Laboratory Animal Care

Vol. 20, No. 2
Part II of Two Parts
Printed in U.S.A.

NATURAL AND INDUCED MALARIAS IN WESTERN HEMISPHERE MONKEYS[1,2]

MARTIN D. YOUNG

SUMMARY • *Plasmodium brasilianum and P. simium are found naturally in western hemisphere monkeys. The infected hosts belong to 9 genera from 5 countries. These parasites have been experimentally induced into 6 species of monkeys which are not natural hosts. Seven species of monkeys have been experimentally infected with one or more of the following species of human malaria, viz., P. vivax, P. falciparum, and P. malariae. The experimental infections differ among the monkey hosts and between these hosts and the human hosts in various respects. The vectors of naturally occurring monkey malarias are not known. Several mosquito species can be infected by human malarias in monkey hosts and can transmit the infections to other monkeys and to man. These new host-parasite-vector relationships present many problems for investigation. Human malaria induced in monkey hosts is a promising model for the study of the malaria of man. Some of the needs to enable the more adequate use of this model were discussed.*

In 1908, more than 60 years ago, *Plasmodium brasilianum*, the first malaria of western hemisphere monkeys was described (17). In 1951 the second and last malaria, *P. simium*, was recorded (7).

Investigations on these monkey malarias proceeded at a slow pace for a long time. In the early 1930's, there was a surge in research activities when Clark started epidemiological work on these parasites at the Gorgas Memorial Laboratory in Panama. This was followed at the same place by the outstanding work of the Taliaferros on biological characteristics and immunity. They also transmitted human malaria for the first time to monkeys, although the model they chose now appears not to have been the best one; the human parasites grew only poorly in the monkey species tried.

Starting about 10 years ago, there was a renewed interest in simian malarias in the New World, mainly by Deane and his associates in Brazil and a few others in other South American countries. During the 1960's, there was increasing appreciation of the New World monkeys as models for various biological studies.

In 1965, it was found at the Gorgas Memorial Laboratory that human *P. vivax* malaria would grow readily in the *Aotus* monkey (30). This led to a spurt of interest and investigations, resulting in a large amount of information being produced rapidly.

It appears propitious now to bring together some of the information on the natural and induced malarias of western hemisphere monkeys with an assessment of the present situation. In compiling the information from the past records, review and summary articles were used where it seemed appropriate. Consultation of the references listed in this report will indicate where a large number of additional citations can be found.

The taxonomy and nomenclature of primates are in a fluid state, according to Napier and Napier (19). To one just beginning to work with primates, this fluid condition is very impressive. One may be confronted with several generic names for what may be the same animal or slightly different

[1] From the Gorgas Memorial Laboratory, Apartado 6691, Panama 5, R. P.
[2] Presented at the Nonhuman Primate Parasite Workshop, Southwest Foundation for Research and Education, San Antonio, Texas, May 26–27, 1969.

variants. For simplicity in this report, the classification given by Napier and Napier (19) is followed and, hopefully, the animals found infected with malaria have been assigned at least to a recognized and acceptable genus. With future clarification of the systematics of the monkeys and with further experimental work with malaria, the specificities of the host-parasite relationships may become more obvious.

Simian Malarias

Only 2 species of malaria have been found naturally in western hemisphere monkeys.

Plasmodium brasilianum has been found in specimens of the following 9 genera, all in the family Cebidae; viz., *Cacajao* (type host), *Chiropotes, Cebus, Ateles, Callicebus, Lagothrix, Alouatta, Brachyteles,* and *Saimiri.* Seventeen or more species belonging to the above genera are hosts, the number depending upon the classification followed. This parasite has been reported in monkeys from Panama, Colombia, Venezuela, Peru, and Brazil (8–11, 13, 18, 20, 25).

Seidelin in Yucatán saw repeatedly in the blood of a captive monkey (*Ateles* sp) ring forms of a red blood cell parasite which he said probably belonged to the genus *Plasmodium* (24). Some reviewers have recorded this as being *P. brasilianum* from *Ateles geoffroyi.*

P. brasilianum infection has been induced experimentally in *Aotus trivirgatus* and *Saguinus geoffroyi,* species not naturally infected (27).

This malaria has not been reported from members of the family Callitrichidae, although probably more than 1000 specimens of the various genera have been examined.

P. simium has been found naturally in *Alouatta fusca* (type host) (7) and *Brachyteles arachnoides* (11). This malaria has been reported only from Brazilian monkeys. It has been induced experimentally in *Saimiri sciureus, Ateles paniscus, Lagothrix lagotricha,* and *Callithrix jacchus* (8).

Human Malarias Induced in Monkeys

The first infection of monkeys with human malaria was *Plasmodium falciparum* transmitted to *Alouatta villosa* (=*A. palliata*) by the injection of parasitized blood (26). The next report of a successful transfer of human parasites to monkeys was 32 years later when Young, Porter, and Johnson (30) demonstrated that *Aotus trivirgatus* could be readily infected with *P. vivax.* This finding was followed by a great deal of testing of the susceptibility of different species of monkeys to various human plasmodia. As a result, so far 7 species of New World monkeys have been shown to be susceptible to 1 or more of the human species. Some general characteristics of human malarias induced in these monkeys follow:

Alouatta villosa (howler monkey). In the few attempts made, we have failed to infect *Alouatta* with Panama vivax from either human or monkey donors (29). This was the first monkey shown to be susceptible to human malaria when the Taliaferros (26) infected 9 specimens with *P. falciparum* from Panama patients. However, the infections persisted for only a short time, the longest being 8 days. Blood subinoculated into another, a single, *Alouatta* produced a transient infection of less than 16 hours. This monkey appears to be one of the least susceptible to human malarias of the Panamanian primates tried.

Aotus trivirgatus (night or owl monkey). This monkey is very susceptible to *P. vivax* malaria (30). Malarious blood readily infects *Aotus,* especially when splenectomized and/or given an immunosuppressant drug. Once established in the *Aotus,* the malaria can then be passed easily to other monkeys, even to those not splenectomized or given drug. Strains have been maintained for 3 years by serial passages.

High parasitemias are produced, much higher on the average than in human syphilitic cases with induced malarias. The infections can be fatal to the monkeys. The parasitemias persist on the average for about 1

month, but some may persist for 3 months or more. Relapses occur. Gametocytes occur in most of the infections.

P. falciparum of Panamanian origin is very difficult to establish in the local *Aotus*. However, Geiman and Meagher (15) readily infected *Aotus* with *P. falciparum* of Ugandian origin. Since then, strains from Malaya and Cambodia have been established in this host [(4); unpublished data at Gorgas Memorial Laboratory; Schmidt, L. H. personal communication]. The infections produce high parasitemias and can be fatal to the monkeys. Gametocytes are produced.

Both the vivax and falciparum infections in monkeys respond readily to drugs such as chloroquine, quinine and thio-bismol.

P. malariae has been established in *Aotus* (16). The parasitemias were persistent. Gametocytes were produced.

Saguinus geoffroyi (titi marmoset). The first report of the susceptibility of *Saguinus* to *P. vivax* of human origin indicated that the induced infections were of a low order (21). Subsequent experience with various strains show that some strains will grow well in this monkey, especially if the inoculation is obtained from a monkey host, usually *Aotus* (23). The parasitemias will reach high levels and may persist for as long as 78 days, although the usual patency is about a month or more. The heaviest infections were obtained in animals which had been splenectomized and/or given an immunosuppressant drug. The monkeys appeared to tolerate the infections well.

S. geoffroyi is also susceptible to *P. falciparum* (22). Four of 32 attempts to induce Panamanian strains resulted in infections. The monkeys were either splenectomized or given an immunosuppressant drug. One splenectomized animal showed a peak parasitemia of 4600 per cmm and the infection persisted for 15 days, the longest duration seen. The highest parasitemia was 22,600 per cmm, but the infection persisted for only 6 days. Two had low parasitemias of short duration. The monkeys tolerated the infections well. Gametocytes were produced.

Saimiri sciureus (squirrel monkey). Deane *et al* (12) reported the infection of a splenectomized *Saimiri* monkey with *P. vivax* blood from a Brazilian patient. A parasitemia with a peak of 28,080 per cmm was produced. The infection persisted for 171 days, at which time it was treated with chloroquine. Subinoculation into another splenectomized *Saimiri* produced a low grade infection lasting for 80 days.

A normal *Saimiri* monkey was susceptible to the Ugandian strain of *P. falciparum* (31). The malarious blood donor was an *Aotus* monkey. The induced infection was of moderate intensity, persisted for 3 weeks, and produced a few gametocytes.

Ateles fusciceps and *A. geoffroyi* (spider monkeys). These 2 species are so similar they will be discussed together. Although in our laboratory we have failed to infect these monkeys by direct inoculation from man, *P. vivax* infections in *Aotus* can be transferred to *Ateles* and from *Ateles* to *Ateles* (29). The infected animals had been splenectomized and/or given immunosuppressant drugs. Most attempts to subinoculate from monkey hosts were successful. High parasitemias were produced but were not fatal to the *Ateles*. The patent periods averaged about 1 month in length but did not persist so long as the vivax infections in the *Aotus*. Gametocytes were produced.

Cebus capucinus (white-faced capuchin). Of the 6 attempts to infect *Cebus* with *P. vivax* from Panamanian patients, none were successful (29). However, when vivax was established in the *Aotus*, 2 of 14 attempts to subinoculate from this host to *Cebus* were successful. The resultant parasitemias were very low, and the average persistence was 5 days.

P. falciparum from Panamanian patients

[a] After the completion of this manuscript, a report was published by Collins, W. E., Contacos, P. G., and Guinn, E. G. Observations on the sporogonic cycle and transmission of *Plasmodium simium* da Fonseca. J. Parasitol. 55: 814–816, 1969. The report contained the following information: "The sporogonous cycle of *P. simium* was completed in *Anopheles freeborni*, *A. maculatus*, and *A. stephensi* which had fed upon an experimentally infected *Saimiri*. Transmission to other *Saimiri* monkeys was accomplished by the first 2 mosquito species." These mosquitoes do not occur in Brazil where natural infections of *P. simium* are found.

also failed to infect 2 *Cebus* (28). But the Uganda strain obtained from *Aotus* donors infected most of the *Cebus* tried. The parasitemias were high, one showing 662,720 parasites per cmm. The monkeys tolerated the infections well. The patent periods were of several weeks duration, one existing for 10 weeks. Gametocytes were produced.

Natural and Experimental Mosquito Vectors of Monkey Malaria[3]

The natural vectors of *P. brasilianum* and *P. simium* are not known. On epidemiological evidence, Deane *et al* (10) suspected that *Anopheles (Kerteszia) cruzi* is a vector of *P. simium malaria*, although they failed to transmit the infection to 3 *Saimiri sciureus* monkeys by injecting sporozoites from naturally infected mosquitoes.

Clark and Dunn (6) apparently were the first to experimentally infect mosquitoes by feeding them upon *Ateles geoffroyi* with a natural infection of *P. brasilianum*. *A. albimanus* and *A. tarsimaculatus* (= *aquasalis*) developed sporozoites, but attempts to infect man by the bites of these mosquitoes failed.

Exotic species of mosquitoes have been infected by *P. brasilianum*. *A. aztecus* and *A. atroparvus* developed sporozoites in the glands after feeding upon a *Saimiri sciureus* with this parasite (14). Sporozoites from *A. aztecus* produced exoerythrocytic bodies but not a blood infection when injected into a *Cebus capucinus*. *Anopheles freeborni* from California was infected by feeding upon an *Ateles geoffroyi* which had acquired its *P. brasilianum* infection naturally in Panama (5). These mosquitoes transmitted the infection to 5 human volunteers.

Experimental Infection of Mosquitoes by Human Malarias Induced in Monkeys

A. albimanus was the first mosquito shown to be capable of transmitting human malaria from a monkey host to another host (30). These mosquitoes were infected by *P. vivax* in an *Aotus trivirgatus*. When these mosquitoes subsequently fed upon 2 human volunteers and an *Aotus* monkey, the humans developed *vivax* infections but the *Aotus* did not.

Further studies showed that most Panama *P. vivax* strains induced in *Aotus*, *Saguinus*, and *Ateles fusciceps* would infect *A. albimanus* (2). *A. albimanus* infected by malarious *Aotus* transmitted the infection to *Aotus* and to *Saguinus*. These mosquitoes infected by *P. vivax* in *Ateles fusciceps* transmitted the infection to other *A. fusciceps* (1). *A. albimanus* generally acquired higher rates of infections from humans with *P. vivax* malaria than from monkeys with *P. vivax*. With *P. vivax* in man the percentage of infections ran up to 91.7% and with *P. falciparum* up to 90.0%.

Other Panamanian mosquitoes which failed to become infected with *P. vivax* in monkeys when companion *A. albimanus* were infected are: *A. oswaldoi*, *A. apicimacula*, and *A. punctimacula*.

Anopheles freeborni. Feeding this exotic mosquito species upon *Aotus* monkeys with Malayan IV strains of *P. falciparum*, Contacos and Collins (4) obtained infections which were transmitted to human volunteers. They failed to infect this mosquito with another *P. falciparum* strain from Malaya (Camp) or with a strain from Uganda. Later another foreign strain of *P. falciparum* from Cambodia, induced in *Aotus*, also infected this mosquito (3).

A. pseudopunctipennis of Panamanian origin when fed simultaneously with *A. albimanus* upon monkeys with induced *P. vivax* malaria became infected but showed lower rates of infection than *A. albimanus* (1). The ratio of infected *albimanus* to *pseudopunctipennis* was about 20:1. The same ratio held when the 2 species were fed together on humans with *P. falciparum*.

Discussion

Of the 16 genera of monkeys in the western hemisphere, only 9 have been shown to harbor malaria parasites. Infected mon-

keys have been reported from only 5 of the 19 countries south of the United States. It seems likely that malaria infections are more widespread in the area and are present in more hosts than the reports now indicate. As more interest develops in the New World monkeys, undoubtedly the number of hosts and geographical ranges will be extended. This prediction is supported by the experimental evidence showing that some species of monkeys not so far found infected in nature can be experimentally infected with simian malarias.

Nearly all of the monkey species found in Panama have been shown to be susceptible to some of the human malarias, especially to *P. vivax* and *P. falciparum*. In addition, the *Aotus* is susceptible to *P. malariae*. Further studies may show additional species of monkeys are susceptible to human malarias.

The donor of the malaria appears to be important. *P. vivax* from man readily infects some of the monkeys but not others. However, once this malaria is established in the monkey, other simian species are more easily infected.

With *P. falciparum* another facet occurs. *P. falciparum* from Panamanian patients is very difficult to establish in Panamanian monkeys. Of more than 100 trials, only a few have been successful, and then only temporarily. The low grade infections produced could not be transferred to other monkeys. But with *P. falciparum* from Africa and Southeast Asia, far removed from the locale of the Panamanian monkey, the infections can be easily transferred from man to some monkeys, produce very high parasitemias which may be fatal, and are readily transferred by serial passages.

The differences in the above host-parasite combinations pose a fascinating problem waiting to be solved, and much important basic information could emerge from the solving process.

P. falciparum induced in monkeys tends to run a parasitological course somewhat similar to that in man, although in some cases the maximum parasitemias in the monkeys can be extraordinarily high.

The parasitemias of *P. vivax* in the most susceptible monkeys can be very high, much higher than those normally seen in human patients. Why the *vivax* parasitemias are much higher in monkeys would be a challenge for investigation.

One gets the impression that gametocytes are produced less regularly in the monkeys than with the same malaria species in man. Also, these gametocytes appear to be less infective to mosquitoes indigenous with the monkey than the same infections in man.

An unusual aspect is the difference in infectivity of malaria to indigenous and exotic mosquitoes. At present, the latter appear to be better vectors for the human malarias induced in monkeys. However, even among the exotic mosquitoes, the situation can vary greatly. While some strains of malaria from Africa and Malaya do not infect mosquitoes well, Contacos and Collins (4) found that another strain from Malaya and one from Cambodia produced heavy infections in *A. freeborni* mosquitoes. Here is an odd example of a malaria from the other side of the world growing in a monkey from South America infecting well mosquitoes originating from California. It seems unlikely that this threesome had ever been associated before in nature, but when all parties were put together, a good host-parasite-vector combination resulted. In contrast, Panamanian vivax malaria growing in Panamanian monkeys only poorly infected the Panamanian mosquitoes tried.

One of the major unsolved problems is to find compatible host-parasite-vector combinations.

The vectors of the monkey malarias in nature are unknown. Various species of anopheline mosquitoes from the area in which the monkeys live should be investigated for their vector ability and importance. This should be done for the experimentally induced malarias in the monkeys, as well as for the naturally occurring infections. Findings in either situation may have significance for the other.

One of the potentially important uses of monkeys is as models for the study of human diseases, especially infectious diseases. It is hoped that they may not only substitute for man in many areas, but may even extend experimental work that will be of significance to man, as well as to the monkey. To do this, the testing of the ability of various species to be hosts for disease agents is one of the first goals.

As a corollary to this, there is a great need for increased knowledge of the biology of monkeys. There is also an increasing need for the development of better husbandry practices. We should look forward to establishing adequate breeding colonies of monkeys for several reasons. One is to provide the scientist with animals whose histories are better known and which are freer from the natural infections acquired in nature. Also important is the need for the conservation of populations of monkeys in their natural environment, hopefully by offsetting this drain on the wild populations by breeding in captivity.

These are only a few of the many problems which have become so apparent recently due to the emergence of the monkey as a host for an increasing number of human diseases, both actual and implied. One of the difficulties now facing the investigator is that of choosing a problem from the many fascinating ones and finding the support to pursue it.

REFERENCES

1. Baerg, D. C., Porter, J. A., and Young, M. D. Sporozoite transmission of *Plasmodium vivax* to Panamanian primates. Am. J. Trop. Med. Hyg. **18:** 346–350, 1969.
2. Baerg, D. C. and Young, M. D. Susceptibility of mosquitoes to human malaria induced in Panamanian monkeys. Mil. Med. **134:** 772–779, 1969.
3. Collins, W. E., Contacos, P. G., Guinn, E. G., Jeter, M. H., and Sodeman, T. M. Monkey to man transmission of *Plasmodium falciparum* by *Anopheles freeborni* mosquitoes. J. Parasitol. **54:** 1166–1170, 1968.
4. Contacos, P. G. and Collins, W. D. Flaciparum malaria transmissible from monkey by mosquito bite. Science **161:** 56, 1968.
5. Contacos, P. G., Lunn, J. S., Coatney, G. R., Kilpatrick, J. W., and Jones, F. Quartan-type malaria parasites of new world monkeys transmissible to man. Science **142:** 676, 1963.
6. Clark, H. C. and Dunn, L. H. Experimental efforts to transfer monkey malaria to man. Am. J. Trop. Med. **11:** 1–7, 1931.
7. Da Fonseca, F. Plasmodio de primato do Brazil Mem. Inst. Osw. Cruz **49:** 543–551, 1951.
8. Deane, L. M. Monkey malaria in Brazil. A summary of studies performed in 1964–66. Rev. Brazil. Biol. **27:** 213–228, 1967.
9. Deane, L. M. and de Almeida, F. B. Natural infections of howler monkey, *Alouatta seniculus straminea*, with *Plasmodium brasilianum* in the state of Amazonas, Brazil. Rev. Inst. Med. Trop. Sao Paulo **9:** 359–360, 1967.
10. Deane, L. M., Deane, M. P., and Ferreira Neto, J. Studies on transmission of simian malaria and on a natural infection of man with *Plasmodium simium* in Brazil. Bull. World Health Org. **35:** 805–808, 1966.
11. Deane, L. M., Ferreira Neto, J., and Goncalves Sitonio, J. Novo hospedeiro natural do *plasmodium simium* e do *plasmodium brasilianum*: O mono, *Brachyteles arachnoides*. Rev. Inst. Med. Trop. Sao Paulo **10:** 287–288, 1968.
12. Deane, L. M., Ferreira Neto, J., and Silveira, I. P. S. Experimental infections of a splenectomized squirrel-monkey, *Saimiri sciureus*, with *Plasmodium vivax*. Trans. Roy. Soc. Trop. Med. Hyg. **60:** 811, 1966.
13. Dunn, F. L. and Lambrecht, F. L. The hosts of *Plasmodium brasilianum* Gonder and von Berenberg Gossler, 1908. J. Parasitol. **49:** 316–319, 1963.
14. Garnham, P. C. C., Baker, J. R., and Nesbitt, P. E. Transmission of *Plasmodium brasilianum* by sporozoites and the discovery of an exoerythrocytic cycle. Parasitologia **5:** 5–9, 1963.
15. Geiman, Q. M. and Meagher, M. J. Susceptibility of a New World monkey to *Plasmodium falciparum* from man. Nature **215:** 437–439, 1967.
16. Geiman, Q. M. and Siddiqui, W. A. Susceptibility of a New World monkey to *Plasmodium malariae* from man. Am. J. Trop. Med. Hyg. **18:** 351–354, 1969.
17. Gonder, R. and Berenberg Gossler, R. von. Untersuchugen uber Malariaplasmodien Affen, Malaria Intern. Arch. Leipzig **1:** 47–56, 1908.
18. Marinkelle, C. J. and Grose, E. S. *Plasmodium brasilianum* in Colombian Monkeys. Trop. Geogr. Med. **20:** 276–280, 1968.
19. Napier, J. R. and Napier, P. H. *A Handbook of Living Primates*, vii & 456 p. Academic Press, London and New York, 1967.
20. Porter, J. A., Johnson, C. M., and de Sousa, L. Prevalence of malaria in Panamanian Primates. J. Parasitol. **52:** 669–670, 1966.
21. Porter, J. A. and Young, M. D. Susceptibility of Panamanian primates to *Plasmodium vivax*. Mil. Med. **131:** 952-958, 1966.
22. Porter, J. A. and Young, M. D. The transfer of *Plasmodium falciparum* from man to the marmoset, *Saguinus geoffroyi*. J. Parasitol. **53:** 845–846, 1967.
23. Porter, J. A. and Young, M. D. Infections of *Plasmodium vivax* in *Saguinus geoffroyi*. In manuscript.
24. Seidelin, H. Notes on some blood-parasites in man and mammals. Ann. Trop. Med. Parasitol. **5:** 501–508, 1911.
25. Serrando, J. A. Infección natural de un araguato, *Alouatta seniculus straminea* por *Plas-*

modium brasilianum. Acta Cient. Venezol. **18:** 12–15, 1967.
26. Taliaferro, W. H. and Taliaferro, L. G. The transmission of *Plasmodium falciparum* to the howler monkey, *Alouatta* sp. I. General nature of the infections and morphology of the parasites. Am. J. Hyg. **19:** 318–334, 1934.
27. Taliaferro, W. H. and Taliaferro, L. G. Morphology, periodicity and course of infection of *Plasmodium brasilianum* in Panamanian monkeys. Am. J. Hyg. **20:** 1–49, 1934.
28. Young, M. D. and Baerg, D. C. Experimental infections of *Plasmodium falciparum* in *Cebus capucinus* (white faced capuchin) monkeys. Mil. Med. **134:** 767–771, 1969.
29. Young, M. D. and Porter, J. A. Susceptibility of *Ateles fusciceps, Ateles geoffroyi* and *Cebus capucinus* to *Plasmodium vivax*. Trans. Roy. Soc. Trop. Med. Hyg. **63:** 203–205, 1969.
30. Young, M. D., Porter, J. A., and Johnson, C. M. *Plasmodium vivax* transmitted from man to monkey to man. Science **153:** 1006–1007, 1966.
31. Young, M. D. and Rossan, R. N. *Plasmodium falciparum* induced in the squirrel monkey, *Saimiri sciureus*. Trans. Roy. Soc. Trop. Med. Hyg. **63:** 686–687, 1969.