HUMAN MALARIA (PLASMODIUM FALCIPARUM) IN OWL MONKEYS (AOTUS TRIVIRGATUS)

by

A. Voller,¹ W. H. G. Richards,² C. M. Hawkey,¹ and D. S. Ridley³

1. Introduction

Direct studies on the human malaria parasites, in particular on Plasmodium falciparum, for which there is no close suitable simian analogue, have been hampered in the past for want of a convenient experimental host.

Taliaferro & Taliaferro (1934) were able to get fleeting P. falciparum parasitaemias in the infant howler monkey (Alouatta spp.) but this animal is not readily available, and is unsuited to captivity (Napier & Napier, 1967).

The splenectomized chimpanzee has also proved susceptible to P. falciparum (Bray, 1958; Rodhain & Jadin, 1964) and has been of value in the elucidation of the exo-erythrocytic cycle of P. falciparum (Bray, 1963) and for chemotherapeutic (Hickman et al., 1966) and immunological studies (Sadun et al., 1966).

The increasing scarcity of chimpanzees raises unopposable moral restrictions on their further employment for all but absolutely essential experimentation. In the last few years the search for new hosts for P. falciparum showed the gibbon (Hylobates lar) to be susceptible to P. falciparum (Ward & Cadigan, 1966) but the infections as reported to date do not closely parallel those in man. Following an extensive survey of Panamanian primates, Porter & Young (1966) found that owl monkeys were susceptible to P. vivax. A year later Guiman & Meagher (1967) showed that this same species of primate would also support infections of P. falciparum.

2. Materials and methods

Aotus trivirgatus, the owl monkey, also known as the night monkey or feline dourocculi, was the experimental host in the present studies. This small monkey is abundant over a wide area in the forests of South and Central America.

We have encountered considerable problems in the maintenance of these animals in captivity, particularly in specimens newly imported. In part the early mortality can be attributed to respiratory tract infections. Some workers in the United States of America (Esparza, personal communication) found generalized infections due to a virus similar to Herpes simplex in their Aotus colonies. There has been no evidence of such infection in our animals in that inoculation of material from moribund specimens into tissue culture and other experimental animals.

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has not resulted in isolation of any viral agents. Extremely abnormal blood coagulation patterns were discovered in one group of three of our animals. These abnormalities characterized by prolonged prothrombin and cephalin times and reductions in factors II, VII, IX and X were corrected by administration of vitamin K. In spite of this therapy this group of animals all succumbed within two months following a severe anaemia which did not respond to vitamin B12, folic acid or iron treatment. At the time of death, all blood coagulation tests were normal but the blood urea and glutamic-oxaloacetic transaminase raised. Post mortem examination indicated extensive centri-lobular necrosis of the liver. The etiology of this disease is, as yet, unknown.

Once these monkeys have been established for a month or two there are few further maintenance problems, although an adequate source of vitamin D3 is essential. A few specimens have been bred in captivity (Int. Zoo Year Book, 1965).

Throughout the present studies the animals were maintained at 75-85°F (23.9-29.4°C) either on a diet of Purina Chow or on a mixture of fruit, eggs, milk, rice and concentrated protein supplement.

Before experimentation the animals were caged in groups of two or three and were transferred to individual cages throughout the experiment. Handling of the monkeys was facilitated by the use of a small perspex catching cage in which they were lightly anaesthetized with halothane. This procedure is safer than frequent anaesthesia with phencyclidine and is preferable to catching the animals by hand which results in their undue excitement.

Infections. The infections were initiated by the intravenous inoculation of P. falciparum infected blood. Several attempts were made to transfer infections directly from man to splenectomized owl monkeys (Table 1) but all were unsuccessful. The studies to be reported here are based on infections of the Camp strain of P. falciparum which originated in West Malaysia and has since been blood passaged in chimpanzees and owl monkeys.

All the infections to be described were initiated by transfer of infected blood either directly from one owl monkey to another or with similar material kept at -70°C with glycerol as a preservative.

**TABLE 1. ATTEMPTS TO INITIATE HUMAN MALARIA PARASITE INFECTIONS IN OWL MONKEYS**

<table>
<thead>
<tr>
<th>Parasite species</th>
<th>Geographical origin</th>
<th>Donor</th>
<th>History of inoculum</th>
<th>Comment</th>
<th>Patent infection in recipient</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. P. falciparum</td>
<td>North America</td>
<td>(Man)</td>
<td>Stored at -70°C</td>
<td>Viability suspect</td>
<td>-</td>
</tr>
<tr>
<td>2. P. falciparum</td>
<td>West Africa</td>
<td>(Man)</td>
<td>Stored at -70°C</td>
<td>Viability suspect</td>
<td>-</td>
</tr>
<tr>
<td>3. P. falciparum</td>
<td>Zambia</td>
<td>(Man)</td>
<td>Stored at 40°C for 24 hrs</td>
<td>Probably viable</td>
<td>-</td>
</tr>
<tr>
<td>4. P. falciparum</td>
<td>Tanzania</td>
<td>(Man)</td>
<td>Injected into monkeys after 4 hrs at 20°C</td>
<td>Probably viable</td>
<td>-</td>
</tr>
<tr>
<td>5. P. falciparum</td>
<td>Tanzania</td>
<td>(Man)</td>
<td>Injected into monkeys after 3 hrs at 20°C</td>
<td>Probably viable</td>
<td>-</td>
</tr>
<tr>
<td>6. P. malariae</td>
<td>Romania</td>
<td>(Man)</td>
<td>Stored at 40°C for 18 hrs</td>
<td>Probably viable</td>
<td>-</td>
</tr>
<tr>
<td>7. P. falciparum</td>
<td>Malaya</td>
<td>(Owl monkey)</td>
<td>Stored at 40°C for 18 hrs</td>
<td>Viable</td>
<td>+</td>
</tr>
</tbody>
</table>

1 This strain was obtained by the courtesy of Dr E. H. Sadun, Walter Reed Army Medical Center, Washington D.C., United States of America.
Parasitology. The course of the malaria infection was monitored by examination of thick and thin blood films, made by ear-prick stained with Giemsa and the parasitaemia was expressed as the number of parasites per 10^6 red blood cells. Rectal temperatures were taken daily.

Haematology. Larger quantities of blood for the serial haematological studies were obtained from the femoral vein.

Packed cell volume was determined using capillary tubes on a micro-haematocrit centrifuge, and haemoglobin by oxyhaemoglobin spectrophotometry. Because only relatively small volumes of blood were available the other tests were limited to platelet counts on five animals and cephalin times, fibrinogen estimations, assays of factors V and VIII and euglobulin lysis times to a single animal.

Platelets were counted by the method of Brecher & Cronkite (1950) using phase contrast microscopy, prothrombin times measured by the method of Quick (1957), cephalin time by the method of Rodman, Barrow & Graham (1958), fibrinogen by the method of Ratnoff & Menzie (1951), Factor V by the method of Stefanini (1950) modified by Wolf (1953), and Factor VIII by the method of Hardisty & Macpherson (1962) and euglobulin lysis times by the method of Buckell (1958). The brain thromboplastin, cephalin and Factor V and VIII deficient substrate plasmas used for these tests was derived from human sources.

Pathology. Post-mortem specimens were removed within five minutes of death and transferred at once to formal saline. Paraffin sections were stained with haematoxylin and eosin.

3. Results and discussion

The failure to achieve successful transfers from man to monkey was probably due to loss of viability of the inoculum during transport from the malaria-endemic areas. Such inocula would in all probability be infectious for man but the initial transfer to the different host probably requires a much greater inoculum. All the successful transfers to owl monkeys of the erythrocytic stages of human malaria have been accomplished when there was immediate transfer from human donor to monkey recipient of a massive inoculum (Reid, personal communication; Geiman & Meagher, 1967).

Morphology of parasites. During the early development of the parasites cycle, accole or applique forms, that is small ring forms applied to the periphery of the red cell (Garnham, 1966) were extremely common, as were small rings with two chromatin masses (Fig. 1a). As the parasitaemia mounted multiple parasitization of red cells was often seen, but it was no more frequent than expected mathematically for independent invasion. Table 2 shows the actual observed figures for random invasion of uninfected and parasitized red cells on the basis of the total observed parasitaemia. This is further evidence against binary fission as a cause of multiple infection of single cells. Reticulocytes and mature red cells were invaded to the same degree.

| TABLE 2. THE DEGREE OF MULTIPLE INFECTION OF SINGLE RED CELLS IN P. FALCIPARUM INFECTIONS IN TWO OWL MONKEYS |
|-----------------------------|-----------------------------|-----------------------------|-----------------------------|-----------------------------|
| Total cells counted | Total infected cells | Single infection of cells | Double infection of cells* | Triple infection of cells |
| Sample 1 | 1 000 | 378 | 266 | 83 (71) | 29 (19) |
| Sample 2 | 1 000 | 101 | 93 | 7 (8) | 1 (1) |

* The figures in brackets are those expected in uninfected red cells and those already infected are equally susceptible to reinvasion.
As the parasites matured and reached the late trophozoite stage, Maurer's clefts became visible in the infected cells and the margins of the cells stained especially heavily.

Although all asexual stages of the parasite, up to and including the mature schizonts (Geiman & Meagher, 1967) are found in the peripheral blood of owl monkeys, there is nevertheless a considerable retreat to the deeper organs as schizogony approaches (Table 3).

Nearing the terminal phases of the infection, mature schizonts become readily detectable in the peripheral blood (Fig. 1b). We have observed between 10 and 37 merozoites per schizont. In Fig. 2 the observed frequency of different numbers of merozoites per schizont is given. For these purposes only mature schizonts with a single pigment mass are considered, in order that cells with more than one parasite should be excluded. The mean number of merozoites per schizont is 21.6, which is close to that reported for human *P. falciparum* infections in Malaya (Field & Shute, 1956). Bray (1958) noted that an African strain which had 12 merozoites per schizont in man had more than double this number in the chimpanzee. Such differences between different strains and hosts may more closely reflect the stage of schizogony at which withdrawal to the internal organs occurs in any particular host-strain combination rather than being a characteristic of the strain only. The finding of Peel & van Hoof (1948) that in the placenta the mean merozoite number is higher than in the peripheral blood would support the thesis that most of the schizonts in the peripheral blood are immature.

Gametocytes were extremely scanty and they were seen on only two occasions in the present studies, and each time they were degenerate. Repeated attempts to infect *Anopheles gambiae* met with complete failure. We feel that these failures can be related to the history of repeated blood passage of this particular strain of *P. falciparum*, rather than to any influence exerted by the host, although the development and infectivity of gametocytes of *P. falciparum* is reduced or abolished in chimpanzees (Bray, 1958) and gibbons (Ward & Cadigan, 1966). Collins & Contacos (1968) have now achieved mosquito passage of a more recently isolated strain of *P. falciparum* from owl monkeys to man and vice versa, although they too were unable to transmit the Camp strain in this way.

**Course of infection.** The parasitaemia and temperature measurements in three untreated blood induced infections of *P. falciparum* in owl monkeys infected with 2000 parasites are given in Fig. 3. The parasite multiplication rate was similar in all three and represents a ten-fold increase in parasitaemia over each 48-hour cycle. In all the owl monkeys we studied, the fulminating infections of this adapted strain of *P. falciparum* led to death unless halted by chemotherapy.

The first untreated infection of *P. falciparum* in non-immune humans results in an acute attack terminated by death, or as is more usual, by immunological recovery. The onset of the disease is marked by recurrent febrile episodes. These may be of the classical tertian type, i.e. every second day, but Shute (personal communication) says that in his experience this is rare and quotidian fevers are the rule in the primary attack, and result from the sporulation of minor broods of the parasite.

In man the parasite density at the onset of fever, the mean fever threshold in non-immune, is given as about $10^4$ parasites/mm$^3$ (Kitchen, 1949) although in some cases it is as high as $10^5$/mm$^3$. The observed fever threshold in our monkeys was $5 \times 10^2$/mm$^3$, $1.85 \times 10^4$/mm$^3$, $8.45 \times 10^4$/mm$^3$, $1.20 \times 10^5$/mm$^3$, and $2.37 \times 10^5$/mm$^3$, which corresponds closely to the human levels. The first clearly defined temperature increase which occurred in all the infected animals did not reach $100^\circ F$ (37.8°C). The subsequent three to four febrile episodes which occurred before death sometimes reached $105^\circ F$ (40.6°C).
### TABLE 3. THE PERIPHERAL BLOOD PARASITAEMIA DURING A SINGLE 48-HOUR CYCLE OF P. FALCIPARUM IN AN OWL MONKEY

<table>
<thead>
<tr>
<th>Parasitaemia (parasites/10⁴ r.b.c.)</th>
<th>Hour 0</th>
<th>Hour 24</th>
<th>Hour 36</th>
<th>Hour 48</th>
</tr>
</thead>
<tbody>
<tr>
<td>Temperature</td>
<td>102.2°F</td>
<td>100.2°F</td>
<td>99.4°F</td>
<td>102.0°F</td>
</tr>
<tr>
<td></td>
<td>39.0°C</td>
<td>37.9°C</td>
<td>37.4°C</td>
<td>38.9°C</td>
</tr>
<tr>
<td>Rings</td>
<td>1 240</td>
<td>375</td>
<td>505</td>
<td>5 120</td>
</tr>
<tr>
<td>Trophozoites</td>
<td>180</td>
<td>735</td>
<td>200</td>
<td>65</td>
</tr>
<tr>
<td>Schizonts</td>
<td>0</td>
<td>0</td>
<td>125</td>
<td>260</td>
</tr>
<tr>
<td>Total all forms of parasite</td>
<td>1 420</td>
<td>1 110</td>
<td>830</td>
<td>5 445</td>
</tr>
</tbody>
</table>

The relationship of the temperature changes to the parasite cycle as determined by blood films is shown in Table 3. The clearly defined febrile episodes of *P. falciparum* infected owl monkeys are usually even more synchronous than those in infected humans (James et al., 1932) and are of much greater magnitude than those observed in induced *P. falciparum* infections in chimpanzees (Hickman et al., 1966) or in gibbons (Ward & Cadigan, 1966). Surprisingly, *P. coatneyi* infections in rhesus monkeys, which in some ways parallel *P. falciparum* in man, are not characterized by clearly defined temperature elevations (Desowitz et al., 1967). Because of this the owl monkey is probably a more suitable animal for some clinical and chemotherapeutic studies on malaria.

Few other clinical symptoms appeared in the infected monkeys apart from progressive weakness approaching death. Most infected animals developed diarrhoea in the terminal stages and the presence of occult blood in the faeces probably reflects haemorrhages of the gastro-intestinal tract resulting from the thrombocytopenia.

The very severe anaemia may well be the main cause of death in *P. falciparum* infected owl monkeys since the packed cell volume drops to about 10% and terminal erythrocyte counts of one to two million/mm³ have been observed (Table 4).

The upper limit of parasitaemia consistent with the life of the owl monkeys is also shown in Table 4. All stages of the parasite were present in the peripheral blood at the time of death with either the young rings or later developmental stages predominating, showing that the animals died at any point in the cycle.

One infected monkey showed symptoms not explicable by anaemia. This animal had severe rigors, diarrhoea and hyperpyrexia when only 1% of the red cells were infected. The rigors ceased after four hours but the animal appeared to be in a state of collapse. Following intramuscular quinine therapy rapid recovery occurred although partial paralysis persisted for a further day. Although this animal suffered a recrudescence two weeks later and parasitaemia rose to a level 10 times higher than that of the initial attack, the only clinical symptoms were mild febrile episodes. It is possible that in this monkey cerebral involvement occurred during the initial attack, a serious manifestation of the disease sometimes seen in non-immune human patients.
## TABLE 4. TERMINAL PARASITOLOGICAL OBSERVATIONS ON OWL MONKEYS DYING OF \textit{P. falciparum} INFECTIONS

<table>
<thead>
<tr>
<th>Stage of parasite</th>
<th>Owl monkeys parasitaemia (parasites/10^4 r.b.c.)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Monkey</td>
</tr>
<tr>
<td></td>
<td>1</td>
</tr>
<tr>
<td>Ring</td>
<td>700</td>
</tr>
<tr>
<td>Trophozoite</td>
<td>5000</td>
</tr>
<tr>
<td>Schizont</td>
<td>1150</td>
</tr>
<tr>
<td>Total</td>
<td>6850</td>
</tr>
<tr>
<td>r.b.c/mm$^3 \times 10^6$</td>
<td>1.2</td>
</tr>
<tr>
<td>P.C.V. %</td>
<td>7.5</td>
</tr>
</tbody>
</table>

* ND = Not done.

**Chemotherapy.** Complete cure of infections with this strain of \textit{P. falciparum} was achieved by a single oral dose of an aqueous suspension of sulfadoxine (sulfadoxine or Fansil) 20 mg base/kg + pyrimethamine (Daraprim) 2 mg base/kg. Treatment of a single infected animal with five doses of quinine 30 mg base/kg intramuscularly, the first given in the afternoon, then morning and evening for two days, depressed parasitaemia for 10 days after which a recrudescence occurred (Fig. 4). Treatment of the recrudescence with a similar quinine regime resulted in radical cure of the infection. This may well have been an example of acquired immunity potentiating a chemotherapeutic agent. One animal with high parasitaemia received chloroquine intramuscularly at 10 mg base/kg. The animal died 24 hours later but blood films made during that day did not reveal the characteristic effect of chloroquine on the morphology of sensitive parasites after exposure to the drug. Chloroquine resistance would not be unexpected because this strain was resistant when isolated from man (DeGowin & Powell, 1965).

**Pathology.** There was considerable variation in the proportion of parasitized red cells in the vessels in different organs. In all the owl monkeys examined the density of parasites was greatest in the heart and lowest in the brain. In the small interstitial capillaries around the muscle fibres of the myocardium, and in the smaller venules (Fig. 5), the parasitization of red blood cells in several animals was almost 100%; in the same animals in the small vessels of the meninges and cerebrum it was only 10–20%. In the lungs and kidneys the degree of parasitization was only slightly higher than in the peripheral blood (approximately 40%). The bone marrow was heavily parasitized in the one animal in which this tissue was examined. In the liver and spleen the number of parasites was difficult to estimate because of the very heavy deposits of malaria pigment.

Occasionally in human infections massive parasitization of red cells is seen in the heart capillaries (Dugeon & Clarke, 1917) but in general there are less parasites here than in other organs (Maegraith, 1948).
TABLE 5. RESULTS OF BLOOD COAGULATION STUDIES CARRIED OUT ON AN OWL MONKEY INFECTED WITH P. FALCIPARUM

<table>
<thead>
<tr>
<th></th>
<th>Before infection</th>
<th>Days before death</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>9-10</td>
</tr>
<tr>
<td>Parasitaemia parasites/10⁴ r.b.c.</td>
<td>0</td>
<td>&lt;1</td>
</tr>
<tr>
<td>Cephalin time</td>
<td>80 secs</td>
<td>*</td>
</tr>
<tr>
<td>Factor V%</td>
<td>240</td>
<td>450</td>
</tr>
<tr>
<td>Factor VIII%</td>
<td>300</td>
<td>200</td>
</tr>
<tr>
<td>Fibrinogen mg/100 ml</td>
<td>390</td>
<td>393</td>
</tr>
<tr>
<td>Platelets x 10⁵/mm³ blood</td>
<td>344</td>
<td>278</td>
</tr>
<tr>
<td>Euglobulin lysis time in hours</td>
<td>&gt;4</td>
<td>&gt;4</td>
</tr>
</tbody>
</table>

* Not done.

A similar predilection of a malaria parasite for the heart as a site of schizontony has been reported for P. coatneyi by Garnham (1965) and for P. knowlesi in rhesus monkeys (Menon, 1939). The mechanism by which differential parasitization of red cells occurs in certain capillaries is not yet understood. It is known, however, that red cells infected with mature parasites or schizonts tend to agglutinate, and this may be one reason for the restriction of schizontony to the internal organs in human falciparum malaria (Garnham, 1966) and the distribution which we have observed in owl monkeys (Table 3).

Blood coagulation studies. A significant reduction in platelet count was found in all of the animals examined. This was closely related to the development of parasitaemia (Figs. 3 and 6). In the animal in which coagulation factors were studied, the cephalin time was prolonged and Factor VIII was grossly reduced at the terminal phase of the infection. There was no change in fibrinogen or Factor V levels and the euglobulin lysis time remained at longer than four hours throughout the course of infection (Table 5).

The progressive thrombocytopenia could be explained by diffuse intravascular coagulation, marrow failure or hypersplenism. When considered in conjunction with the prolonged cephalin time and reduction in Factor VIII, diffuse intravascular coagulation is the most probable explanation. Factor VIII was depleted in all the human patients studied by Dennis et al. (1967) and although it might be expected that Factor V and fibrinogen would also be reduced, normal values were found in three out of 15 and 22 out of 31 patients respectively by these workers. In Aotus monkeys the fibrinogen may be protected by the unusually high levels of plasma antithrombin present in this species (Hawkey, unpublished observation, 1969).

The presence of occult blood in the faeces of Aotus monkeys in the terminal stages of P. falciparum and the cerebral symptoms which developed in one infected animal, strengthen the premise that diffuse intravascular coagulation was present as a result of the infection. This condition has been described in human patients with malarial infections and is thought to result from red cell breakdown in the circulating blood with release of a phosphoidal substance which activates the coagulation mechanism (Quick, 1957) and adenosine diphosphate which causes platelets to aggregate (Gaarder et al., 1961). In its intermediary stages this situation can give rise to the formation of platelet and fibrin microthrombi. Although these are usually cleared rapidly by the reticuloendothelial and fibrinolytic systems, and are not detectable clinically or histologically, their production involves depletion of
circulating levels of platelets and clotting factors giving rise to a haemorrhagic condition. This may be the cause of the haemorrhage and thrombosis found in many organs of patients dying with falciparum malarial infections. Renal, hepatic and splenic thrombosis and cerebral thrombosis complicated by haemorrhage has been described in these patients by Dudgeon & Clark (1917) and Spitz (1946) and haemorrhagic symptoms in patients with untreated P. falciparum have been noted by Cadigan et al. (1967).

Devakul, Harinasuta & Reid (1966) have demonstrated an increased turnover of 131I labelled fibrinogen in patients with acute, drug resistant P. falciparum infection, and Cadigan et al. (1967) and Dennis et al. (1967) have found thrombocytopenia, prolongation of the cephalin time and prothrombin time and reduction of coagulation factors during the acute phase of the infection. In some patients the abnormalities were partially corrected by treatment with heparin (Dennis et al., 1967). These findings strongly suggest that in malaria platelets and clotting factors become depleted as a result of diffuse intravascular coagulation.

Our results suggest that a similar condition occurs in owl monkeys infected with P. falciparum, which closely parallels the situation in humans infected with this same parasite, although so far there is no histological evidence that the condition progresses to the stage of occlusive thrombosis.

The similarity of the pattern of the disease in man and Aotus suggest that this animal may be useful for the clinical study of P. falciparum for which none of the other mammalian malaria model systems are satisfactory.

**RESUME**

Les auteurs exposent la méthode qu'ils ont utilisée pour provoquer une infection à P. falciparum chez le nyctipitheque (Aotus trivirgatus) et décrivent l'évolution de l'infection. Ils ont procédé par inoculation intraveineuse de sang infecté par la souche Camp de P. falciparum, originaire de la Malaisie occidentale, après passage de cette souche sur des chimpanzés et sur Aotus. Les infections ont été provoquées soit par transfert direct de sang infecté d'un nyctipitheque à un autre, soit en utilisant ce même sang conservé à -70°C et additionné de glycérine comme conservateur. Les tentatives faites pour transférer directement l'infection de l'homme à des Aotus splénectomisés ont échoué à cause, croit-on, d'une perte de viabilité de l'inoculum au cours du transport depuis les régions de paludisme endémique.

On a généralement observé de petites formes annulaires à la périphérie des hématies au début du cycle évolutif du parasite. Vers le stade terminal de l'infection, les schizontes mûrs devenaient facilement décelables dans le sang périphérique avec une moyenne de 21,6 merozoïtes par schizonte (minimum 10, maximum 37). Les gamétocytes étaient extrêmement peu nombreux, peut-être à cause des passages répétés de la souche dans le sang.

Chez tous les nyctipithèques étudiés, l'inoculation de cette souche modifiée de P. falciparum a provoqué des infections foudroyantes et la mort toutes les fois que l'évolution n'a pas été modifiée par la chimiothérapie.

Le seuil fébrile observé correspondait au niveau enregistré chez l'homme. La première élévation de température nettement établie était toutefois inférieure à 37,8°C: les poussées ultérieures atteignaient parfois 40,6°C, avec une amplitude beaucoup plus grande que dans le cas des infections à falciparum provoquées chez le chimpanzé. On n'a guère observé d'autres symptômes cliniques qu'un affaiblissement progressif. Aux stades terminaux, il s'est produit de la diarrhée avec présence occulte de sang dans les fèces, provenant sans doute d'hémorragies du tube digestif provoquées elles-mêmes par une thrombocytopenie. La mort était probablement causée par une anémie aiguë, le volume globulaire tombant à 10 % de sa valeur normale tandis que la numération finale des globules rouges n'était plus que de un million/mm³. A part les
gamètocytes, on a observé dans le sang, au moment de la mort, des formes de parasites correspondant à toutes les phases du cycle évolutif.

La guérison complète a été obtenue par l'administration orale d'une dose unique d'une association de sulfadoxine (20 mg-base/kg) et de pyriméthamine (2 mg-base/kg) en suspension aqueuse. Le seul animal traité à la quinine a reçu au total 150 mg de la base/kg en trois jours. Il y a eu diminution de la parasitémie pendant 10 jours, puis une recrudescence. Un autre singe a été traité par injection intramusculaire de chloroquine (10 mg de la base/kg), mais apparemment sans effet puisque l'animal a succombé dans les vingt-quatre heures. Il est vrai qu'on avait utilisé une souche résistant à la chloroquine chez l'homme.

L'autopsie a révélé que la densité des parasites était maximale dans les capillaires et les petites veines du myocarde où, dans certains cas, 100 % des hématies étaient parasitées, alors que, chez les mêmes animaux, 10 à 20 % seulement d'entre elles étaient parasitées dans les petits vaisseaux des méninges et du cerveau.

Des études ont été faites sur la coagulation du sang. On a noté chez tous les sujets une diminution sensible du nombre des plaquettes, étroitement liée au développement de la parasitémie, avec prolongation du temps de coagulation par la céphaline et une forte réduction du facteur VIII à la phase terminale de l'infection. Aucun changement n'a été observé en ce qui concerne les taux du fibrinogène ou du facteur V ni dans le temps de lyse de l'euglobuline.

La similitude du tableau de la maladie chez l'homme et chez Actus fait penser que ce singe pourrait être utilisé avec profit pour certaines études cliniques et chimiothérapeutiques sur le paludisme à falciparum.

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FIG. 1
THE MORPHOLOGY OF P. FALCIPARUM FROM THE PERIPHERAL BLOOD OF AN OWL MONKEY

A Early phase of infection

B Terminal phase of infection
FIG. 2

THE FREQUENCY OF DIFFERENT MEROZOITE NUMBERS PER SCHIZONT IN A
P. FALCIPARUM INFECTION IN AN OWL MONKEY
FIG. 3

PARASITAEMIA, PLATELET COUNTS AND TEMPERATURE MEASUREMENTS OF 3 OWL MONKEYS INFECTED WITH P. FALCIPARUM
FIG. 4

THE COURSE OF INFECTION OF P. FALCIPARUM IN AN OWL MONKEY FOLLOWING TREATMENT OF THE PRIMARY ATTACK

LOG. PARASITES \(10^4\) R.B.C.

QUININE

QUININE

QUININE

DEGREES FAHRENHEIT

DEGREES CENTIGRADE

DAYS AFTER INFECTION

35 36 37 38 39 40

95 96 97 98 99 100 101 102 103 104

2 4 6 8 10 12 14 16 18 20 22 24 26 28 30 32 34
FIG. 5
A SECTION OF THE HEART OF AN OWL MONKEY INFECTED WITH P. FALCIPARUM

WHO 90636
FIG. 6

THE RELATIONSHIP OF PLATELET COUNTS TO PARASITAEMIA IN OWL MONKEYS INFECTED WITH P. FALCIPARUM
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