

Title: Transmission of Plasmodia to Heterologous Hosts

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Species of the genus Plasmodium have been considered to be highly restrictive in their host parasite relationships. There is, however, a growing body of evidence that some plasmodia may infect heterologous hosts. The ultimate aim is to adapt the human malarials to a convenient laboratory animal such as the white rat or mouse. In addition, the processes by which plasmodia may be induced to infect normally non-susceptible hosts can give valuable information on the dynamics of host parasite relationships.

This report summarizes the results of a number of experiments on experimental transmission of primate malarials to heterologous hosts.

1. Attempts to infect the white rat with primate malarials. (K. Pavanand and R.S. Desowitz)

a. P. inui: Three splenectomized and two intact young adult rats (weight: 50-70 gm.) were inoculated via the tail vein with a washed saline suspension of erythrocytes infected with P. inui (484/10,000 rbc). Blood smears were made daily for five days and weekly thereafter. One splenectomized rat showed ring forms 48 hours after inoculation and was negative thereafter. All other animals remained negative.

b. P. coatneyi in immuno-suppressively treated rats: Two rats, one splenectomized and one intact, were given per os 5-7 mg./kg of Purinethol daily for 6 days. The course was begun two days before being infected with P. coatneyi via the tail vein. Two other untreated animals (one splenectomized and one intact) were inoculated with the infected monkey blood at the same time. No parasites were found in any of the rats during the one month that blood films were examined.

c. Gibbon malaria:

The first trial employed four adult rats. One splenectomized and one intact rat were treated with Purinethol as described in the preceding experiment. One intact and one splenectomized rat were left untreated. All animals were intravenously inoculated with heavily infected gibbon blood (parasitaemia 1086/10,000 rbc). Blood smears were taken daily. The intact, untreated rat showed pigment in the leukocytes three days after inoculation but no parasites were detectable. On the 6th day many well-developed ring forms were seen. The blood was negative thereafter. All other animals were negative throughout the two week period of observation.

The second experiment employed younger rats weighing between 40-60 gm. Immunosuppressive was not given in this trial. Five rats were splenectomized and four remained intact. Blood from gibbon P-9 (parasitaemia 876/1000 rbc) was washed twice, resuspended in sterile physiologic saline and 0.7-1.5 ml. and inoculated via the tail vein. All rats showed parasites in the blood up to 48 hrs after inoculation. In one rat (Intact) a single ring form was again seen on the 21st day.

The third series of trials employed newborn rats not older than 24 hrs. Ten newborn rats were inoculated with 0.50 ml. of heavily infected blood from gibbon P-9 (parasitaemia 1004/10,000 rbc). Parasites were seen in blood films made 28 and 93 hrs. after inoculation. On the 13th day three animals remained alive and in two of these what appeared to be very young pre-ring stage forms (chromatin dot

with small solid cytoplasmic appendage, merozoite-like in appearance and size) were seen. The infections in these animals were very scanty. That these forms were probably parasites and not artifacts was indicated by their absence in ten newborn control rats inoculated with the blood of an uninfected gibbon.

A second group of 11 newborn rats were inoculated intraperitoneally with blood from gibbon P-9 (parasitaemia 618/10,000 rbc). One animal showed parasites in the peripheral smear within 16 hrs. and all others at 24 hrs. Nearly mature schizonts resembling P. jefferyi were seen in the film from one animal at 93 hrs. and in another at 141 hrs. Three animals were sacrificed 5 days after inoculation and the pooled blood inoculated into 6 newborn rats. Twenty-four hours after inoculation, two rats were dead, smears made from heart blood of one rat revealed mature schizonts. Daily blood films from the remaining rats were negative until the 9th day when very scanty young forms resembling merozoites were seen in all animals. One erythrocyte was found to be doubly infected with these forms. No parasites were seen after the 9th day. One of these rats was sacrificed on the 9th day and its blood inoculated into 5 newborn rats. Four of these rats (2nd subpassage) showed a scanty parasitaemia of young pre-ring forms 17 hrs. after inoculation. In one rat a developing trophozoite was seen on the 4th day.

These subpassages in new born rats are shown schematically below.

Gibbon P9
6 Days
1st gibbon to rat passage
5 days
1st rat to rat subpassage
9 days
2nd rat to rat subpassage
(positive 4th day)
24 days total sojourn in rat host

These results of these experiments indicate that it might be possible to adapt gibbon malaria (probably P. jefferyi) to the newborn rat. While the survival time of gibbon erythrocytes in the rat is as yet unknown, the finding of parasites 24 days and two subpassages later would make this factor unlikely, at least at the later subpassages. The presence of what appeared to be very young forms is evidence that at least one schizogonic cycle had occurred. The reason for the better success with gibbon malaria as compared to monkey malarias is as yet unknown. However, preliminary experiments have shown a greater compatibility between rat and gibbon bloods than between rat and monkey.

2. Primates as heterologous hosts. (R.S. Desowitz and F. Cadigan). These experiments have two objectives: 1. To determine the biologic relationships between the plasmodia of higher and lower primates. 2. To determine if infection with one might provide some immunologic protection against another, particularly between apparently related species such as P. coatneyi and P. falciparum, or P. inui and P. malariae.

a. Attempts to infect gibbons with P. coatneyi. Preliminary observations on the cross-immunity between the two species.

Two gibbons were inoculated intravenously with 2 ml. blood from a P. coatneyi infected rhesus. One gibbon S9 had not been experimentally infected previously and was considered "clean" while gibbon P9 had been infected with P. falciparum although the animal no longer had parasites in the peripheral blood films. Thirty-six days after inoculation a scanty parasitaemia was seen in gibbon S9. The parasitaemia continued at this low level for two weeks and then disappeared. One month after the last positive blood film the animal was inoculated with blood from the "gibbon line" of P. falciparum. The animal became infected with a course of parasitaemia no different than "non-immune" controls.

P9 showed a P. falciparum infection for 44 weeks. Approximately 6 months after the last positive blood film it was challenged with P. coatneyi. This animal did not subsequently become infected.

Further trials on cross immunity engendered by these two plasmodia are now in progress.

b. Attempts to infect rhesus monkeys with gibbon malaria: Two rhesus monkeys were inoculated intravenously with blood from a heavily infected gibbon. Neither became infected.

c. A comparison of the infection following inoculation of gibbon malaria into animals previously infected with P. vivax and P. falciparum with animals not previously inoculated revealed no evidence of any degree of protection afforded by either P. vivax or P. falciparum against the gibbon malaria.

d. To determine how specific is the susceptibility of gibbons to malaria, several splenectomized white-capped gibbons (Hylobates lar pileatus) have been obtained and inoculated with P. vivax and P. falciparum. Results thus far indicate that this sub-species is far less susceptible than Hylobates lar lar at least insofar as patent peripheral parasitemia is concerned. No conclusions can be made as this study has been in progress for only a short time.