# COMPARISON OF CHLOROQUINE, QUINACRINE (ATABRINE), AND QUININE IN THE TREATMENT OF ACUTE ATTACKS OF SPOROZOITE-INDUCED *VIVAX* MALARIA (CHESSON STRAIN) [1]

PRELIMINARY REPORT

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Chloroquine, 7-chloro-4-(4-diethylamino-1-methyl-butylamino)-quinoline, a synthetic antimalarial agent developed during the war, has proved superior to atabrine and quinine (1). The following investigations were designed to supplement other observations in induced malaria and field observations by a study of the relative effectiveness of chloroquine, quinacrine, and quinine in the treatment of acute attacks of standardized sporozoite-induced *vivax* infections.

## PROCEDURES AND METHODS

### *General*

Details of the general procedure and the plan of investigation are reported elsewhere (2). Thirty-nine healthy Caucasian volunteers at Stateville Penitentiary were infected with South Pacific *vivax* malaria (Chesson strain) by the bites of ten infected *Anopheles quadrimaculatus* mosquitoes or by injection of their infected salivary glands.[3] The Chesson strain (3) was chosen because it is characterized by a high relapse rate, short latent interval between relapses, and almost complete absence of delayed primary attacks (4, 5, 6). Evaluation of results could be made with a small number of subjects because the complete history of each infection was known; and because the effect of immunity on relapse rate was minimized by utilizing presumably susceptible individuals living in a non-endemic area, and by limiting therapeutic trials to primary attacks and early relapses. In addition, treatment was initiated early in most of the attacks.[4]

### *Drug administration*

*Chloroquine.* A total of 2 grams of base (equivalent to 3.2 grams of the diphosphate) was administered over a period of one week. After an initial dose of 0.4 gram of base, followed by two doses of 0.2 gram at four-hour intervals, the daily maintenance was 0.2 gram.

*Quinacrine.* After an initial dose of 0.4 gram of quinacrine dihydrochloride (approximately 80 per cent base) followed by three of 0.2 gram at four-hour intervals, the daily maintenance dose was 0.4 gram. The total amount of quinacrine for seven days was 3.4 grams of the salt.

*Quinine.* Two groups of subjects received quinine; one for seven days and one for 14 days. In the first group, a total of 11–12 grams of base (approximately equivalent to 13–15 grams of the hydrochloride or sulfate) was administered over seven days. In the second group, a total of 21–23 grams of base was administered over a 14-day period.

### *Analysis of drugs in plasma*

Whole blood was centrifuged for 15 minutes at 2,000 r.p.m.; the plasma was separated and recentrifuged for

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[1] This investigation was carried out under a contract, recommended by the Committee on Medical Research, between the Office of Scientific Research and Development and the University of Chicago. The work was planned in cooperation with the Panel on Clinical Testing of Antimalarials of the Board for Coordination of Malarial Studies. This work was further aided by the participation of Army Medical Officers assigned to the project by the Surgeon General, U. S. Army.

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[3] The mosquitoes used in these studies were provided by Dr. Clay G. Huff and Dr. Frederick A. Coulston of the Department of Bacteriology and Parasitology. They supervised infection of mosquitoes, inoculation of volunteers and determined the intensity of infection in the salivary glands of the mosquitoes.

[4] The investigations reported in this paper would not have been possible except for the enthusiastic cooperation of the inmates and the administrative staff of Stateville Penitentiary.

an hour at the same speed in order to insure complete removal of the components of the buffy coat. The anticoagulant used was potassium oxalate. Quinine and quinacrine were estimated in the plasma by the methods of Brodie and Udenfriend (7, 8). Chloroquine was estimated by a modification (9) of the method of Brodie, *et al.* (10).

## RESULTS

*Time necessary for the clearance of parasites during treatment.* Chloroquine cleared the blood of parasites more rapidly than quinacrine, which in turn required less time than quinine to produce negative thick smears of the peripheral blood. Three-quarters of the patients treated with chloroquine had negative thick smears two days after therapy had been instituted; with quinacrine, three days were necessary, and with quinine, four days (Figure 1). Patients treated with chloroquine on the average became afebrile sooner than those receiving the other drugs.

*Relapse rate.* The Chesson strain, studied under these conditions, has a higher relapse rate (5) than that usually seen in naturally acquired Southwest Pacific *vivax* malaria in non-immune military personnel during World War II (11). In these studies the rate was approximately the same with each drug (Figure 2 and Tables I, II, III, IV). Two patients did not relapse after treatment with quinine. One of the patients had a markedly prolonged prepatent period (57 days) and the other a long parasitic latent period (39 days) following treatment of the preceding attack. Analysis of data from over one hundred clinical attacks studied under these conditions has revealed that patients in whom the prepatent period exceeds 14 days or the preceding latent period 30 days have much lower relapse rates than do patients in whom these periods are shorter (5).

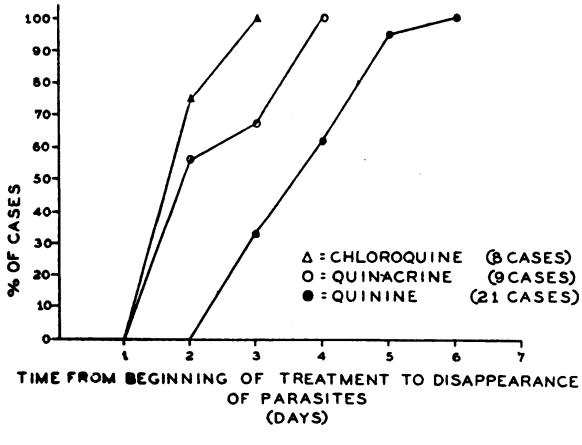


FIG. 1

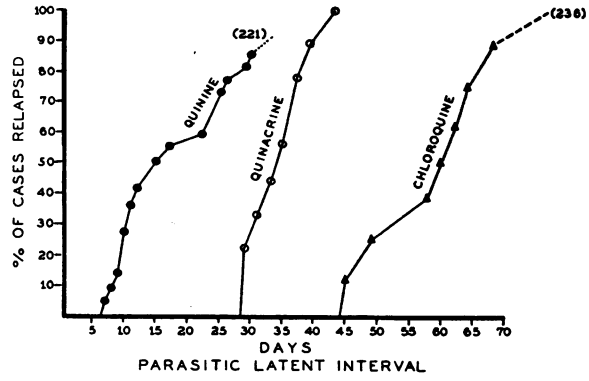


FIG. 2

*Parasitic latent period after termination of treatment.* The median length of the period of parasitic latency following termination of therapy with chloroquine was almost twice as great as that following quinacrine and four times as great as that after quinine (Figure 2, Tables, I, II, III, IV). In 50 per cent of the patients treated with quinine, parasitemia recurred in 15 days or less. With quinacrine, 50 per cent relapsed within 34 days, whereas, with chloroquine, 60 days elapsed before parasitemia reappeared in one-half the group.

One of the patients treated with quinine for 14 days did not undergo parasitic relapse until 221 days, and one of the patients treated with chloroquine did not relapse until 236 days. The former subject had a latent period of 38 days after his primary attack and probably presented a less severe challenge to the drug. The latter patient was permitted to undergo an unusually long febrile course both during the primary attack and first relapse, and, therefore, acquired immunity may have been a factor in prolonging his latent period after treatment with chloroquine.

*Drug dosage and concentration of drug in the plasma.* The dosages of drugs used in these studies were probably as high as can be given in large scale treatment with reasonable expectation of avoiding severe toxicity. With quinine, in fact, moderate symptoms of cinchonism were encountered.

The average of the mean plasma drug levels of

TABLE I

*Therapeutic effect of quinine, administered for seven days*

| Patient | Nature of attack* | Range of plasma concentration | 7-day mean plasma concentration | Parasite clearance | Interval from end of therapy to relapse | |
|---|---|---|---|---|---|---|
| | | | | | Fever | Parasites |
| | | *mgm./L.* | *mgm./L.* | *days* | *days* | *days* |
| 1 | P | 5.9–18 | 9.7 | 5 | 8 | 7 |
| 2 | P | 4.0–16 | 9.4 | 5 | 12 | 10 |
| 3 | P | 8.8–27 | 16 | 4 | 19 | 15 |
| 4 | $P_v$ | 3.9–11 | 8.1 | 4 | 10 | 10 |
| 5 | $P_v$ | 3.3–16 | 8.4 | 6 | 12 | 9 |
| 6 | $P_v^{12}$ | 5.1–19 | 12 | 5 | 15 | 11 |
| 7 | $P_e^{11}$ | 6.8–15 | 10 | 5 | 14 | 11 |

* P indicates a primary attack. The numerical exponents indicate the number of days of fever and/or parasitemia experienced by certain patients treated late during the course of an attack. The subscripts $_v$ and $_e$ indicate, respectively, intravenous or intracutaneous inoculations of dissected mosquito salivary glands.

all patients receiving quinine (8 mgm. per liter) was about 1⅓ times the concentration necessary to obtain a therapeutic effect (12). For the group receiving quinacrine the average plasma concentration (71 gamma per liter) was slightly greater than twice the minimum effective therapeutic level (12). In the group receiving chloroquine, however, the mean plasma concentration attained (96 gamma per liter) was at least six times the minimum required to eradicate the blood forms of the parasite (13).

TABLE II

*Therapeutic effect of quinine, administered for 14 days*

| Patient | Nature of attack* | Range of plasma concentration | 14-day mean plasma concentration | Parasite clearance | Interval from end of therapy to relapse | |
|---|---|---|---|---|---|---|
| | | | | | Fever | Parasites |
| | | *mgm./L.* | *mgm./L.* | *days* | *days* | *days* |
| 1 | P | 5.1–16 | 7.9 | 4 | 13 | 10 |
| 2 | P | 5.0–13 | 7.3 | — | — | — |
| 3 | P | 6.9–21 | 10 | 5 | 12 | 8 |
| 4 | $R_1$ | 5.3–16 | 6.6 | 5 | 16 | 12 |
| 5 | $R_1$ | 0.5–3.6 | 2.6 | 5 | 228 | 221 |
| 6 | $R_2$ | 4.1–11 | 6.0 | 4 | 32 | 25 |
| 7 | $R_2$ | 5.2–9.6 | 6.8 | 3 | 21 | 15 |
| 8 | $R_3$ | 4.0–8.8 | 6.0 | 3 | — | 25 |
| 9 | $R_3$ | 4.0–9.6 | 5.4 | 3 | — | 22 |
| 10 | $R_3$ | 3.4–5.9 | 4.9 | 3 | 20 | 17 |
| 11 | $R_3$ | 8.4–9.4 | 8.8 | 3 | 34 | 29 |
| 12 | $R_3$ | 4.6–7.8 | 6.4 | 4 | 34 | 26 |
| 13 | $R_3$ | 6.9–11 | 7.6 | 3 | 35 | 30 |
| 14 | $R_3$ | 7.0–13 | 10 | 3 | 30 | 25 |
| 15 | $R_3$ | 5.6–8.5 | 7.1 | 4 | — | — |

* P indicates a primary attack. $R_1$, $R_2$, and $R_3$ indicate first, second and third relapses.

## DISCUSSION

The two groups of patients treated with quinine differed significantly from each other in mean parasite clearance time and mean parasitic latent interval after therapy. The seven-day group showed a longer clearance time and a shorter latent interval than the 14-day group. The seven-day group consisted entirely of primary attacks whereas the 14-day group contained a large percentage of third relapses. The attacks treated with chloroquine were primaries and first and second relapses. They were probably intermediary in severity of infection between the two quinine groups. Furthermore, the difference in means between the quinine groups were smaller than the differences in means of all the quinine-treated patients and those treated with quinacrine and chloroquine. For these reasons, the two quinine series were considered as one group for purposes of comparison.

TABLE III

*Therapeutic effect of quinacrine, administered for seven days*

| Patient | Nature of attack* | Range of plasma concentration | 7-day mean plasma concentration | Parasite clearance | Interval from end of therapy to relapse | |
|---|---|---|---|---|---|---|
| | | | | | Fever | Parasites |
| | | *gamma/L.* | *gamma/L.* | *days* | *days* | *days* |
| 1 | P | 40– 90 | 61 | 3 | 34 | 29 |
| 2 | P | 92–105 | 151 | 2 | 44 | 39 |
| 3 | $P^{19}$ | 43– 77 | 65 | 4 | 39 | 37 |
| 4 | $P_L^{21}$ | 52– 80 | 59 | 2 | — | 43 |
| 5 | $P_v$ | 39– 95 | 62 | 2 | 29 | 29 |
| 6 | $P_v$ | 46– 70 | 54 | 2 | — | 35 |
| 7 | $P_v$ | 70–117 | 83 | 4 | — | 33 |
| 8 | $P^{22}$ | 35– 61 | 43 | 4 | 37 | 31 |
| 9 | $R_1$ | 50– 79 | 62 | 2 | 39 | 37 |

* P indicates a primary attack. $R_1$ indicates first relapse. The numerical exponents indicate the number of days of fever and/or parasitemia experienced by certain patients treated late in the course of an attack. The subscripts $_v$ and $_L$ indicate, respectively, intravenous or lymph gland inoculations of dissected mosquito salivary glands.

In the series here reported, chloroquine cleared the peripheral blood more rapidly in the majority of cases, than quinacrine and quinine. These results are consonant with findings obtained elsewhere in a large group of naturally occurring attacks (15). However, due to the small number of cases and to the fact that parasite clearance time was measured in days rather than in hours,

TABLE IV

*Therapeutic effect of chloroquine, administered for seven days*

| Patient | Nature of attack* | Range of plasma concentration | 7-day mean plasma concentration | Parasite clearance | Interval from end of therapy to relapse | |
|---|---|---|---|---|---|---|
| | | | | | Fever | Parasites |
| | | *gamma/L.* | *gamma/L.* | *days* | *days* | *days* |
| 1 | $R_1$ | 57–100 | 77 | 2 | 50 | 45 |
| 2 | $R_1$ | 69–103 | 83 | 2 | 52 | 49 |
| 3 | $R_1$ | 100–164 | 129 | 2 | 70 | 64 |
| 4 | $P_e$ | 62–103 | 85 | 3 | 63 | 58 |
| 5 | $R_2$ | 58– 88 | 74 | 2 | — | 60 |
| 6 | P | 95–152 | 125 | 3 | 71 | 68 |
| 7 | $R_2$ | 70–130 | 110 | 2 | — | 62 |
| 8 | $R_1^{26}$ | 73–110 | 89 | 2 | 241 | 236 |

* P indicates a primary attack. $R_1$ and $R_2$ indicate first and second relapses. The numerical exponent indicates the number of days of parasitemia experienced by one patient treated late in the course of an attack. The subscript $_e$ indicates subcutaneous inoculation of dissected mosquito salivary glands.

the difference between the mean parasite clearance time here reported between chloroquine and quinacrine, could not be proved statistically significant. On the other hand, the differences in mean clearance time observed between quinine and quinacrine and between quinine and chloroquine were sufficiently great for the size and dispersion of the series, for them to be considered due to factors other than chance.

The parasitic latent interval following treatment with chloroquine was longer than that following quinacrine and quinine. Furthermore, the margin between the minimal plasma concentration of chloroquine required to eradicate trophozoites and the concentration that may be attained without encountering toxicity is much greater than are the margins for quinacrine and quinine.

The long period of latency following treatment with chloroquine may be attributed, in part, to its slow rates of excretion and degradation; but the long persistence of an effective suppressive plasma concentration is also due to the fact that many times the minimal effective plasma concentration may safely be attained during therapy.

The results that we have obtained using experimental sporozoite-induced infection are in accord with previous studies in artificially induced infection (4, 13, 14) and with concurrent studies in naturally acquired malaria (15).

SUMMARY

1. Chloroquine has been tested in a small series of infections with highly relapsing Chesson strain of Southwest Pacific *vivax* malaria, under controlled conditions and compared with quinine and quinacrine.

2. Both chloroquine and quinacrine cleared the blood of parasites in most of the cases more rapidly than did quinine.

3. The relapse rate after treatment with all three drugs was about the same, 90 per cent or over.

4. The latent period following therapy for 50 per cent of patients treated with quinine, quinacrine, and chloroquine, was 15 days, 34 days, and 64 days, respectively.

5. Chloroquine was superior to quinacrine and quinine for the treatment of this series of acute attacks of *vivax* malaria.

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