

Subtitle: Protection Afforded by Previous Infection with Homologous and Heterologous Strains of P. falciparum

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Previous studies in chimpanzees have shown that a South East Asian strain of P. falciparum would not protect against infection with a West African strain of the same parasite. Since the effectiveness of any attempts at active immunization would be dependent upon the homogeneity of the strains in the area where immunization would be done, we attempted to determine the degree to which several isolates of P. falciparum from a relatively small area would show cross-protection.

METHODS

The isolates of P. falciparum were obtained from patients at the Phrabuddhabat Hospital and the nearby Passive Detection Center of the National Malaria Eradication Project (Region I) located in Saraburi province in Central Thailand. The exact locations of the patients homes are not known, but are all from the general area from which patients are drawn or roughly from 40 miles north to 20 miles south along the mountain range in which these institutions are located. All of the original infections in gibbons were started as blood transfers from the human cases except P10, S1, S3 which were sporozoite induced infections. Subsequent gibbon passage was by blood transfers. Each isolate in gibbons was given a letter designation. Table 1 lists the previous infection histories of the gibbons which had been previously infected. Animals P1, P2, P3, P7, P10, P13, S2, S10, S12, S30 had reached a 1% level of parasitemia at some time during their infection.

Animals used were juvenile white-handed gibbons (Hylobates lar lar) which were wild-caught in Thailand. After a period of adjustment to captivity, all animals were splenectomized and then treated with 17.5 mg/kg chloroquine and 10.5 mg/kg of primaquine. No malarial parasites were observed before or after surgery in any of these animals. Before challenge with a second infection of P. falciparum, all animals had been free of peripheral parasitemia for at least several months.

Blood smears, both thick and thin, were done daily on all gibbons and stained with Giemsa stain. Parasite counts were made on the basis of the number of trophozoites per 500 white blood cells and converted to number of parasites per cubic millimeter by using the mean white blood count.

The following schemata of challenge were used.

1. In test I, isolate B was used to challenge an animal previously infected with isolate B (P3) and a virgin animal (S39).
2. In test II, isolate B was used to challenge animals previously infected with isolate B (S2), A (S7) and J (S10).
3. In test III, isolate B was used to challenge one animal with two prior inoculations of A (P1), one animal with two prior inoculations of B (P3), one animal with a single inoculation of B (P2) and two virgin animals (S59, S60).
4. In test IV, isolate B was inoculated into animals which had previously been infected with A (S30), B (P7), C (P11), D (P13), F (P10), G (S1), H (S3), L (S12), N (S25), O (S23), and S13 (a virgin).

5. In test V, isolate A was used to challenge an animal infected with A (S41), one infected with B (S60), one which was infected twice with B (P2), one which had had three inoculations of B (P3), an animal which had had Q (S58) and a virgin animal (S79).

TABLE 1

	Isolate	Gibbon Passage	Number of weeks of parasitemia	Remarks
P1	A	First/Third	27/9	Rechallenged
P2	B	Second	28	
P3	B	First	36	
P7	B	First	54	
P10	F	First	12*	Sporozoite induced
P11	C	First	37	
P13	D	First	37	
S1	G	First	14*	Sporozoite induced
S2	B	Second	27	
S3	H	First	14*	Sporozoite induced
S7	A	Second	27	
S10	J	Second	14	
S12	L	First	23	
S13	—	—	—	
S23	O	First	12	
S25	N	First	19	
S30	A	Third	21	
S39	—	—	—	
S41	A	Third	41	
S58	Q	Second	19	
S59	—	—	—	
S60	—	—	—	

* End of observation period, full duration not known

RESULTS

The parasitemia curves are shown graphically in figures 1-5. It can easily be seen in each test that prior infection with the homologous isolate confers a marked degree of protection upon challenge. Although re-infection occurs, the parasite levels are lower by 1.2 logs than the control and in the two tests in which there was a long follow-up, the total days of detectable parasitemia differed markedly as well.

In the three tests in which animals previously infected with isolate A were challenged with isolate B there was evidence of partial protection as shown by a delay in reaching high levels (1% parasitemia) compared to the controls. This was particularly marked in the case of P1 (Fig 3) which had been infected twice with isolate A before challenge with B. Conversely, an animal previously infected with isolate B (S60 in Fig 5) showed a delay in building up the level of parasitemia when challenge with isolate A. It is interesting to note that P2 which had had two infections with B showed a greater degree of protection to isolate A than did S60 and that P3 which had had three previous infections with B showed as much protection as did S41 which had been infected with the homologous strain.

From Figure 4 it can be seen that infection with isolates C, G, N, and O conferred no protection against challenge with isolate B. Gibbon S3 which had had previously been infected with isolate H showed the same response to challenge with B as did P7 for which B was an homologous challenge. It is possible that these two isolates are an identical strain. Isolate F(P10) did not confer any protection in the first 30 days but may have done so subsequently. An intermediate type of response was shown by gibbons P13 and S12 (isolates D and L which showed protection of over 1 and 0.6 logs respectively but which continued to have low levels of parasitemia for a prolonged period.

From these data we would surmise that at least four strains are represented here by isolate A, B and H, D and L, and the group C, G, O and N. The latter group could easily represent more than one strain, of course, since they are grouped on the basis of no protection against B. The results with isolate F are such that no relative position in terms of the other isolates can be assigned. Isolate J (Figure 2) while dissimilar to A and B was not tested in comparison with the other strains and although its pattern resembles that of P7, no prediction of relationship can be made from these data.

In summary, one can say that of eleven isolates compared with B, only one (H) gave cross-protection of a degree similar to the homologous isolate and only two (D and L) gave persistent partial protection. Isolates A and F delayed the peak of parasitemia which possibly could be of value as an adjunct of chemotherapy.

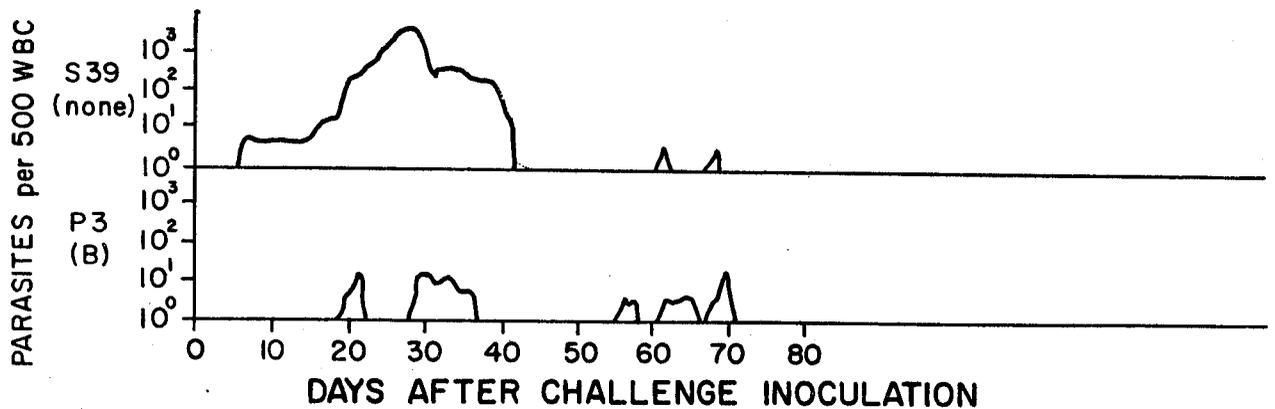


FIGURE 1. TEST I HOMOLOGOUS CHALLENGE WITH ISOLATE B

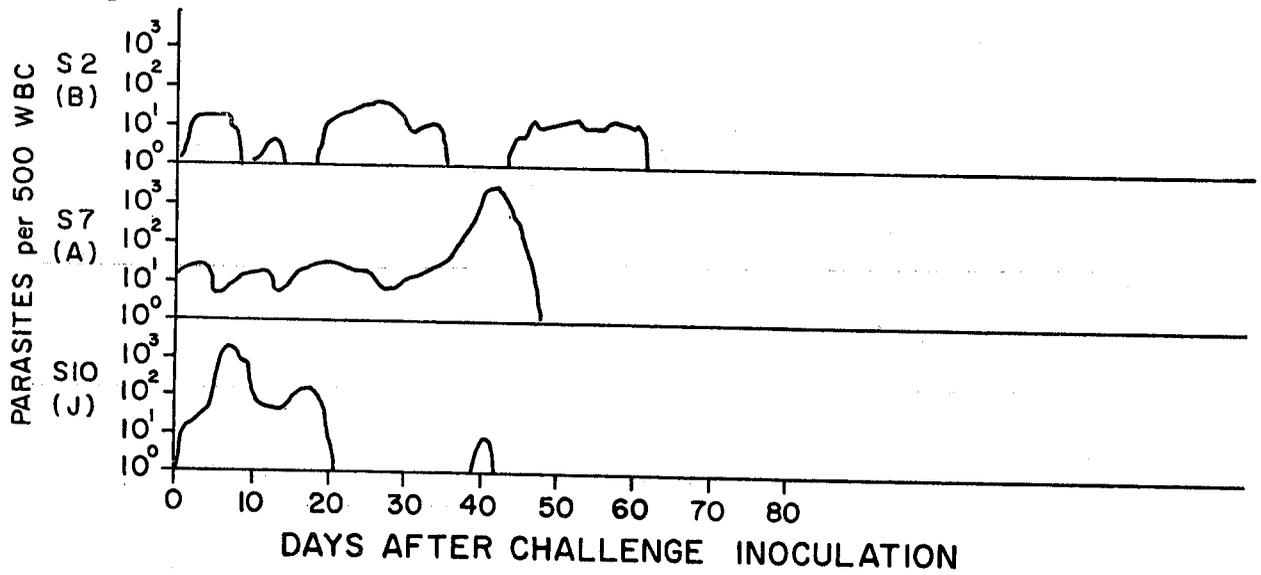


FIGURE 2. TEST II HOMOLOGOUS AND HETEROLOGOUS CHALLENGE WITH ISOLATE B

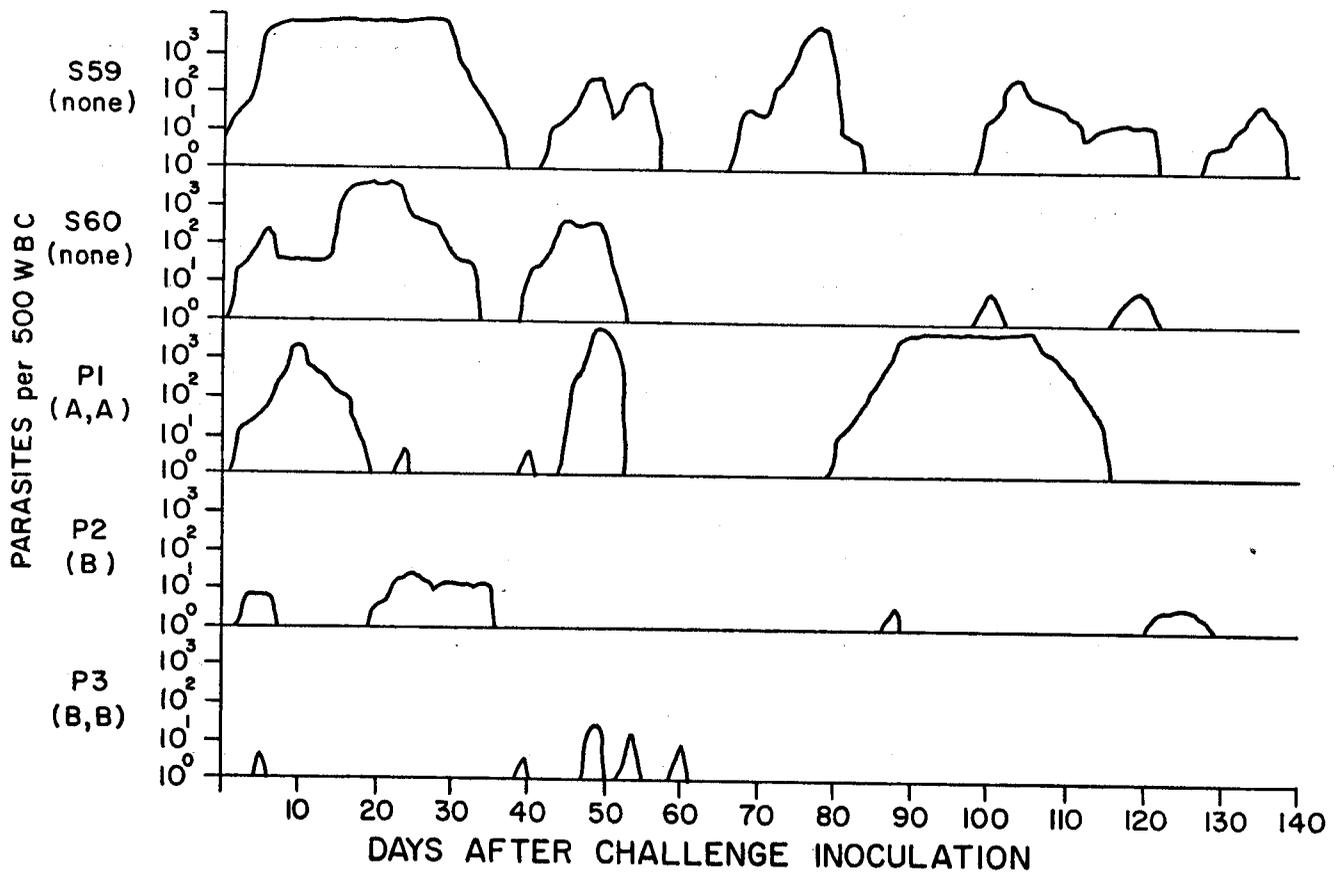


FIGURE 3. TEST III HOMOLOGOUS AND HETEROLOGOUS CHALLENGE WITH ISOLATE B

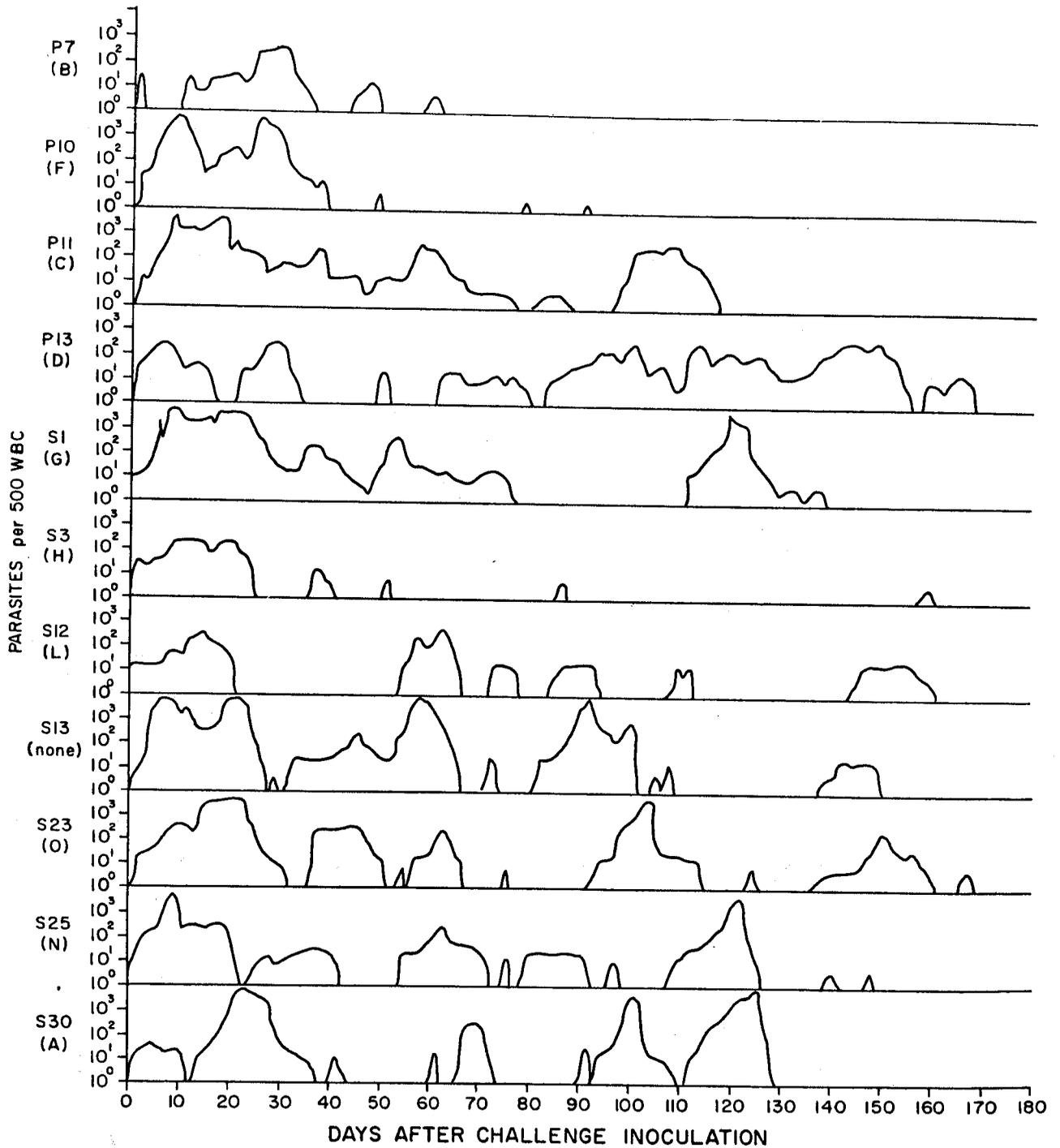


FIGURE 4. TEST IV HOMOLOGOUS AND HETEROLOGOUS CHALLENGE WITH ISOLATE B

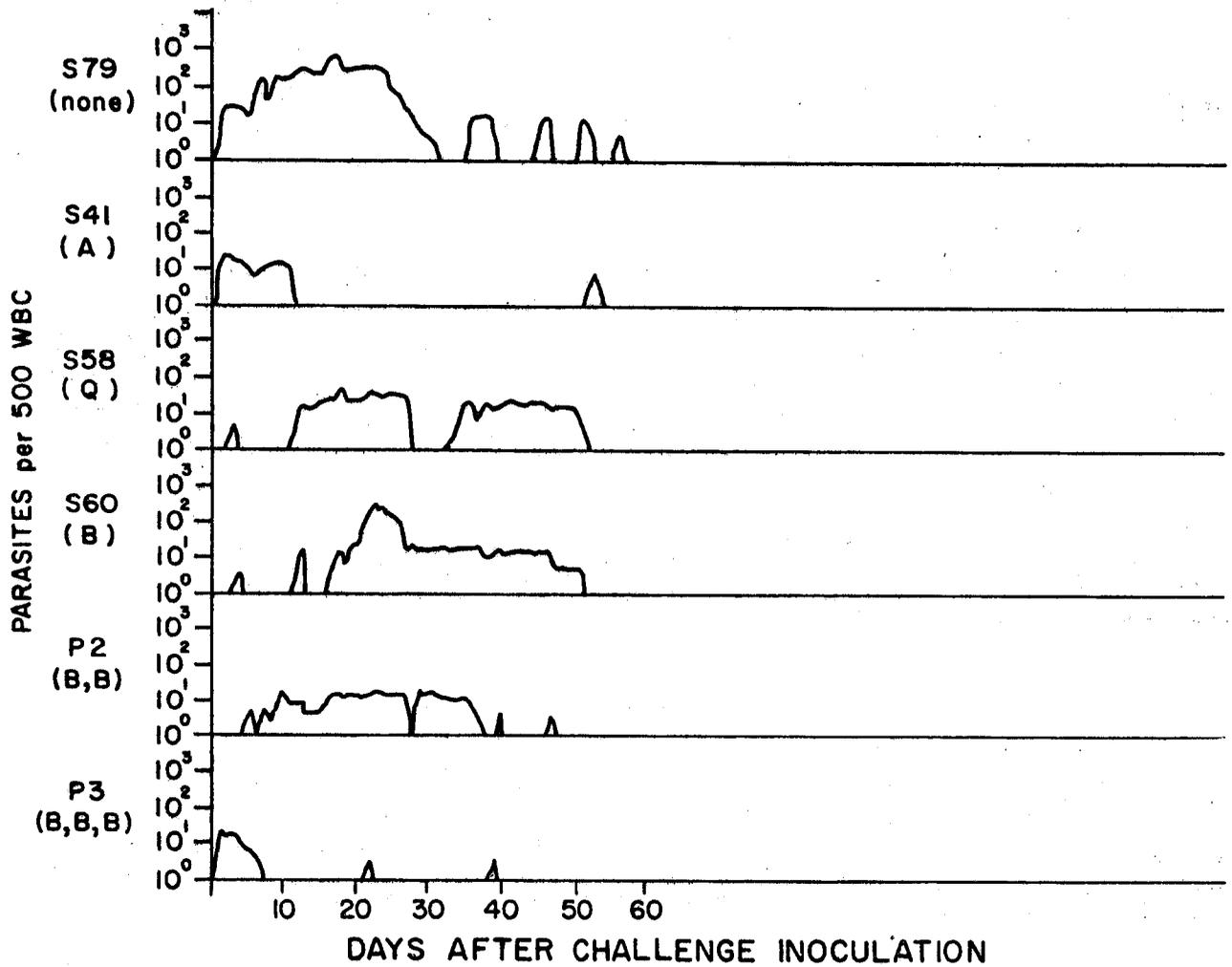


FIGURE 5. TEST V HOMOLOGOUS AND HETEROLOGOUS CHALLENGE WITH ISOLATE A