SUSCEPTIBILITY OF *ANOPHELES ALBIMANUS* TO PRIMATE AND AVIAN MALARIAS

DON E. EYLES

U. S. Department of Health, Education and Welfare, Public Health Service, National Institutes of Health, National Institute of Allergy and Infectious Diseases, Laboratory of Parasite Chemotherapy *

INTRODUCTION. In our experimental studies on monkey and avian malarias, it has been necessary to seek efficient vectors for the various species. Except for the report of Clark and Dunn (1) of a few mosquitoes experimentally infected with *Plasmodium brasilianum,* we could not obtain any information on the susceptibility of *Anopheles albimanus* to species of malaria other than those of man.

Since *A. albimanus* is readily maintained in an insectary and lives well under laboratory conditions, we have tried it on several species of malaria and have compared it with other vectors. This report describes the degree to which *A. albimanus* is susceptible to *Plasmodium cynomolgi* subspecies *bastianellii,* and *P. inui* in rhesus monkeys *(Macaca mulatta)* and to *P. gallinaceum* in the chicken. A note on *Anopheles freeborni* and *P. gallinaceum* is also included.

METHODS. *Anopheles albimanus* eggs were obtained from Dr. H. G. Simkover of the Shell Development Company, to whom we are indebted. The mosquitoes thrived in the insectary.

Plasmodium cynomolgi subspecies *bastianellii* and *P. inui* were maintained in rhesus monkeys. The feedings on *P. inui* were done on infected, splenectomized monkeys as splenectomized monkeys develop more intense parasitemias. Mosquitoes were fed when gametocytes were present in blood smears. The infections in *A. albimanus* were compared with infections in companion lots of *A. quadrimaculatus.* The lots of the two species were fed consecutively on the monkeys with as little delay as possible.

P. gallinaceum was maintained in various breeds of domestic chickens. The infections in *A. albimanus* were compared with infections in *Aedes aegypti.* One lot of *A. freeborni* was also fed on a chicken with *P. gallinaceum.*

Progress of the mosquito infections was followed by the examination of dissected midguts about a week after feeding. If oöcysts were found on the guts, the remainder of the lots (if any remained) were kept for transmission attempts. Transmission was attempted by the injection of dissected salivary glands, as most of the infections in *A. albimanus* were of a low grade, and transmission by bite would have been less certain.

OBSERVATIONS. *Plasmodium cynomolgi bastianelli:* Fourteen lots of *A. albimanus* were fed on monkeys infected with this species of malaria. For comparison, lots of *A. quadrimaculatus* were fed on the same monkeys. The results of the comparisons are summarized in Table 1.

It is evident that under the conditions of our experiments, *A. albimanus* is nearly insusceptible to *P. cynomolgi bastianellii.* Ten of the 14 lots showed no infection whereas the comparable lots of *A. quadrimaculatus* were lightly to moderately infected. The four lots which contained a few lightly infected *A. albimanus* were fed under the most auspicious circumstances. This is shown by the heavy infections obtained in the companion lots of *A. quadrimaculatus,* the mosquitoes of which bore an average of 65 to 469 oöcysts per midgut.

There were 22 mosquitoes of the *A. albimanus* Lot No. 275 remaining after midgut dissection. The glands of these were removed on the 14th day after feeding and injected intravenously into an

* From the Section on Cytology, located at the Medical School of the University of Tennessee, 874 Union Avenue, Memphis 3, Tennessee.

TABLE 1.—A comparison of the susceptibility of *Anopheles albimanus* and *A. quadrimaculatus* to infection with *Plasmodium cynomolgi bastianellii*

Anopheles albimanus				*Anopheles quadrimaculatus*			
Lot No.	No. dissected	Percent infected	Average oöcysts*	Lot No.	No. dissected	Percent infected	Average oöcysts*
247	17	0	0	246	20	55	12
252	18	0	0	251	20	45	16
255	21	0	0	254	20	55	7
257	8	0	0	256	10	70	34
260	17	0	0	261	20	85	26
275	24	8	0.2	274	10	90	469
278	22	0	0	277	10	90	24
280	20	0	0	281	10	50	4
296	20	5	0.1	295	10	90	358
298	20	0	0	297	11	63	8
301	20	5	0.2	300	10	90	129
302	20	5	4	303	10	70	65
306	20	0	0	307	10	60	11
310	19	0	0	311	12	30	3
All lots	266	2.1	—	All lots	183	65	—

* Average oöcysts per mosquito midgut.

uninfected monkey to determine if transmission would result. In addition, 31 mosquitoes of Lot No. 278 were also injected in spite of the fact that no oöcysts had been seen when the midguts were examined. The monkey which received these glands became infected, showing 5,925 parasites per mm.^{3} on the 16th day after inoculation. This is the first record of the transmission of a simian malaria by *A. albimanus*.

Plasmodium inui. Seven lots of *A. albimanus* were fed on splenectomized monkeys infected with *P. inui*. Companion lots of *A. quadrimaculatus* were fed for comparison as is shown in Table 2. Two of the seven lots showed very light infection of the *A. quadrimaculatus* mosquitoes but no infections among the corresponding *A. albimanus*. One infected *A. albimanus* mosquito was found in one lot, but no infected mosquitoes were found in the corresponding *A. quadrimaculatus* lot.

Even though it was demonstrated that *A. albimanus* can become infected with *P. inui*, it is obviously not superior to *A. quadrimaculatus*. Neither mosquito appears to be an efficient vector of this species;

TABLE 2.—Comparison of the susceptibility of *Anopheles albimanus* and *A. quadrimaculatus* to infection with *Plasmodium inui*

Anopheles albimanus				*Anopheles quadrimaculatus*			
Lot No.	No. dissected	Percent infected	Average oöcysts*	Lot No.	No. dissected	Percent infected	Average oöcysts*
283	20	0	0	284	22	9	0.1
287	20	0	0	284	20	5	0.1
290	20	0	0	289	18	0	0
291	20	5	0.1	292	20	0	0
308	20	0	0	309	20	0	0
315	20	0	0	314	10	0	0
322	20	0	0	323	20	0	0
All lots	140	0.7	–	All lots	130	2.3	–

* Average oöcysts per mosquito midgut.

however, low gametocyte counts may have contributed to the lack of success.

No transmission trials were attempted.

Plasmodium gallinaceum. It was previously known that several species of *Anopheles* could become infected with *P. gallinaceum.* Table 3 summarizes the infection, and were injected subcutaneously into an uninfected chicken. This chicken showed parasites eight days later. This is the first demonstration that *A. freeborni* may serve as an experimental transmitter of *P. gallinaceum;* however, oöcysts on the midgut have been noted previously (2).

TABLE 3.—Comparison of the susceptibility of *Anopheles albimanus* and *Aedes aegypti* to infection with *Plasmodium gallinaceum*

Anopheles albimanus				*Aedes aegypti*			
Lot No.	No. dissected	Percent infected	Average oöcysts*	Lot No.	No. dissected	Percent infected	Average oöcysts*
265	21	5	0.2	266	10	90	84
269	18	0	0	267	10	70	39
277	10	20	1.9	276	12	100	41
305	19	0	0	304	10	70	18
325	20	10	6.2	326	10	100	100
All lots	88	6.0	—	All lots	52	87	—

* Average oöcysts per mosquito midgut.

results of five feedings of *A. albimanus* on this species. For comparison, lots of *Aedes aegypti* were fed on the same infected chickens.

All of the *A. aegypti* lots became infected and the oöcyst counts ran on the average from 17 to 99 oöcysts per midgut. Three of the five *A. albimanus* lots also showed infection but the oöcyst counts were very low and only a small proportion of the mosquitoes bore cysts.

Oöcysts on the *albimanus* midguts were smaller and less advanced in development than those seen in *A. aegypti.* Transmission was not effected when nine pairs of dissected glands from *A. albimanus* Lot No. 277 were injected subcutaneously into an uninfected chicken.

One lot of *A. freeborni* was fed on *P. gallinaceum.* This feeding was coupled with those of the last comparison of Table 3. Sixty percent of the mosquitoes became infected with an average of 95 oöcysts per midgut. The infection was similar in intensity to that found in *Aedes aegypti* fed on the same chicken (lot no. 326).

Eleven pairs of glands were dissected from mosquitoes of this lot 14 days after

DISCUSSION. The main purpose of the experiments described was to make a selection of mosquitoes for experimental studies on malaria. These results are made available for those who may require such information.

A. albimanus was found nearly useless as a practical experimental vector of all of the species of malaria tried. Previous experimental work with this species has shown it to be an efficient transmitter of coindigenous *Plasmodium falciparum,* but a poor transmitter of strains of *P. falciparum* from other regions (3, 4). Other work with this species has indicated that it is not an efficient vector of other strains of human malarias except for coindigenous strains of *P. vivax* and *P. ovale* (5, 8).

The lack of success with *Plasmodium inui* parallels the lack of success of many investigators in finding mosquito hosts of this species. Garnham did succeed in obtaining good infections in *Anopheles atroparvus* (6) and previously we have found moderate infections in *A. freeborni* (7).

In the future we hope to use *A. albimanus* in attempts to transmit *Plasmodium*

brasilianum with which it is coindigenous, but so far we have been unable to obtain a strain of the latter.

SUMMARY. *Anopheles albimanus,* as compared with *A. quadrimaculatus,* was an inefficient experimental vector of *Plasmodium cynomolgi bastianellii,* but transmission was accomplished with this species.

Neither *A. albimanus* nor *A. quadrimaculatus* appeared to be efficient vectors of *Plasmodium inui.*

A. albimanus showed midgut infections with oöcysts of *P. gallinaceum* in a few instances, but there were fewer oöcysts than in *Aedes aegypti* and they appeared retarded in their development. One attempt at transmission failed.

A. freeborni became infected with *P. gallinaceum* and transmission was accomplished.

References

1. CLARK, H. C. and DUNN, L. H. 1931. Experimental efforts to transfer monkey malaria to man. Am. J. Trop. Med., 11(1):1–7.

2. EYLES, D. E. 1952. Studies on *Plasmodium gallinaceum.* IV. A comparison of the susceptibility of *Aedes aegypti, Anopheles quadrimaculatus* and *Anopheles freeborni.* Am. J. Hyg., 56(1): 71–77.

3. JEFFERY, G. M., EYLES, D. E., and YOUNG, M. D. 1950. The comparative susceptibility of *Anopheles quadrimaculatus* and two strains of *Anopheles albimanus* to a Panama strain of *Plasmodium falciparum.* J. Nat. Malaria Soc., 9(4): 349–355.

4. EYLES, D. E., and YOUNG, M. D. 1950. The comparative susceptibility of *Anopheles albimanus* and *Anopheles quadrimaculatus* to a South Carolina strain of *Plasmodium falciparum.* J. Inf. Dis., 87:189–193.

5. YOUNG, M. D. 1948. Adaptability of exotic malaria parasites to indigenous anophelines. Proc. Fourth Int. Congr. Trop. Med. and Malaria, 1: 672–680.

6. GARNHAM, P. C. C. 1951. The mosquito transmission of *Plasmodium inui* Halberstaedter and Prowazek, and its pre-erythrocytic development in the liver of the Rhesus monkey. Trans. Royal Soc. Trop. Med. and Hyg., 45(1):45–52.

7. EYLES, D. E. 1960. *Anopheles freeborni* and *A. quadrimaculatus* experimental vectors of *Plasmodium cynomolgi* and *P. inui.* J. Parasit., in press.

8. JEFFERY, G. M. 1954. The Donaldson strain of malaria. 3. The infection in the mosquito. Am. J. Trop. Med. and Hyg., 3:651–659.