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Antimalarial Effects of Rifampin in *Plasmodium vivax* Malaria

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The antimalarial effects of rifampin in 60 adults with *Plasmodium vivax* malaria were assessed. There were three treatment groups: rifampin (20 and 15 mg/kg of body weight per day for 1 and 4 days, respectively; n = 5); rifampin followed by primaquine (15 mg of base per day for 14 days; n = 25), and chloroquine (25 mg of base per kg over 3 days) followed by primaquine (n = 30). All patients were hospitalized till clearance of fever and parasites, and 45 patients stayed in the hospital for 1 month. Despite initial clearance of fever in all patients and a ≥6-fold reduction in parasitemia per 48-h life cycle, rifampin alone was not effective: all five patients had subsequent R2-like parasitological responses. All patients treated with rifampin-primaquine cleared both fever and parasitemia, but the therapeutic responses were slower than those following treatment with chloroquine-primaquine. Final fever clearance times were significantly longer (mean [standard deviation] = 43 [35] versus 27 [19] h; P = 0.046), and the parasite clearance times (to 50 and 90% of admission parasite counts and to a level undetectable in a peripheral blood smear) were also significantly greater (P = 0.053 to <0.001). However, reappearance of infection occurred in only one patient treated with rifampin-primaquine. The results of this study suggest that rifampin at the usual therapeutic doses has partial activity against blood stages of *P. vivax* in humans but that used alone it is insufficient for cure. Rifampin might therefore be of value in combination antimalarial therapy.

Several antibiotics have antimalarial activity in vitro and in vivo (3, 4, 6, 11). The tetracycline group is now used widely in combination with quinoline drugs in treatment, and doxycycline alone or in combination is used in prophylaxis. Clindamycin has also proved of value in South America (9). Multidrug resistance in *Plasmodium falciparum* continues to increase in many tropical areas (7, 12), while *Plasmodium vivax* in some parts of Indonesia has become resistant to chloroquine (2, 15). Fortunately, cure rates with a 7-day quinine-and-tetracycline regimen in uncomplicated falciparum malaria remain above 90% in Thailand, where multidrug resistance is a particular problem (7, 10, 14). But there are limited options for the future, and new antimalarial drugs are needed. Rifampin has antimalarial activity both in vitro (6, 16) and in vivo in murine malaria (1, 16). The present study was conducted to evaluate the antimalarial effect of rifampin in humans. Patients with *P. vivax* malaria were studied in this pilot investigation because the risks of possible inadequate treatment are acceptable, as the infection is nearly always benign.

MATERIALS AND METHODS

Patients. The study was conducted with adult male patients with *P. vivax* malaria admitted to the Bangkok Hospital for Tropical Diseases, Thailand, during 1992 and 1993. Informed consent was obtained from each subject before recruitment. Exclusion criteria were a known allergy to one of the study drugs, a history of taking any antimalarial drugs within the previous 48 h, admission urine that was positive for sulfonamides (lignin test) or 4-aminoquinolines (Wilson Edeson test), or glucose-6-phosphate dehydrogenase deficiency. The study was approved by the ethics committee of the Faculty of Tropical Medicine, Mahidol University, Bangkok, Thailand.

Management. After clinical assessment and confirmation of the diagnosis, baseline blood samples were taken for routine hematological and biochemical tests. Patients were then randomized to three different oral treatment groups. These were a group which received rifampin alone, one which received sequential rifampin and primaquine, and a control group which received standard treatment with sequential chloroquine and primaquine. Details concerning part of the control group (n = 25) will be reported elsewhere (13). Rifampin (Merrell Dow Pharmaceuticals Inc.; 300 mg per tablet) was given orally as one dose as close as possible to 20 mg/kg of body weight per day followed by 15 mg/kg/day for the next 4 days. Primaquine (Thai Government Pharmaceutical Organization; 7.5 mg of base per tablet) was given at a dose of 15 mg of base per day for 14 days at the end of the rifampin course. Chloroquine phosphate (Thai Government Pharmaceutical Organization; 300 or 450 mg of base) was given at a dose of 10 mg (base)/kg followed every 8 h by 5 mg/kg for three doses. Oral acetaminophen (0.5 to 1 g) was given if fever was ≥38°C. Vital signs were recorded every 4 h until resolution of fever and thereafter every 6 to 12 h. Fever clearance time was the time taken for body temperature to fall below 37.5°C and to remain below this value for >24 h.

Laboratory investigations. Parasite counts were performed every 12 h till clearance and daily thereafter. Counts were expressed as numbers of parasites per μl of blood and were calculated from the numbers of parasitized cells per 1,000 erythrocytes in a thin film stained with Giemsa's or Field's stain or per 200 leukocytes in a thick film. Parasite clearance times were calculated as the times from the start of antimalarial treatment until the asexual malaria parasite counts fell to 50% (PCT_{50}) and to 90% (PCT_{90}) of the admission value and to a level undetectable in a peripheral blood smear (PC_T). Routine

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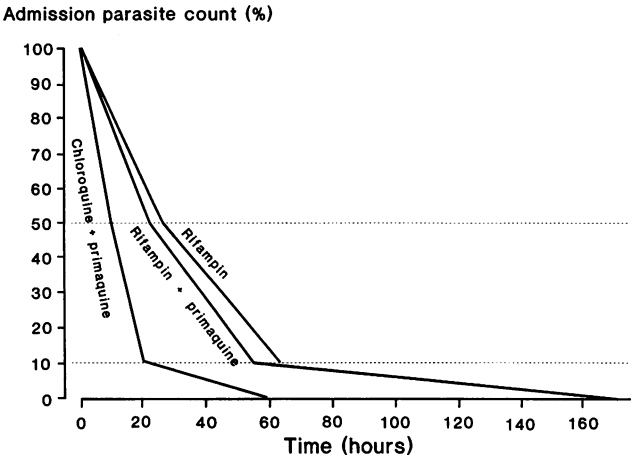

FIG. 1. Parasite clearance kinetics in *P. vivax* malaria following three different treatments. Data are means of PCT_{50}, PCT_{90}, and PC_T.

biochemical and hematological tests were performed at the time of admission and were repeated weekly thereafter.

All patients were hospitalized till clearance of both fever and parasites. Patients who failed to respond to rifampin were subsequently treated with the standard dose of chloroquine and primaquine. Reappearance of vivax malaria was assessed in patients who remained in the hospital for at least 1 month.

Statistical analysis. The data from each group were compared by one-way or multifactor analysis of variance (SSPS personal computer, version 4.0 software). Correlations were assessed by the method of Pearson.

RESULTS

A total of 60 adult male patients with *P. vivax* infection, aged between 15 and 50 years (mean [standard deviation (SD)] = 24 [7] years) were included in the study. The majority of the patients (48 cases, or 80%) had a history of previous attacks of malaria and had been in an area of malaria endemicity before admission; 17 patients came from the western border and 43 from the eastern border of Thailand. Of the 60 patients enrolled (5 treated with rifampin, 25 treated with rifampin-primaquine, and 30 treated with chloroquine-primaquine), 45 patients remained in the hospital for at least 1 month. The remaining 15 patients were unwilling to stay and were discharged after recovery. There were no significant differences among the treatment groups in age distribution ($P = 0.44$) or other baseline clinical or laboratory characteristics.

Clinical response. All treatment regimens were well tolerated. Following treatment, fever cleared in all patients and the overall mean (SD) time to fever clearance was 34 (27) h. The five patients treated with rifampin alone had another one to two attacks of high-grade fever (>38°C) 12 to 30 days after treatment. All the other patients remained afebrile during the 1-month hospital stay. Fever clearance times were significantly longer for patients treated with rifampin-primaquine (mean [SD] = 43 [35] h) than for patients treated with chloroquine-primaquine (27 [19] h; $P = 0.046$).

Parasitological responses. After the start of treatment, there was an initial reduction in parasitemia in all treatment groups and all but the five patients in the rifampin-only group became aparasitemic. The parasitemia reduction rates in the rifampin groups, both with and without primaquine, were lower than those following chloroquine-primaquine treatment (Fig. 1).

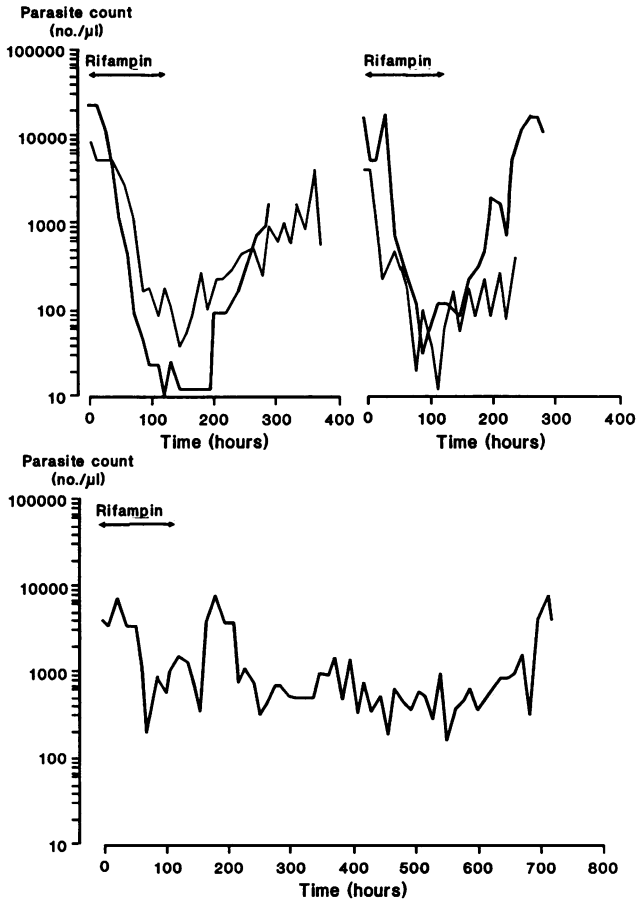

FIG. 2. Serial parasite counts for five patients with vivax malaria treated with rifampin alone.

The mean (SD) ratios of parasite counts at 48 h (one cycle) and 96 h (two cycles) to baseline counts in the rifampin group were 0.138 (0.174) and 0.017 (0.029), respectively. These ratios were similar to those for patients treated with rifampin-primaquine, i.e., 0.161 (0.05) at 48 h and 0.021 (0.28) at 96 h. However, ratios for both groups were significantly higher than ratios for the group treated with chloroquine-primaquine, i.e., 0.021 (0.053) at 48 h and clear by 96 h ($P < 0.001$). Thus, rifampin induced a ≥6-fold reduction in parasitemia per 48-h cycle compared with a 50-fold reduction induced by chloroquine. After completion of the 5-day rifampin treatment, four patients in the rifampin-only group had a rapid rise in parasitemia associated with another attack of fever. The other patient had fluctuating persistent parasitemia with another two attacks of fever but remained in good general condition (Fig. 2). All five patients developed high fevers associated with rising parasitemia, and all were treated subsequently with the standard chloroquine and primaquine regimen, beginning variously at 9, 12, 12, 16, and 30 hospital days. After these five cases had clearly failed therapy, further recruitment to the treatment group receiving rifampin alone was terminated.

Patients who received the sequential rifampin and primaquine regimen had significantly longer parasite clearance times (PCT_{50}, PCT_{90}, and PC_T) than those treated with chloroquine and primaquine ($P = 0.053$ to <0.001) (Fig. 1). The overall mean (SD) parasite clearance time for the rifampin-primaquine group (PC_T = 171 [54] h) was almost three times longer than for the group treated with chloroquine-primaquine (62

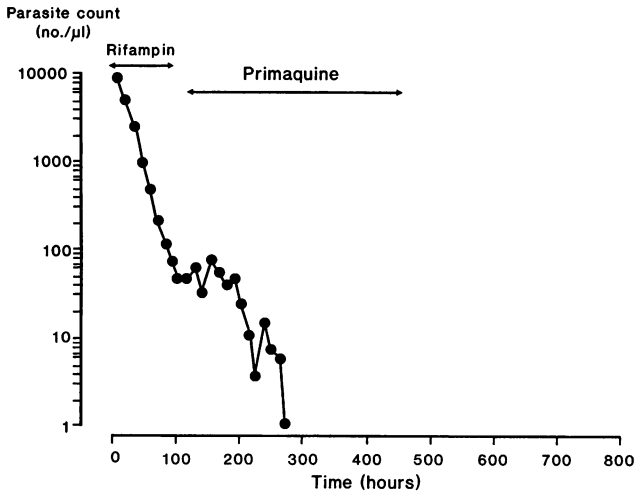

FIG. 3. Serial parasite counts for 25 patients treated with the sequential combined regimen of rifampin followed by primaquine. Data are geometric means.

[22] h; $P < 0.001$) (Fig. 1 and 3). The PCT_{50} and PCT_{90} of patients treated with rifampin (with or without primaquine) were similar, although the rifampin-only group did not subsequently clear their parasitemias (Fig. 2). The overall parasitological responses (PCT_{50}, PCT_{90}, and PC_T) were correlated with fever clearance times ($r = 0.43$ to 0.65; $P < 0.001$).

Subsequent clinical course. During 1 month of hospital stay by the 45 patients (5 treated with rifampin, 20 treated with rifampin-primaquine, and 20 treated with chloroquine-primaquine), reappearance of infection occurred in only one patient treated with rifampin-primaquine (on the 28th hospital day). This patient was subsequently treated successfully with chloroquine-primaquine. During the 1-month period, there were no significant differences in the weekly hematological or biochemical findings in any of the treatment groups. In particular, changes in hematocrits were similar in the three groups.

DISCUSSION

Rifampin has been used extensively for the treatment of tuberculosis and leprosy since 1969. Although there has been no clinical trial against malaria in humans, rifampin has been known to have activity in vitro against all erythrocytic stages of *P. falciparum* (6, 16). Rifampin is also active against the murine malarias *Plasmodium chabaudi* and *Plasmodium berghei* in vivo (1, 16). In bacteria, rifampin inhibits DNA-dependent RNA synthesis through its effects on the beta subunit of eubacterial RNA polymerase (8, 17). The antimalarial action of rifampin may be similar because plasmodium parasites (including *P. falciparum* and *P. vivax*) contain an organellar circular DNA molecule that encodes the beta subunit of a prokaryocyte-like RNA polymerase (5). In this study, rifampin at the usual therapeutic dose of 15 to 20 mg/kg/day was active against the blood-stage infection of human *P. vivax* malaria. During rifampin treatment, parasitemia decreased ≥6-fold per 48-h asexual life cycle, but in the five patients treated with rifampin alone, parasitemia did not clear (an R2-like response). It is possible that a larger dose or a longer course of rifampin might improve the parasitological response, as in mouse models (1, 16), although this would be impractical, costly, and possibly toxic. On the basis of these reduction rates (~0.15 per cycle), it would take approximately 2 weeks of treatment to clear all parasites from the body. Treatment with a sequential combined regimen of rifampin and primaquine cleared fever and parasitemia in all patients, and late reappearance of infection occurred in only one patient. However, compared with results of treatment with chloroquine-primaquine, fever and parasite clearance times of these patients treated with rifampin-primaquine were long (and in the majority, clearance occurred during treatment with primaquine). Primaquine alone at these doses is effective against vivax malaria, with similar long parasite clearance times (13).

Many antibiotics have some antimalarial activity, but only clindamycin and tetracycline are widely used for the treatment of chloroquine-resistant *P. falciparum* malaria. Given alone, the tetracyclines act too slowly in treatment. In a study of 12 patients with vivax malaria, 2 to 4 g of tetracycline alone per day administered for 7 to 14 days gave parasite clearance times of 144 to 240 h. Furthermore, despite these relatively high doses, 4 of 12 patients required more than one course of treatment (3). Thus, rifampin appears to be similar to these other antibiotics in having relatively weak and slow antimalarial effects. But it could be synergistic with other more potent antimalarial drugs if given in combination. Quinine and tetracycline remain more than 90% effective against multidrug-resistant falciparum malaria in Thailand (10). Rifampin is more costly and is relatively more toxic, but in contrast to tetracycline it may be given to pregnant women and to young children. Short courses, as in the present study, are usually well tolerated. Further studies are needed to establish the role of rifampin and its related compounds in combination with other antimalarial agents for the treatment of multidrug-resistant *P. falciparum*.

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