

COMPARISON OF *PLASMODIUM FALCIPARUM* INFECTIONS IN PANAMANIAN AND COLOMBIAN OWL MONKEYS

RICHARD N. ROSSAN, JAMES S. HARPER III, DAVID E. DAVIDSON, JR.,*
ALFONSO ESCAJADILLO AND HOWARD A. CHRISTENSEN

Gorgas Memorial Laboratory, P.O. Box 935, APO Miami 34002 and

*Division of Experimental Therapeutics, Walter Reed Army Institute of Research,
Washington, DC 20307-5100

Abstract. Parameters of blood-induced infections of the Vietnam Oak Knoll, Vietnam Smith, and Uganda Palo Alto strains of *Plasmodium falciparum* studied in 395 Panamanian owl monkeys in this laboratory between 1976-1984 were compared with those reported from another laboratory for 665 Colombian owl monkeys, studied between 1968-1975, and, at the time, designated *Aotus trivirgatus griseimembra*. The virulence of these strains was less in Panamanian than in Colombian owl monkeys, as indicated by lower mortality rates of the Panamanian monkeys during the first 30 days of patency. Maximum parasitemias of the Vietnam Smith and Uganda Palo Alto strain, in Panamanian owl monkeys dying during the first 15 days of patent infection, were significantly higher than in Colombian owl monkeys. Panamanian owl monkeys that survived the primary attack had significantly higher maximum parasitemias than the surviving Colombian owl monkeys. Peak parasitemias were attained significantly earlier after patency in Panamanian than in Colombian owl monkeys, irrespective of the strain of *P. falciparum*. More Panamanian than Colombian owl monkeys evidenced self-limited infection after the primary attack of either the Vietnam Smith or Uganda Palo Alto strain. The duration of the primary attacks and recrudescences were significantly shorter in Panamanian than in Colombian owl monkeys. Mean peak parasitemias during recrudescence were usually higher in Panamanian owl monkeys than in Colombian monkeys. Differences of infection parameters were probably attributable, in part, to geographical origin of the two monkey hosts and parasite strains.

It was demonstrated¹⁻⁴ that eight strains of *Plasmodium falciparum* would regularly infect normal owl monkeys from northern Colombia. Such infections have been used in evaluation of antimalarial drugs and other biological studies. The Vietnam Oak Knoll, Vietnam Smith, and Uganda Palo Alto also will regularly infect Panamanian owl monkeys. This report is concerned with a quantitative comparison of the course of untreated malaria infections in monkeys of these two geographic origins.

The three strains of *P. falciparum*, Vietnam Oak Knoll, Vietnam Smith, and Uganda Palo Alto, were studied serially from 1976 to 1984 in 395 Panamanian owl monkeys. These strains were passaged in 665 Colombian owl monkeys from 1968 to 1975. Consequently, the parasite passage levels in these monkey subspecies were different.

MATERIALS AND METHODS

Monkeys—nomenclature and husbandry

Previously, Panamanian and northern Colombian owl monkeys were classified as belonging to the same subspecies, *Aotus trivirgatus griseimembra*.⁵ A recent revision⁶ of the genus *Aotus* separated them into two subspecies: Panamanian monkeys, (K VIII, K IX), *A. lemurinus lemurinus*, and Colombian monkeys (K II, K III, K IV), *A. l. griseimembra*. As this proposed nomenclature is tentative, the animals will be referred to in this report as Panamanian and Colombian owl monkeys.

Panamanian owl monkeys were obtained on the Isthmus of Panama, in areas west of the Panama Canal. Upon arrival at the laboratory, each animal was examined, weighed and sexed, identified by a metal neck tag with an accession number, administered thiabendazole orally (100 mg base/kg) and vaccinated with attenuated *Herpes simplex* and *Herpes tamarinus* viruses of atten-

uated virulence (New England Regional Primate Research Center, Southborough, Massachusetts). Both vaccines were used until July 1981; since then only *H. simplex* has been given.

The animals were housed as male-female pairs in quarantine rooms in wire mesh cages, measuring 31 × 24 × 28 in. Room temperatures varied seasonally from 26°C to 33°C, with relative humidities of 75% to 87%. A ventilation system provided not less than 15 air changes per hour. Illumination cycles in the rooms were white light from midnight to noon, and red light from noon to midnight.

About one month after arrival, each monkey was tattooed with its identification number and a thick blood film examined to exclude naturally occurring plasmodial infections. The animals remained in the quarantine room for a minimum of 90 days before being transferred to areas devoted to housing monkeys for malaria studies. The weight of the monkeys when inoculated ranged from 700–800 g.

From 1976 to 1981, the diet on five days of the week consisted of commercial high protein monkey chow, bananas, apples and oranges. On the remaining two days, a liquid diet was offered (2 cups sugar, 16 oz. high protein baby cereal, 24 oz. orange juice concentrate, 4 eggs with shells, 4 bananas, 8 cc multivitamin solution [Vi-Daylin, Ross], 1 cc vitamin B₁₂, 2 folic acid tablets, and water to make 1 gallon). From 1981 to the present, the daily diet consisted of a mixture of moistened commercial high protein monkey chow and commercial marmoset diet, supplemented with bananas, apples, oranges, carrots, cooked rice and oatmeal, and high protein baby cereal. Water was available continuously.

Parasite strains and related procedures

Colombian owl monkey-adapted *P. falciparum* strains were used in these studies: Uganda Palo Alto, Vietnam Oak Knoll, and Vietnam Smith. Both the Vietnam Oak Knoll and Vietnam Smith strains were obtained through the courtesy of Dr. Leon H. Schmidt, University of Alabama at Birmingham, Birmingham, Alabama. Dr. Alister Voller, Nuffield Institute of Comparative Medicine, London, England, kindly provided the Uganda Palo Alto strain. All strains were received as cryopreserved blood.

The Vietnam Oak Knoll and Vietnam Smith strains were each inoculated intraperitoneally into a splenectomized Colombian owl monkey while

the Uganda Palo Alto strain was inoculated into an intact Panamanian owl monkey. Once parasitemias were established in the initial recipients, parasites were serially passaged in intact Panamanian monkeys.

Donor monkeys were bled (0.5 to 1.0 ml) from the femoral vein or artery, using 0.1 ml of a 2.5% solution of sodium citrate as anticoagulant. Infected blood was diluted with sterile chilled normal saline so that 1.0 ml contained either 5×10^4 or 5×10^6 parasites. Inoculum was injected into the saphenous vein of each recipient to induce infection.

To reduce monkey requirements for strain maintenance, an inoculum of 5×10^4 parasites was used. Animals serving as untreated controls for drug evaluation studies were inoculated with 5×10^6 parasites. The prepatent period in the Panamanian owl monkeys that received 5×10^4 parasites was longer than in those inoculated with 5×10^6 parasites. Data presented in Table 1 indicate infection parameters in the monkeys inoculated with either 5×10^4 or 5×10^6 parasites of the Uganda Palo Alto, Vietnam Smith, or Vietnam Oak Knoll strain of *P. falciparum*. The peak parasitemias and the time required for their attainment were equivalent in monkeys, dying or surviving, after inoculation with either number of parasites. The patent day of death was also similar between the two groups infected with one of the three strains of falciparum malaria.

Blood was collected by lancing the marginal ear vein, and films were stained with Giemsa. Evaluation of parasitemia was as follows: negative, if no parasites were seen after examining a thick blood film for at least five minutes; and <10 parasites/mm³, if parasites could be demonstrated only on a thick blood film. The number of parasites/mm³ was determined by the Earle-Perez method.⁷

Blood films were prepared daily from the day after inoculation until parasites could no longer be detected for \geq five consecutive days. Thereafter, films were made twice weekly. If a recrudescence occurred, blood films were again prepared on a daily basis. The infection was considered "cured" if no parasites were observed for ≥ 100 days.

Conversion of parasite counts per 10⁴ erythrocytes to number of parasites per mm³

Maximum parasitemias in Colombian owl monkeys were published as the number of par-

TABLE I
Effect of inoculum size on infection parameters of three strains of *Plasmodium falciparum* in Panamanian owl monkeys

Strain	Inoculum		No. monkey inoc.	Fatal infections				Self-limited infections			
	No. troph.	No. monkey inoc.		No.	Days* death (±SD)	Peak par. no./mm ³ × 10 ³ (±SD)	Day of peak (±SD)	No.	Peak par. no./mm ³ × 10 ³ (±SD)	Day of peak (±SD)	No. without recrud.
Vietnam Oak Knoll	5 × 10 ⁴	43	34	14.1 (7.7)	894 (453)	9.5 (2.9)	9	691 (380)	9.7 (2.4)	7	2
	5 × 10 ⁶	59	36	14.8 (7.4)	886 (416)	9.4 (3.1)	23	595 (278)	9.7 (2.6)	15	8
Vietnam Smith	5 × 10 ⁴	66	19	14.4 (3.5)	1,148 (486)	10.6 (1.5)	47	670 (457)	11.8 (3.6)	22	25
	5 × 10 ⁶	106	28	13.6 (3.5)	1,058 (602)	10.4 (2.1)	78	606 (381)	10.6 (2.0)	50	28
Uganda Palo Alto	5 × 10 ⁴	53	24	12.8 (3.8)	1,385 (634)	9.6 (2.3)	29	729 (438)	9.2 (2.4)	22	7
	5 × 10 ⁶	68	16	13.0 (4.4)	1,330 (416)	9.5 (1.3)	52	654 (375)	9.5 (2.9)	45	7

* Day after onset of patent parasitaemia.

asites per 10⁴ erythrocytes.² Normal Colombian owl monkeys were considered to have a hematocrit of 45 ± 3% and 5 × 10⁶ erythrocytes per mm³ (L. H. Schmidt, University of Alabama, personal communication). At the time of maximum parasitemia the mean hematocrit, based upon an unspecified number of spot checks, was 15 ± 3%, or 1.67 × 10⁶ erythrocytes per mm³, and this figure was used to convert number of parasites per 10⁴ erythrocytes to number of parasites per mm³.

Statistical evaluation

We used a two-tailed *t*-test to compare the population means for various parameters in Panamanian and Colombian owl monkeys, respectively. We assumed that the samples were independent and the sample means were unbiased estimates of the population means.

RESULTS

Effects of inoculum size

The data presented in Table I show the influence of inoculum size upon diverse infection parameters of three *P. falciparum* strains in Panamanian owl monkeys. The peak parasitemia and the day of its occurrence after the onset of patency were essentially identical in monkeys inoculated with either 5 × 10⁴ or 5 × 10⁶ trophozoites of the Uganda Palo Alto, Vietnam Smith and Vietnam Oak Knoll strains, irrespective of whether the monkeys died or survived the parasite challenge. Additionally, the inoculum size had no effect upon the day of death after the onset of patency.

The effect of inoculum size on mortality rate was variable. For the Vietnam Smith strain the mortality rate of monkeys inoculated with 5 × 10⁴ or 5 × 10⁶ parasites was essentially identical, 28.8% and 26.4%, respectively. The mortality rate of monkeys inoculated with 5 × 10⁴ parasites of the Vietnam Oak Knoll strain was apparently greater (79.1%) than those inoculated with 5 × 10⁶ parasites (61%), but this difference was not significant, (*P* = >0.73). There was a significant difference (*P* = <0.025) between the mortality rates of monkeys inoculated with 5 × 10⁴ parasites or 5 × 10⁶ parasites of the Uganda Palo Alto strain, 45.3% and 23.5%, respectively.

The data in Table I also indicate that, in the case of the Vietnam Smith and Uganda Palo Alto

TABLE 2

Mortality of Panamanian and Colombian owl monkeys following infection with three strains of *Plasmodium falciparum*

Strain	Total	Number of monkeys (Percent)			Survivors Self-limited infections
		Malaria fatalities			
		Patent day			
		<15	>15-30	≥30	
Panamanian owl monkeys					
Vietnam Oak Knoll	102	57 (55.9)	9 (8.8)	4 (3.9)	32 (31.4)
Vietnam Smith	172	35 (20.3)	12 (7.0)	—	125 (72.7)
Uganda Palo Alto	121	32 (26.4)	8 (6.6)	—	81 (67.0)
Colombian owl monkeys*					
Vietnam Oak Knoll	283	149 (52.7)	84 (29.7)	17 (6.0)	33 (11.7)
Vietnam Smith	156	46 (29.5)	41 (26.3)	6 (3.8)	63 (40.4)
Uganda Palo Alto	226	133 (58.8)	46 (20.4)	8 (3.5)	39 (17.3)

* The data in all tables for Colombian owl monkeys were derived from a previous publication.⁷

strains, fewer recrudescences occurred among monkeys that had received the larger inoculum.

The infection parameter data for the two groups of Panamanian owl monkeys were combined for presentation in Tables 2 through 6.

Mortality

A comparison of the mortality of the two owl monkey subspecies to three strains of *P. falciparum* is indicated in Table 2. Overall, 157 (39.7%) Panamanian owl monkeys died during patency in contrast with 530 (79.7%) Colombian owl monkeys.

The percentage of deaths attributable to infection with the Vietnam Oak Knoll strain, during the first 15 days of patency, was approximately the same for Panamanian and Colombian owl monkeys. After patent day 16, fewer Panamanian owl monkeys than Colombian owl monkeys succumbed to infection with the Vietnam Oak Knoll strain. A total of 70 (68.6%) Panamanian monkeys and 250 (88.3%) Colombian owl monkeys died of infection with this strain of *P. falciparum*.

Infections of the Vietnam Smith strain were less virulent in the Panamanian owl monkey than in the Colombian owl monkey. During the first 30 days of patency, 47 (27.3%) Panamanian owl monkeys died, while during this same period of

patency 87 (55.8%) of the Colombian owl monkeys died. No deaths of Panamanian monkeys occurred after patent day 30; six Colombian owl monkeys died during this period.

As indicated by the number of monkeys that died during the first 30 days of patency of the Uganda Palo Alto strain, infections were less lethal for Panamanian owl monkeys (33%) than for Colombian owl monkeys (79.2%). After patent day 30, no Panamanian monkeys died, while eight Colombian owl monkeys died.

The percentage of Panamanian owl monkeys with self-limited infections was greater than that of Colombian owl monkeys for each of the three *P. falciparum* strains.

Parasitemias

The peak parasitemias in Panamanian and Colombian owl monkeys that died during patency and in the survivors with self-limited infections are shown in Table 3. Panamanian owl monkeys infected with the Vietnam Oak Knoll strain, that succumbed during the first 15 days of patency or between patent day 16 to 30, had lower mean peak parasitemias than Colombian owl monkeys. There was, however, no significant difference between the parasitemias of the two groups of owl monkeys, $P = <0.6$ and $P = <0.2$, respectively.

TABLE 3

Peak parasitemias in infections with three strains of *Plasmodium falciparum* in Panamanian and Colombian owl monkeys

Strain	Peak parasitemia per mm ³ × 10 ³ —mean (±SD)			Survivors Self-limited infections
	Malaria fatalities			
	Patent day		≥ 30	
≤ 15	> 15–30	≥ 30		
Panamanian owl monkeys				
Vietnam Oak Knoll	975 (427)	527 (243)	496 (45)	622 (314)
Vietnam Smith	1,297 (485)	629 (400)	—	630 (412)
Uganda Palo Alto	1,510 (488)	776 (415)	—	681 (401)
Colombian owl monkeys				
Vietnam Oak Knoll	1,007 (374)	645 (301)	293 (116)	243 (147)
Vietnam Smith	921 (407)	689 (301)	496 (371)	213 (90)
Uganda Palo Alto	1,264 (591)	1,055 (523)	573 (204)	385 (221)

The mean peak parasitemia in Panamanian owl monkeys that died during the initial 15-day patent period after challenge with the Vietnam Smith strain was significantly higher ($P = <0.001$) than in the comparable group of Colombian owl monkeys. Panamanian and Colombian monkeys, infected with the Vietnam Smith strain, that died during patent days 16 to 30 had essentially the same peak parasitemias ($P = >0.5$).

The mean peak parasitemia for Uganda Palo Alto infections in Panamanian monkeys that died during the first days of patency was significantly higher ($P = <0.05$) than in Colombian owl monkeys. For the monkeys that died during patent days 16 to 30, the mean peak parasitemia was lower in Panamanian than in Colombian monkeys, but not statistically significant ($P = >0.1$).

As indicated in Table 3, the Panamanian owl monkeys that survived the primary attack with each *P. falciparum* strain, had higher parasitemias than did the Colombian owl monkeys. These differences were highly significant ($P = <0.001$).

Time to attain peak parasitemias

The infection parameter presented in Table 4 compares the time required to achieve peak parasitemia, once patency was established, in Panamanian and Colombian owl monkeys. Regard-

less of the strain of *P. falciparum*, fatality during the primary attack, or survival, as indicated by self-limited infection, peak parasitemias were attained significantly earlier in Panamanian owl monkeys than in Colombian owl monkeys. In all instances, except one, the P values were <0.001 . The exception ($P = <0.05$) was for monkeys infected with the Vietnam Oak Knoll strain that died during patent days 16 to 30.

Self-limited infections

The distribution of monkeys with self-limited infections attained with either the primary attack or recrudescence is indicated in Table 5. A total of 148 Panamanian owl monkeys (62.2%) evidenced self-limited infections following the primary attack, while infections in 78 (57.8%) Colombian owl monkeys were self-limited with the primary attack. Following the first recrudescence, infections were self-limited in 64 (26.9%) of the Panamanian owl monkeys and in 39 (28.9%) of the Colombian owl monkeys.

Self-limited infections after the second recrudescence were achieved in 24 (23.8%) Panamanian and 13 (9.6%) Colombian owl monkeys. The infection (Vietnam Smith) in one (0.4%) Panamanian owl monkey was self-limited following the third recrudescence, in contrast to five (3.7%) in Colombian monkeys.

An Uganda Palo Alto infection in one (0.4%)

TABLE 4

Time to attain peak parasitemias in infections with three strains of *Plasmodium falciparum* in Panamanian and Colombian owl monkeys

Strain	Day from initiation of patency to peak — mean (\pm SD)			Survivors Self-limited infections
	Malaria fatalities			
	Patent day			
	≤ 15	$> 15-30$	≥ 30	
Panamanian owl monkeys				
Vietnam Oak Knoll	8.9 (1.6)	14.0 (5.3)	7.8 (1.3)	9.7 (2.6)
Vietnam Smith	10.4 (1.8)	11.2 (2.1)	—	11.1 (2.8)
Uganda Palo Alto	9.3 (1.8)	10.6 (2.3)	—	9.4 (2.7)
Colombian owl monkeys				
Vietnam Oak Knoll	11.4 (1.6)	16.6 (2.9)	17.0 (2.9)	16.2 (4.3)
Vietnam Smith	11.7 (1.4)	17.3 (2.7)	21.3 (7.4)	14.4 (4.0)
Uganda Palo Alto	10.5 (1.7)	15.7 (3.1)	13.4 (3.3)	14.9 (4.0)

Panamanian owl monkey was self-limited after the fourth recrudescence; all infections in Colombian owl monkeys were self-limited after the third recrudescence.

While the number of Panamanian and Colombian owl monkey survivors, following primary attack of infection with the Vietnam Oak Knoll strain, was essentially the same (32 and 33, re-

spectively), the infection in 12 Panamanian owl monkeys was self-limited after the primary attack as it was in 18 Colombian owl monkeys. More Panamanian than Colombian owl monkeys experienced self-limited infection after the first and second recrudescence of the Vietnam Oak Knoll strain. The infection in one Panamanian owl monkey was self-limited after the

TABLE 5

Self-limited infections in Panamanian and Colombian owl monkeys infected with one of three strains of *Plasmodium falciparum*

Strain	Total	No. (percent) self-limited infections attained at				
		Primary attack	Recrudescence			
			1	2	3	4
Panamanian owl monkeys						
Vietnam Oak Knoll	32	12 (37.5)	12 (37.5)	7 (21.9)	—	1 (3.1)
Vietnam Smith	125	70 (56.0)	40 (32.0)	14 (11.2)	1 (0.8)	—
Uganda Palo Alto	81	66 (81.5)	12 (14.8)	3 (3.7)	—	—
Colombian owl monkeys						
Vietnam Oak Knoll	33	18 (54.6)	9 (27.3)	3 (9.0)	3 (9.0)	—
Vietnam Smith	63	35 (55.6)	22 (34.9)	5 (7.9)	1 (1.6)	—
Uganda Palo Alto	39	25 (64.1)	8 (20.5)	5 (12.8)	1 (2.6)	—

TABLE 6
Infection characteristics in Panamanian and Colombian owl monkeys infected with one of three strains of Plasmodium falciparum and exhibiting self-cure

Strain	Patent period: mean no. days (\pm SD)/no. monkeys				Subpatent period: mean no. days (\pm SD)				Mean peak parasitaemia/mm ³ $\times 10^4$ (\pm SD)				
	Recrudescence				Recrudescence				Recrudescence				
	Primary attack	1	2	3	4	Primary at- tack-1	1-2	2-3	3-4	1	2	3	4
Panamanian owl monkeys													
Vietnam	31.0	10.7	14.4	—	11.0	32.8	26.1	11.0	31.0	64	16	0.24	1.0
Oak Knoll	(8.0)/12	(2.1)/12	(4.3)/7	—	(-)/1	(16.9)	(9.1)	(-)	(-)	(71)	(33)	(-)	(-)
Vietnam	25.0	11.6	13.6	9.0	—	33.3	25.2	38.0	—	28	7	0.5	—
Smith	(4.4)/70	(4.2)/40	(3.6)/14	(-)/1	—	(14.6)	(8.1)	(-)	—	(58)	(13)	(-)	(-)
Uganda	24.1	9.5	9.7	—	—	40.2	32.0	—	—	44	5	—	—
Palo Alto	(4.8)/66	(3.8)/12	(1.7)/3	—	—	(14.2)	(3.0)	—	—	(70)	(6)	—	—
Colombian owl monkeys													
Vietnam	45.1	19.9	19.5	21.3	—	20.9	24.2	17.7	—	13	6	4	—
Oak Knoll	(23.5)/18	(5.9)/9	(8.3)/3	(7.0)/3	—	(8.3)	(9.6)	(3.1)	—	(9)	(4)	(3)	—
Vietnam	31.6	19.5	18.2	16.0	—	22.4	23.7	14.0	—	9	1.0	0.3	—
Smith	(3.7)/35	(5.4)/22	(5.2)/5	(-)/1	—	(9.2)	(9.9)	(-)	—	(6)	(0.8)	(-)	—
Uganda	33.1	19.4	12.8	13.0	—	23.6	29.0	36.0	—	53	3	0.2	—
Palo Alto	(4.6)/25	(5.5)/8	(2.0)/5	(-)/1	—	(10.3)	(12.0)	(-)	—	(27)	(3)	(-)	—

fourth recrudescence; three Colombian monkeys attained self-limited infection status following the third recrudescence.

Panamanian and Colombian monkeys infected with the Vietnam Smith strain had a similar rate of self-limited infection after the primary attack. A total of 55 (44%) Panamanian owl monkeys and 28 (44.4%) Colombian monkeys, following recrudescences 1, 2 and 3, had self-limited infections.

Approximately twice as many Panamanian owl monkeys as Colombian owl monkeys survived the primary attack of the Uganda Palo Alto strain. The infection in 66 Panamanian owl monkeys was self-limited with the primary attack, in contrast to 25 Colombian monkeys. Self-limited infection after recrudescences 1 and 2 was observed in 15 Panamanian monkeys, and in 14 Colombian owl monkeys after recrudescences 1 through 3.

Patent and subpatent periods

Diverse infection characteristics associated with recrudescing infections, length of the primary attack and recrudescences, interval between recrudescences, and peak parasitemias are compared in Table 6. Although the mean duration of the primary attack in Panamanian owl monkeys infected with the Vietnam Oak Knoll strain was 14 days shorter than in similarly infected Colombian monkeys, this difference was not significant ($P = >0.05$). The length of the first recrudescence of the Vietnam Oak Knoll strain in Panamanian monkeys was significantly shorter ($P = <0.001$) than in Colombian owl monkeys, but the difference for the length of second recrudescence was not significant ($P = >0.2$).

Primary attacks and first recrudescences were of significantly ($P = <0.001$) shorter duration in Panamanian owl monkeys infected with the Vietnam Smith strain than in Colombian owl monkeys, as was the length of the second recrudescence ($P = <0.05$).

A comparison of the mean duration of the primary attack and first recrudescence of infections with the Uganda Palo Alto strain indicated that both of these parameters were significantly ($P < 0.001$) shorter in Panamanian monkeys than in Colombian owl monkeys. The duration of the second recrudescence with this strain was shorter in Panamanian monkeys than in Colombian monkeys, but not significantly so ($P > 0.05$).

A single Panamanian owl monkey, infected with the Vietnam Oak Knoll strain, experienced self-cure after the fourth recrudescence, while three Colombian owl monkeys had self-limited infections following the third recrudescence with the Vietnam Oak Knoll strain. One Panamanian owl monkey, infected with the Vietnam Smith strain, had a third recrudescence of nine days duration, in contrast with a Colombian owl monkey, in which a third recrudescence lasted 16 days. No Panamanian owl monkeys, infected with the Uganda Palo Alto strain, exhibited a third recrudescence.

The subpatent periods between the end of the primary attack and the beginning of the first recrudescence, and between the first and second recrudescence in Panamanian owl monkeys infected with the Vietnam Oak Knoll strain, were longer than in Colombian owl monkeys. These differences, however, were not significant, $P > 0.05$ and $P < 0.8$, respectively.

For infections of the Vietnam Smith strain in Panamanian owl monkeys, the subpatent period between the primary attack and first recrudescence was significantly longer ($P < 0.01$) than in Colombian monkeys. There was no significant difference ($P < 0.8$) between the durations of the subpatent period between the first and second recrudescence.

Comparison of the subpatent periods following infection of the Uganda Palo Alto strain indicated that the time between the end of the primary attack and the first recrudescence was significantly longer ($P < 0.02$) in the Panamanian than in the Colombian owl monkey. The subpatent period between the first and second recrudescence was not significantly different ($P < 0.7$) between the two monkey subspecies.

Peak parasitemias during recrudescence

Peak parasitemias for recrudescences in Panamanian and Colombian owl monkeys are presented in Table 6. The mean peak parasitemias in Panamanian owl monkeys were higher than comparably infected Colombian monkeys during the first recrudescence of infection with the Vietnam Oak Knoll or Vietnam Smith strains, but not for infection with the Uganda Palo Alto strain. During the second recrudescence, the mean peak parasitemia in Panamanian owl monkeys was greater than in Colombian owl monkeys for

each of the three strains. Because the standard deviations were greater than the mean peak parasitemias in Panamanian monkeys, no meaningful statistical analysis was possible.

DISCUSSION

The principal objective of this report was to present a quantitative comparison between the course of untreated *P. falciparum* infections (three strains) in the two owl monkey subspecies, of distinct geographic origin, which are the most frequently used for experimental human malarial studies. Similarities and differences of infection parameters in Panamanian and Colombian owl monkeys may be summarized as follows:

1. The Panamanian owl monkey was less susceptible to infection with each of the three strains as mortality during the first 30 days of patency was less than in the Colombian monkey. Only during the first 15 days of patent infection of the Vietnam Oak Knoll strain did more Panamanian than Colombian monkeys die of malaria.
2. Peak parasitemias in Panamanian owl monkeys, infected with either the Vietnam Smith or Uganda Palo Alto strain and dying during the first 15 days of patency, were significantly higher than in Colombian owl monkeys that died during the same period. All Panamanian monkeys that survived the primary attack of the three strains had significantly higher maximum parasitemias than did surviving Colombian owl monkeys.
3. Peak parasitemias, either in fatal or self-limited infections, were achieved significantly earlier after patency in Panamanian than in Colombian owl monkeys, regardless of the strain of *P. falciparum*.
4. After the primary attack of either the Vietnam Smith or Uganda Palo Alto strain, more Panamanian than Colombian owl monkeys evidenced self-limited infection. Self-limited infections occurred more frequently in Panamanian than in Colombian monkeys after the first recrudescence, regardless of the malaria strain. Following the second recrudescence, more Panamanian than Colombian owl monkeys, infected with either the Vietnam Oak Knoll or Vietnam Smith strain, experienced a self-limited infection.
5. The duration of the primary attack and the first and second recrudescences were significantly shorter in Panamanian than in Colombian owl monkeys, except for the duration of the primary attack and second recrudescence of Vietnam Oak Knoll infection.

The subpatent periods between the end of the primary attack and first recrudescence and between the first and second recrudescences were longer in Panamanian than in Colombian monkeys. Only the duration of the subpatent period between the primary attack and first recrudescence of the Vietnam Smith and Uganda Palo Alto strains was significantly greater in Panamanian owl monkeys.

The mean maximum parasitemias in Panamanian owl monkeys, during the first and second recrudescences of the Vietnam Oak Knoll and Vietnam Smith strain, were higher than in Colombian monkeys. During the first recrudescence of the Uganda Palo Alto strain, the mean maximum parasitemia was lower in Panamanian than in Colombian owl monkeys, and during the second recrudescence of this strain, the maximum parasitemias were essentially the same in both monkeys.

The basis for diversities of infection parameters in these two owl monkey subspecies has not been defined, however, two factors may be excluded as having affected the observations presented in this report. No naturally-acquired plasmodium infections, of either human or monkey origin, have been reported^{2,8} in owl monkeys, thus eliminating the possibility of immunity acquired prior to experimental use. Comparison of husbandry practices for the Panamanian owl monkey with those outlined¹ for the Colombian owl monkey indicate that while qualitative differences exist, none can account for the variations of infection parameters between the two monkeys.

Few direct comparisons of experimentally-induced *P. falciparum* infections in owl monkeys of Panamanian and Colombian origin have been published. In one such report,⁹ four of four Panamanian owl monkeys and seven of nineteen Colombian owl monkeys survived infection with the Uganda Palo Alto strain. Inoculation of Vietnam Oak Knoll parasites into 10 owl monkeys of Panamanian origin produced potentially lethal infections in all recipients. Of three Colombian owl monkeys infected with the Vietnam Oak Knoll strain, one animal survived. The authors

concluded that the country of origin of the owl monkeys, rather than the karyotype, determined host susceptibility to lethal malarial infections.

Our experience with the Panama II strain illustrates that owl monkey karyotype may not be the sole determinant of susceptibility to *P. falciparum* strains. In attempts to adapt *P. falciparum* from Panamanian patients, 75 indigenous owl monkeys were inoculated with human blood.¹⁰ No infections were reproduced in any of the monkeys. A blood sample from a patient who had acquired falciparum malaria in Panama was sent to another laboratory where the Panama II strain was established in a Colombian owl monkey.¹¹ We inoculated parasites from the second infected Colombian monkey into a splenectomized Panamanian owl monkey and subsequently established infections in normal Panamanian owl monkeys (personal communication). What parasite modification(s) occurred, in this Panamanian strain during two passages in Colombian owl monkeys, that facilitated adaptation to Panamanian owl monkeys is unknown.

In another report,¹² each of three normal Colombian owl monkeys was inoculated intravenously with 7.5×10^5 parasites of the Uganda Palo Alto strain: one monkey died on day 8 after challenge with a parasitemia of 60%, another monkey died on day 15 after inoculation with a parasitemia of 54.4%, and the third monkey survived, with a maximum parasitemia of 25%. This 66% mortality rate during the first 15 days of patency is comparable to that (58.8%) reported² for a group of 226 Colombian owl monkeys infected with the Uganda Palo Alto strain, and greater than the 26.4% mortality in a group of 121 Panamanian owl monkeys.

Although almost 20 years have elapsed since the report¹³ of the first successful adaptation of *P. falciparum* to an owl monkey from Colombia, no definitive explanation has been forthcoming as to why monkeys of the genus *Aotus* are susceptible to falciparum malaria. This susceptibility, as demonstrated in the present report, varies quantitatively but moderately so, according to the country of origin of both the owl monkey and the strain. These differences in infection parameters between Panamanian and Colombian owl monkeys have not invalidated the use of the former for the evaluation of new antimalarial drugs.

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