

Subtitle: Effect of P. falciparum Infection on Serum Biochemistry Values of the Gibbon

Investigators:

MAJ Francis C. Cadigan, Jr., MC  
Verachat Chaicumpa, DVM  
Sanit Puhomchareon

### CHEMISTRY

Since white-handed gibbons (Hylobates lar lar), unlike chimpanzees continue to have intermittent parasitemia for as long as twelve months after P. falciparum infection produced by blood inoculation and most can be expected to have recurrent high peaks of parasitemia for at least three to four months and since there is no disease produced and thus no treatment required to keep the animals alive, it was felt that this animal would make a good model for study of the long term effects of P. falciparum on the serum chemistries.

### METHODS

Ten splenectomized juvenile white-handed gibbons (Hylobates lar lar) were inoculated with P. falciparum which had been maintained by blood passage in gibbons for approximately fifteen months. Gibbons P10,P11,S1,S13,S23,S25,S30, and S67 all received sixteenth passage material. Gibbon S77 received seventeenth passage material (from S67) and gibbon 76 received twentieth passage material. All inoculations were with fresh heparinized blood and the dose was approximately  $10^8$  parasites. The donor animal for sixteenth passage material was blood group A, the recipients were AB,B,AB,B,B,B,B, respectively. The donor for S77 was group B; gibbon S77 is group A. The donor for S76 was group A; gibbon S76 is also group A.

Gibbon P10,P11,S1 and S30 had been previously inoculated successfully with other strains of P. falciparum. Gibbons S23 and S25 had been inoculated with blood from two patients who were originally thought to have P. vivax infections but were subsequently appeared to be mixed infection with P. vivax predominating. These gibbons are described in detail elsewhere.

After inoculation the animals were examined daily for evidence of overt disease and, for the first sixty days of infection, rectal temperatures were taken daily. Thick and thin smears of peripheral blood were made daily and stained with Giemsa stain. Parasites counts were recorded in terms of the number of trophozoites per 500 WBC. Blood was drawn at 7 day intervals for determination of hematocrite, BUN, cholesterol, bilirubin, thymol turbidity, alkaline phosphatase, SGOT, SGPT, creatinine, total protein and electrophoresis. Blood chemistries were done in the same manner as described by Desowitz et al. in another report in this series. Studies on S76 and S77 were for a period of three months, the remainder were studied for six months.

### RESULTS

All infections were patent on peripheral blood smear by day 6 (Table 1) and reached a 1% parasitemia level by day 21. All of those followed for six months except P10 continued to show significant parasitemia levels for at least 120-days. Figures 1 and 2 show representative patterns of the biochemical changes compared to peripheral parasitemia.

At no time in the course of infection did any gibbon show evidence of any symptoms attributable to malaria. No change in behaviour, appetite or attitude was noted. Minor fluctuations in body temperature occurred as was reported in a previous series but no clinically significant correlation could be made with the stage or degree of infection.

TABLE 1

	First patent	Parasitemia over 1%	No of parasites in inoculum
P10	Day 2	Day 8	$12.8 \times 10^7$
P11	Day 2	Day 8	$12.8 \times 10^7$
S1	Day 1	Day 7	$12.8 \times 10^7$
S13	Day 1	Day 6	$12.8 \times 10^7$
S23	Day 1	Day 18	$12.8 \times 10^7$
S25	Day 1	Day 8	$12.8 \times 10^7$
S30	Day 2	Day 21	$12.8 \times 10^7$
S67	Day 2	Day 4	$9.6 \times 10^7$
S76	Day 6	Day 13	$6 \times 10^7$
S77	Day 6	Day 15	$18 \times 10^7$

In every instance, a drop in hematocrit followed peaking of peripheral parasitemia. The lowest hematocrit noted was 28% in S1. Detailed analysis of hematologic response to falciparum infection in the gibbon is given elsewhere.

The blood urea nitrogen, direct and total bilirubin, thymol turbidity, and creatinine levels fluctuated during the observation period but showed no correlation with parasitemic curves and no positive trend except that the BUN increased in the first two weeks after infection in all animals and showed an overall tendency to continue to rise in gibbons P11, S13, S25, S30 and S67.

Cholesterol levels dropped consistently, at about the same time that the hematocrit fell, to levels as low as one-third of pre-infection levels. Although the serum cholesterol dropped with subsequent rises in peripheral parasitemia, the decrease was not as marked as on the original parasite peak.

The transaminase levels showed no significant fluctuation in the six gibbons which had been infected previously, but marked increases in both SGOT and SGPT occurred in the four animals which had not been inoculated with malaria previously. There was a lag of at least several days after peak parasite levels were attained before the enzyme values increased. Since determinations were done at 7 day intervals, it is not possible to define the response further at this time.

Seven of the ten animals showed a slight but definite rise in total protein during the study period. Serum albumen levels showed a consistent rise from pre-infection levels in the first few weeks and then remained fairly constant. Globulin levels decreased slightly in most of the animals during the first month and then gradually increased. The A/G ratio increased during the first few weeks of infection, then gradually fell to slightly higher than pre-infection levels.

In the globulin fractions, there was no major change or trend in the alpha-2 globulin. Beta globulin decreased slightly during the first ten days in six animals and then with minor fluctuations remained at the same level. Gamma globulin showed a slight decline in the first ten days in all but two animals, then in six animals began to rise after 40 days and remained stabilized after 100 days. Two animals had transitory rises at 70-80 days. In most instances the gamma globulin increased 50-100% above pre-infection levels.

Alkaline phosphatase levels decreased in six cases, rose in three cases and remained essentially unchanged in two. Two of the six cases which showed a rise (S76 and S77) had very marked rises with a peak in the third week followed by a drop to normal by the fifth week.

GIBBON P-10

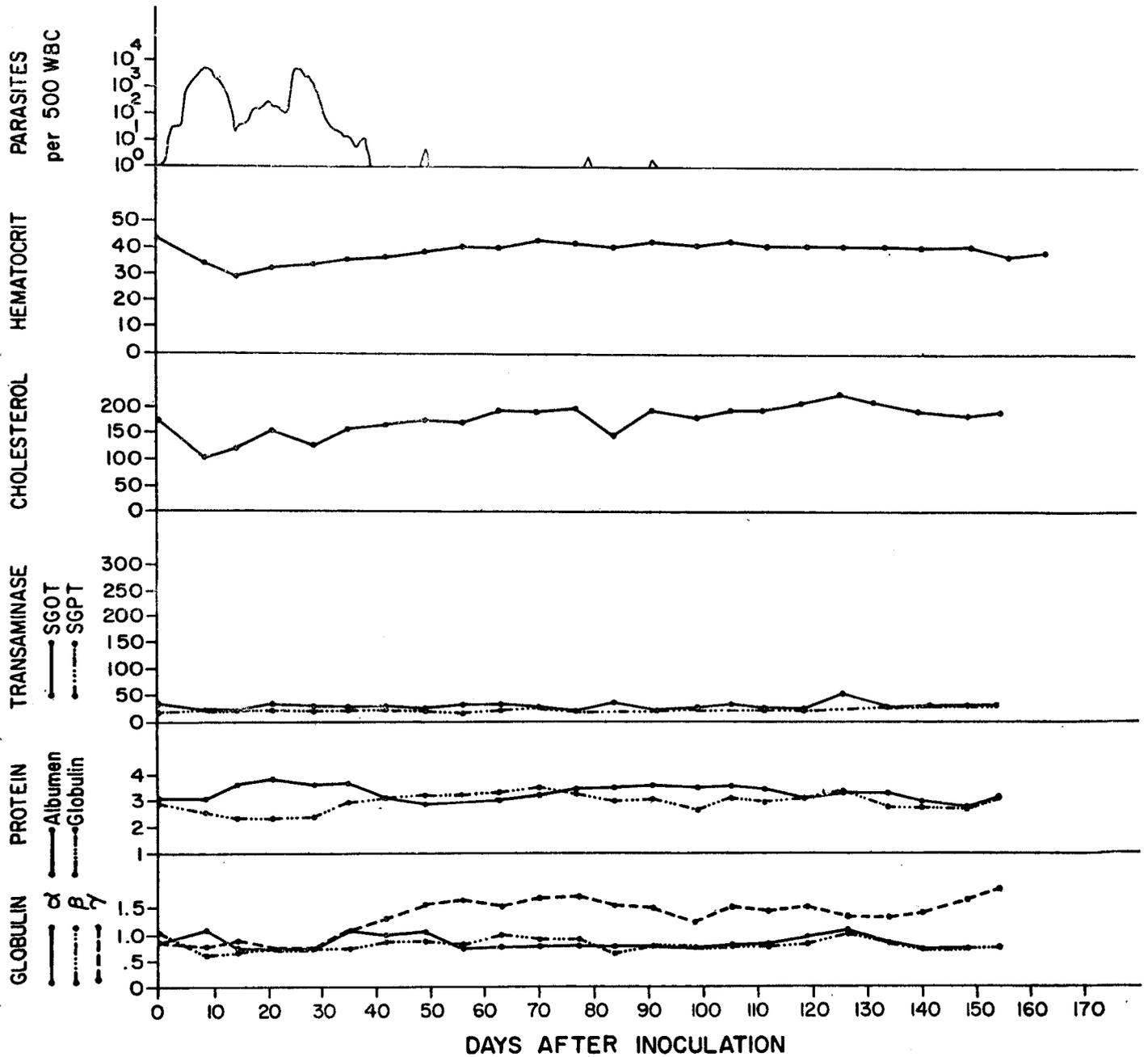


FIGURE 1. COMPARISON OF HEMATOCRIT AND SERUM CHEMISTRY VALUES WITH PERIPHERAL PARASITEMIA IN A GIBBON PREVIOUSLY INFECTED WITH AN HETEROLOGOUS STRAIN OF P. FALCIPARUM.

GIBBON S-77

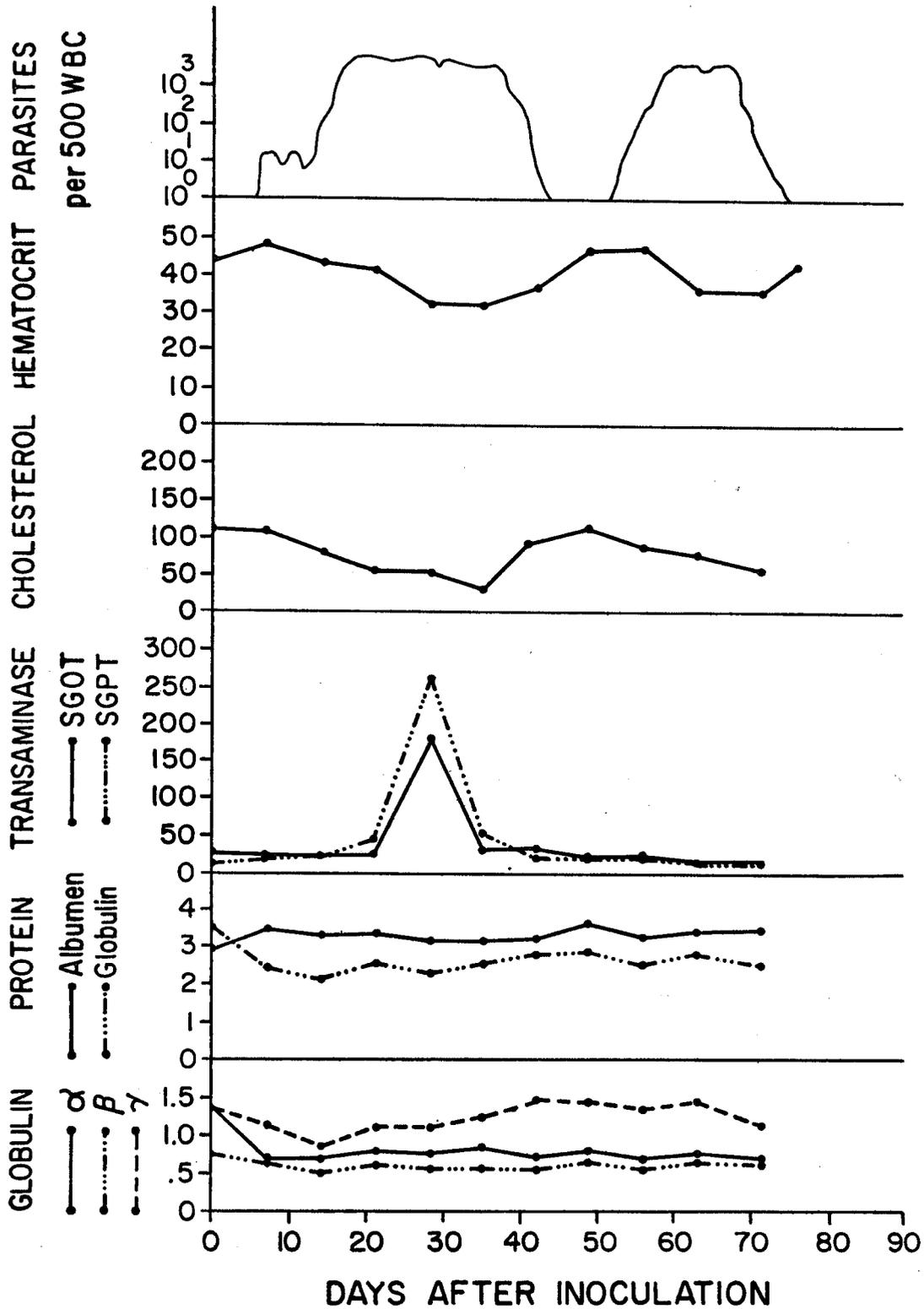


FIGURE 2. COMPARISON OF HEMATOCRIT AND SERUM CHEMISTRY VALUES WITH PERIPHERAL PARASITEMIA IN A GIBBON IN ITS FIRST INFECTION WITH P. FALCIPARUM.