

Subtitle: Plasmodium falciparum in the Gibbon (Hylobates lar lar). Pattern of infection.

Investigators:

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The white-handed gibbon (Hylobates lar lar) of Thailand has previously been reported to be susceptible to both blood and sporozoite induced infection with P. falciparum. This paper reports more prolonged observation of infections, and the influence of factors such as multiple passage, blood types and intercurrent infections on the pattern of parasitemia in splenectomized gibbons.

METHODS

The animals used were juvenile white-handed gibbons (Hylobates lar lar) from Thailand. All animals were splenectomized and treated with chloroquine and primaquine. No animals had detectable parasitemia before or after surgery.

Blood grouping was done with human anti-A and anti-B sera. Previous studies by Wiener et al. (1) have shown that the gibbon erythrocyte A and B antigens, while not identical with the human antigens are quite similar and that they react with human antisera. Crossmatch was done in the major mode (human cells into gibbon sera) and without the addition of albumen.

Blood smears were made daily and stained with Giemsa stain. Parasite counts were made in terms of number of trophozoites or gametocytes per 500 white blood cells and converted to number per cubic millimeter by using the mean white blood count.

RESULTS

Effect of blood group compatibility. Of eighty-four gibbons tested for the ABO blood group, none were group O, twenty-two were group A, thirty-six were group B and twenty-six were group AB. Crossmatches between human erythrocytes and gibbon sera of the same blood group were grossly incompatible. Similarly crossmatches between human O negative cells and gibbon sera of the three different groups (A,B,AB) were incompatible on gross examination. Thus there was no evidence to support the possibility that there was a prolonged survival time of human erythrocytes in the gibbon and that prolonged survival was the reason for success in transferring P. falciparum infection from humans to white-handed gibbons. Despite the incompatibility of human cells with gibbon sera, the fourteen attempts to pass P. falciparum by blood transfer from humans to gibbons were all successful.

On the other hand, in the gibbon to gibbon transfers there is evidence that blood group compatibility has an effect on the speed with which peripheral parasitemia becomes patent and upon the speed with which the initial peak is reached. Table 1 shows that although all transfers were successful and all reached a parasitemia level of 1% in the first month when there was transfer from an infected gibbon into recipient gibbon which had an incompatible group (e.g. A into B or AB into A) there was a delay in the mean prepatency period and a delay in reaching the 1% parasitemia level. It can be seen that this was not so great as to be of significance in the selection of animals.

Effect of inoculum size. It is also apparent from Table 1 that the size of the inoculum is of at least equal importance as the compatibility of blood. In one instance in which an identical inoculum was put into five gibbons simultaneously, patency occurred in the compatible recipients on days 4, 6 and 10, and in the incompatible recipients on days 4 and 6. Peaks occurred in the compatible animals on days 21, 20, and 31 respectively, and in the incompatible animals on days 26 and 21 respectively.

Duration of infection. Peripheral parasitemia is usually detectable throughout the first eight to ten weeks of infection and thereafter is variable in occurrence. Parasitemia may not be detectable on peripheral smear for several weeks or even, less often, several months and then reappear. The mean overall duration of detectable parasitemia is thirty-one weeks or about seven months. In individual animals it has varied from as short as six to as long as seventy-two weeks. In the two strains which have undergone multiple gibbon passage, there has as yet been no indication that the duration of infection was affected. One of these, strain B, is now in its twenty-second gibbon passage but only the first eleven passages have been followed for a sufficient period to comment on duration of the infection. Strain A in four passages has not changed its mean duration.

Pattern of infection. Five gibbons which were inoculated simultaneously with the same dose of *P. falciparum* (strain A) had similar parasitemia curves. These are shown in figure 1. It can readily be seen that the basic pattern is the same in each animal. As infection continued, succeeding periods of high peripheral parasitemia tended to be of shorter duration, to occur at greater intervals and to attain lower maximum levels.

Intercurrent infection. Naturally occurring upper respiratory infection, herpes simplex infection and superficial fungal infections have not affected the levels of parasitemia, nor have they resulted in malaria disease. Gibbons inoculated with dengue types 1 and 2 virus, although showing evidence of infection (serologic changes or viremia), did not show alteration of the pattern of the malaria infection.

Effect of multiple passage. There is no evidence in the twenty-second passage of one strain nor in the fourth or fifth passage of two other strains that there is any great alteration in the pathogenicity of the parasite. No disease symptoms attributable to malaria infection have occurred even in the presence of a 8% parasitemia. No significant change in the time before patency nor before reaching a 1% parasitemia level has been noted. Chloroquine resistance has been maintained through twenty-two passages. As mentioned previously, the duration of infection has not been altered by multiple passage.

The effect of multiple passage on gametocytemia seems to be an increase in the number of gametocytes and the number of days on which they are found. Precise quantitation of this has not been done yet, but a definite trend is obvious. In addition, whereas the gametocytes in early passages were all very immature, now many of them are more nearly mature and on two days in the 14th passage, a few morphologically mature gametocytes were seen. Attempts to infect mosquitoes from gibbons have been unsuccessful. Details are listed under another study report. The frequency with which schizonts are noted in the peripheral blood has increased markedly.

Table 1. Effect of ABO compatibility and size of inoculum on infection

Patent on Day	Compatible		Patent on Day	Incompatible	
	1% Parasitemia on Day	Inoculum* Parasites x 10 ⁶		1% Parasitemia on Day	Inoculum* Parasites x 10 ⁶
1	3	300	1	6	300
1	3	300	1	6	300
1	4	300	1	10	300
1	4	300	2	6	300
1	4	150	5	15	15
1	5	300	6	13	40
1	18	300	6	15	150
2	5	300	6	25	50
2	9	40	7	14	300
2	18	300			
3	8	40			
3	10	60			
6	31	1			
Mean 1.9	9.4		3.9	12.2	

* Numbers have been rounded off for ease in reading table.

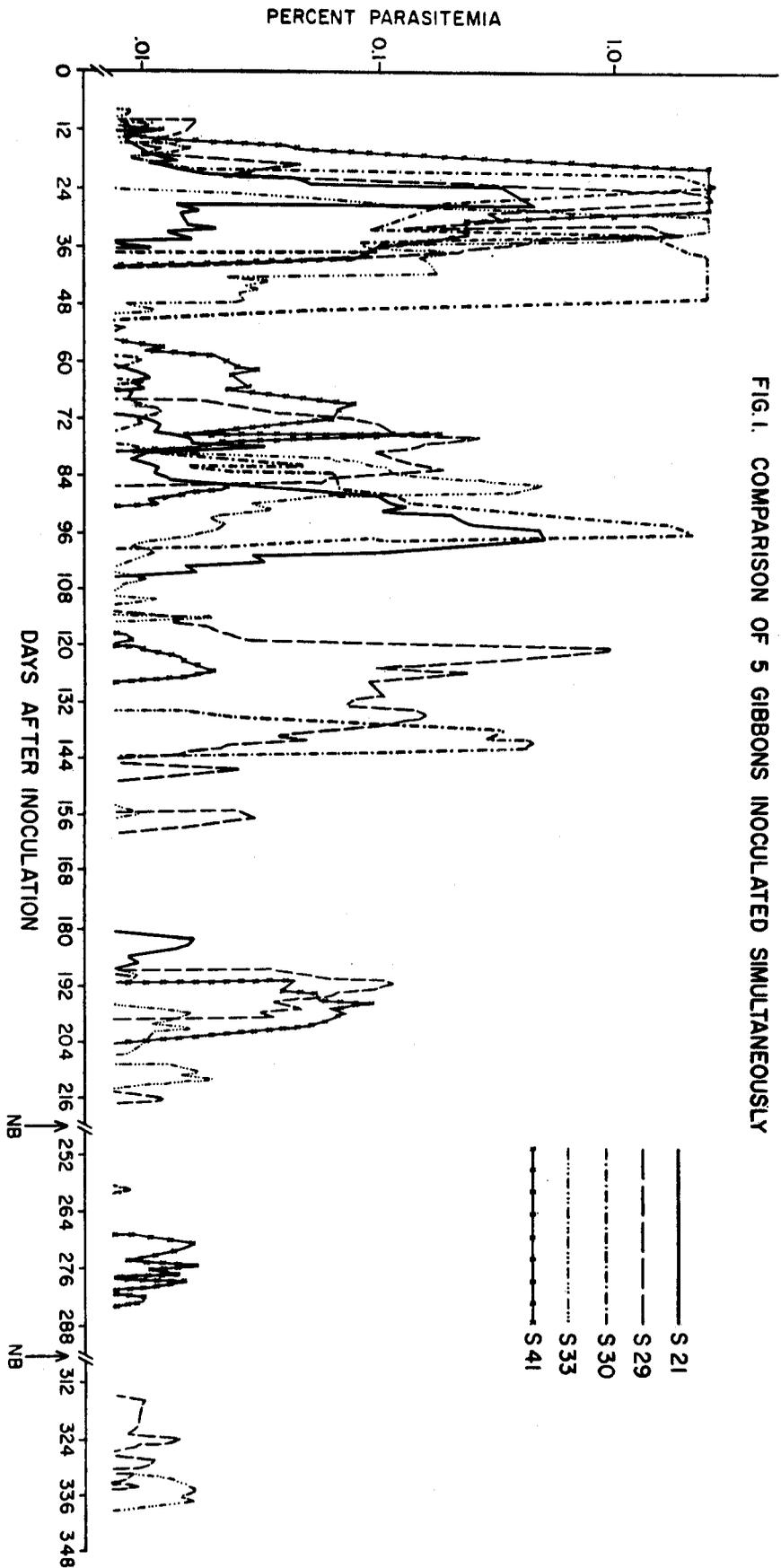


FIG. 1. COMPARISON OF 5 GIBBONS INOCULATED SIMULTANEOUSLY