 96.7, 92.3, 100 and 95.2%, respectively. There was no significant difference of cure rates among various regimens. A total of 16 *P. vivax* and 1 *P. malariae* reinfections were reported among the study groups during the secnd half of the follow-up period, 14 of which were from the groups administered short action drugs (artesunate, artemether or quinine). The results suggested that either artesunate 600 mg or artemether 640 mg in combination with mefloquine 1,250 mg over a period of two days should be considered as alternative regimens for treating uncomplicated multi-drug resistant falciparum malaria.

INTRODUCTION

Plasmodium falciparum in Thailand is highly resistant to chloroquine (Bunnag *et al*, 1986), sulfadoxine-pyrimethamine and amodiaquine (Pinichpongse *et al*, 1982). There is increasing resistance to alternative antimalarials: quinine (Bunnag *et al*, 1987), mefloquine (Ketrangsee *et al*, 1992) and mefloquine/sulfadoxine/pyrimethamine (Thimasarn *et al*, 1995) especially along the Thai-Cambodian and Thai-Myanmar borders. The combination of quinine and tetracycline, the current regimen, which is now being administered for mefloquine failure cases, is effective but compliance is a problem faced by malaria clinics at the field level.

Alternative drug combinations are urgently needed for the treatment of drug resistant strains of falciparum malaria and to improve the patient compliance, *ie* such regimens need to be effective and with a short treatment course.

Artesunate and artemether (qinghaosu derivatives) are effective antimalarial drugs with a rapid onset of action that destroys asexual parasites at an early stage of development. The efficacy of these drugs has been shown in many clinical trials in China, Myanmar and Thailand (Guoqiao *et al*, 1982; Jiang *et al*, 1982; Bunnag *et al*, 1991a). Artesunate and artemether rapidly cleared parasites with virtually no side-effects, but the treatment needs to be at least 5 days. Although artesunate and artemether are promising in the treatment of multiple drug resistant parasites, with a long course of treatment, they unlikely to be successful with ambulatory treatment.

Mefloquine is an antimalarial with long half-life (Karbwang *et al*, 1990; Desjardin, 1979; Schwartz, 1982), that has been recommended as an oral single dose for the treatment of falciparum malaria (WHO, 1984). An effective short treatment course is possible with the combination of either artesunate or artemether with mefloquine. As mefloquine has been reported to act at the large ring and early

growing trophozoite stages, a later action than that of artemether (Guoqiao *et al*, 1984), the potential usage of such combinations would be high if their efficacies are evident. Special reference is made to the usefulness of these combinations for patients at peripheral levels where the early management of severe falciparum malaria is generally impossible.

The role of artemether and artesunate is to prevent the development of the parasites to the more pathological phase of infection , *ie* severe malaria, while mefloquine would prevent recrudescence. It has been shown previously that the combination of mefloquine, sulfadoxine-pyrimethamine and qinghaosu in the treatment of uncomplicated falciparum malaria resulted in radical cure (Guoqiao *et al*, 1984). In this context we compared the efficacy of mefloquine alone and various combinations of mefloquine with artesunate or artemether in the treatment of patients having uncomplicated multidrug resistant falciparum malaria. The outcome of the study made available base line information on future alternative regimens for operational use by the Malaria Control Program.

The objectives of the study were:

1) To assess the efficacy of oral artesunate and artemether in combination with mefloquine in the treatment of uncomplicated falciparum malaria in an area where mefloquine resistance is reported to occur.

2) To compare the tolerance of mefloquine alone and to that of artesunate or artemether when either one is combined with mefloquine.

MATERIALS AND METHODS

The study was conducted during July 1993-December 1994 in four malaria clinics in Bo Rai, Trat Province and Pong Numron, Chanthaburi Province (Fig 1) situated along the Thai-Cambodian Border where falciparum multidrug resistant malaria is known to occur. The four malaria clinics being located in the same locality where parasites are presumed belonging to similar strains.

Study population

The criteria to enroll patients were as follows:

1. Symptomatic adult individuals having contracted *P. falciparum* from Trat and Chanthaburi Provinces or across the international border in Cambodia, within 50 km radius from the malaria clinics.

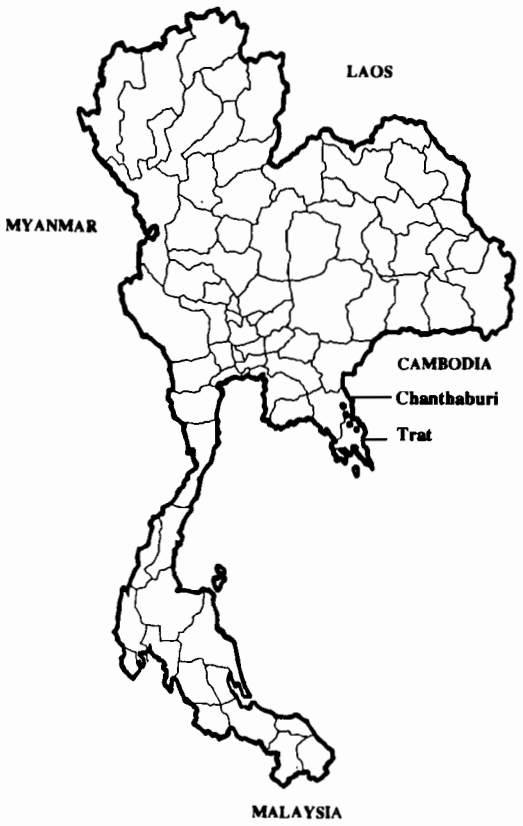

Fig 1--Map of Thailand showing study sites.

2. The asexual parasitemia range 500-400,000 per μl blood.

3. The general conditions of patients well, no sign of complications.

4. Adult males and females over 15 years included, pregnant women excluded.

5. The patients had no history of antimalarial drug intake over the past 2 weeks prior to the study. (Urine examination to detect antimalarial drugs was not carried out).

6. The patients agreed to remain in malaria free area for the whole period of 4 weeks, and agreed to return for post treatment checking and signed the consent forms.

Sample size and patient allocation

Patients were randomly allocated to the following therapeutic regimens. All four malaria clinics had fixed quota sample size, depending on their average annual patient attendance.

Group A: Initial dosage of artesunate 300 mg to be followed by mefloquine 750 mg after 6 hours interval. Patients were given take home treatment of artesunate 300 mg to be taken the following morning with a further mefloquine 500 mg after a 6 hours interval (total mefloquine 1,250 mg, artesunate 600 mg).

Group B: Initial dosage of artemether 320 mg to be followed by mefloquine 750 mg after 6 hours interval. Patients were given to take home artemether 320 mg to be taken the following morning with mefloquine 500 mg after a 6 hours interval (total mefloquine 1,250 mg, artemether 640 mg).

Group C: Artesunate 300 mg initial dose followed by 100 mg daily for 4 days (total 700 mg).

Group D: Artemether 320 mg initial dose followed by 120 mg daily for 4 days (total 800 mg).

Group E: Quinine 600 mg 3 times daily plus tetracycline 500 mg 2 times daily for 7 days.

Group F: Mefloquine 750 mg initial dose followed by a further 500 mg the following morning (total 1,250 mg).

Group G: Artesunate 300 mg initial dose followed by mefloquine 750 mg 1 hour later. Patients were given to take home artesunate 300 mg to be taken the following morning (total artesunate 600 mg, mefloquine 750 mg).

(Artesunate 50 mg tablet, Guilin company, China)

(Artemether 40 mg capsule, Kunming company, China)

(Mefloquine 250 mg tablet, Lariam®, Hoffman La Roche, Basel, Switzerland).

Post treatment follow-up

This study was a field-orientated trial. Each patient was observed at his/her intake of the first dosage of regimen and requested to remain a few hours at the clinics in order to observe, as a precaution, any possible side effects and vomiting.

It was not possible to ensure complete patient compliance in drug intake, however, the malaria clinics were requested to determine the intake together with history of possible entry into malaria transmission areas, with particular attention to cases that exhibit recrudescence from Day 7 onward.

Patients were asked to return for blood examination on Days 1, 2, 7, 14, 21 and 28 (Day 0 = first day of treatment).

Treatment for recrudescence

All recrudescent cases whose blood showed no decrease or increased in parasitemia on Day 2 or remained parasitemic up to the Day 7 or those giving a recrudescence during the follow-up period were retreated, under supervision, with quinine/tetracycline for 7 days (as Regimen E) and the study was concluded.

Quality control of blood film examination

All blood slides examined by trained malaria clinic workers were collected and rechecked by skilled microscopists at the Regional laboratory, Center for Malaria Region 5, Nonthaburi.

Statistical analysis

Differences in proportion were analyzed using chi-square and Fisher's exact tests.

RESULTS

394 cases were initially enroled in the study, of which 12 were excluded due to treatment regimen error. 51.2% contracted malaria from Trat and Chanthaburi Provinces while 47% contracted malaria from the adjacent areas in Cambodia; 1.8% were of unknown origin. 382 cases were included for final analysis of which 189 were from malaria clinics in Bo Rai, Trat Province, while 194 were from Pong Num Ron, Chanthaburi Province. 88.8% were adult males with mean age equal to 30.3 ± 11.2, range 15 to 70 years. Initial parasite density

prior to treatment was 26,264 ± 41,890, range 576-311,200 per μl (Table 1).

Results of treatment

Cure rates in all treatment groups were higher than 90% (Table 2). All cases giving a recrudescence were of an RI responses with the exception of a single RII case in Group E (Q7T7). A total of 16 *P. vivax* and 1 *P. malariae* infections were reported among the study groups during the second half of the follow-up period. These cases were excluded from the final data analysis shown in Tables 1 and

Table 1

Patient biodata.

Regimen	No.	Age (yr)	Initial parasite density (per μl) X ± SD	range
A	80	30.4 ± 9.8	29,007 ± 36,398	640-278,400
B	45	27.4 ± 9.2	24,506 ± 45,147	680-296,000
C	55	27.9 ± 10.8	20,445 ± 25,945	600-151,600
D	42	30.9 ± 10.8	16,155 ± 17,970	576-78,872
E	47	30.1 ± 11.4	17,736 ± 29,034	640-144,800
F	45	33.3 ± 13.4	23,696 ± 48,648	624-280,480
G	68	31.8 ± 12.6	42,703 ± 61,471	600-311,200
Total	382	30.3 ± 11.2	26,264 ± 41,890	576-311,200

Table 2

Results of treatment by various regimens.

Regimen	N	Response						Cure rate* on Day 28 (%)
		S	RI	RII	RIII	S/RI	U	
A	80	75	1	0	0	2	2	98.7
B	45	34	1	0	0	5	5	97.1
C	55	46	1	0	0	1	7	97.9
D	42	30	1	0	0	3	8	96.7
E	47	36	2	1	0	2	6	92.3**
F	45	41	0	0	0	2	2	100
G	68	59	3	0	0	2	4	95.2***

*Cure rate = $\frac{S}{N\text{-}S/RI\text{-}U} \times 100$

**E versus F, p = 0.59261; ***F versus G, p = 0.42619

Remarks : Evaluation of treatment results

S = Blood slides were negative for asexual forms from Day 7 to Day 28

RI = Initial disappearance of parasitemia followed by recrudescence within 28 days.

RII = Parasitemia reduction (Asexual parasitemia of *less than* 25% of pretreatment count on Day 2) but did not disappear within 7 days.

RIII = No significant reduction (Asexual parasitemia of *equal or more than* 25% of pretreatment count on Day 2) of parasitemia within 7 days.

S/RI = Blood slides were negative on Day 7 and patients dropped out from the study before Day 28.

U = Unclassified results of treatment due to reinfection by other malaria species during study period and other reasons.

Table 3

Proportions of patients having various side effects.

Regimen	No.	Nausea*	Vomit	Abd	Diarrh	Headache	Skin	CVS	CNS	Others
A	73	28.7	19.1	12.3	10.96	32.9	-	1.4	24.7	5.5
B	45	22.2	13.3	17.8	11.1	28.9	6.7	2.2	26.7	8.9
C	55	20.0	7.3	9.1	3.6	21.8	3.6	1.8	18.2	5.4
D	42	11.9	7.1	11.9	7.1	47.6	-	2.4	16.6	7.1
E	47	21.3	10.6	2.1	-	36.2	4.3	-	14.9	8.5
F	47	38.3	8.5	12.8	6.4	44.7	-	-	19.2	-
G	68	20.6	5.9	5.9	4.4	42.6	-	-	8.8	-
B mixed	12	41.7	25.0	25.0	8.3	41.7	-	-	25.0	25.0

*$X^2 = 12.72$; p-value = 0.0792

2. It was noted that most of these infections were from the groups administered short action drugs (4 cases with artesunate alone, 6 cases with artemether alone, 4 cases with Q7T7, and one each in Groups A, F and G).

Side effects

The side effects of medications as reported on Day 1 were analyzed (Table 3). Patients tolerated all regimens very well. Headache and nausea were common symptoms in all treatment groups and were probably attributed to the disease itself. Gastrointestinal symptoms were observed in all treatment groups. Patients who received mefloquine and quinine did not have any more GI side effects than the others who received artesunate or artemether alone. Patients from Group F who received mefloquine 1,250 mg in divided doses had more nausea than the others but there is no statistically significant difference. Palpitation and mild skin rash were reported in a few cases in both mono drug and combinations. Insomnia was reported in all treatment groups.

All side effects were mild and self recovering within 7 days. There were 12 patients who unintentionally received mixed drugs, artesunate and artemether, due to technical error, had a little higher rates of GI symptoms and all had either sensitive or S/RI responses.

DISCUSSION

The study was carried out in the area located on the Thai-Cambodian border, termed the epicenter of *P. falciparum* multidrug resistance in Asia. Mefloquine 15 mg/kg body weight alone or in combination with sulfadoxine/pyrimethamine failed to cure patients with falciparum malaria in this area and also on the western border with Myanmar, the similarity considered due to population migration (Thimasarn *et al*, 1995; Fontanet *et al*, 1993). Many clinical trials have confirmed that artesunate and artemether were effective against acute uncomplicated falciparum infections patients in the Hospital for Tropical Diseases, Bangkok (Bunnag *et al*, 1991a, b). Unfortunately, artesunate and artemether need to be administered for at least 5-7 days if they were used alone (Bunnag *et al*, 1991b). Combination of oral artesunate followed by long-acting drugs such as mefloquine yielded high cure rates (Looareesuwan *et al*, 1992), a regimen however not practical for use within the Malaria Control Program where patients are treated on OPD basis. High dose of artesunate up to 800 mg over two day period followed by mefloquine 15 mg/kg body weight was effective (cure rate over 90%) and well tolerated (Looareesuwan *et al*, 1994). A study with oral artesunate 700 mg over 5 days was shown to be effective and better tolerated than the combination of quinine-tetracycline given for 7 days (Karbwang *et al*, 1994).

Field trials conducted in Chanthaburi, east Thailand, showed that a single oral dose of either artesunate or artemether improved cure rates of mefloquine when used for the treatment of multidrug resistant falciparum malaria (Bunnag *et al*, 1995, 1996).

The study faced many limitations: patient drop

out that caused uneven distribution of cases in each group, uneven dose of artesunate and artemether groups due to nonavailability of 50 mg tablet artemether, long period of study due to decreasing trend of malaria incidence and other operational difficulties.

In the present study although there were no differences of cure rates among various regimens, it was noted that those regimens that contained mefloquine seemed to prevent other infections appearing (*P. vivax* and *P. malariae*) during the follow-up observation period somewhat better than those without mefloquine. The appearance of infections of other species during the period of study, however, has given indication of a relatively high degree of transmission in these areas and shows the difficulty to prevent reinfection under such field conditions. A point which needs explanation is that related to the administration of mefloquine together with either artesunate or artemether rather than sequential combination of these drugs followed by mefloquine. Artemisinin derivatives have been claimed to interfere with blood levels of mefloquine (Karbwang, personal communication) and, therefore, should be taken prior to the administration of mefloquine. The Control Program aims for the highest possible cure rates while restricting the amount of drugs to be taken home by patients thus ensuring compliance of the full regimen, and also to prevent possible distribution of strict government controlled antimalarial drugs into the community. Although mefloquine blood levels were not measured we considered that the initial dosage of artesunate and artemether given prior to the first dose of mefloquine would substantially reduce parasitemia and therefore patients would tolerate mefloquine better than those who received mefloquine as a mono-drug treatment.

In conclusion, we therefore recommend that either artesunate 600 mg or artemether 640 mg, in combination with mefloquine 1,250 mg over a period of two days be considered as alternative regimens for treating uncomplicated multidrug resistant falciparum malaria.

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