

Longevity Studies on Malaria Infected *Anopheles*

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OBJECTIVE : To test the effects of human malaria parasites on the longevity of the host mosquito.

BACKGROUND : Most previous investigations on the effect of malaria parasites on the mosquito host have been inconclusive. De Buck and Swellengrebel (2) found greater mortality among heavily infected mosquitoes in relation to lightly infected ones. However, Boyd (1) and Ragab (3) concluded that malaria parasites have no detrimental effects on the longevity and vitality of the host mosquito. More recently, Schiefer *et al.* (4) found that reductions in the flight capabilities of infected *Anopheles stephensi* mosquitoes were positively correlated to the severity of *Plasmodium cynomolgi* infection. During the past several years AFRIMS investigators, collaborating on malaria drug testing in the rhesus monkey model, have noted a detrimental effect on mosquitoes infected with *P. cynomolgi*. Procedures during this study called for the production of maximum numbers of sporozoites in the laboratory vector, *An. balabacensis*.

Observations indicated that high oocyst numbers and subsequently high sporozoite numbers led to increased mortality in the host mosquitoes. The highest mortality was observed on day 2 and day 12 post-feeding, probably corresponding with the initial time of invasion of the midgut by ookinetes and later the rupturing of the oocysts on the midgut and the invasion of the salivary glands by sporozoites of *P. cynomolgi*. In order to determine whether increased mortality occurred in anopheline mosquitoes infected with human malaria, a study was designed to compare the longevity of uninfected mosquitoes with those infected with *P. vivax*. A reduction in vector longevity could be important information in the study of the epidemiology of human malaria.

METHODS : Sixty laboratory-reared *Anopheles balabacensis* (Thai Strain) and 60 *An. maculatus* (IMR Strain) were fed on human malaria patients in Phra Phutthabat or Kanchanaburi on the day of hospital admission and days 1, 7, 14, and 21 of follow-up. Ten engorged mosquitoes of each species were then withheld for longevity studies and the remainder were used for dissections and other experiments. Mosquitoes were also fed each day on uninfected volunteers as a simultaneous control. The mosquitoes had been reared and maintained either at the AFRIMS Phra Phutthabat insectary, or at the Bangkok insectary to be transported to the study site in Kanchanaburi. Females of both species were 4 to 5 days old on the day of feeding and were dissected 7 and 14 days post-feeding. Midguts and salivary glands were examined for oocysts and sporozoites respectively, with oocyst and sporozoite indices being determined at the time of dissection. Infected and control specimens were maintained as the original lot (10 specimens) in small screened paper cups supplemented daily with a multivitamin syrup

solution for nutrition. All cups were kept in the same room, so that both infected and control mosquitoes were exposed to the same temperature and humidity.

RESULTS : A total of 22 infected lots and 19 uninfected were compared for *balabacensis*, and 20 infected lots and 13 uninfected lots for *maculatus*. The life span for both infected and uninfected *balabacensis* females ranged from 1-51 days, while that for *maculatus* ranged from 2-34 days. Mean survival days for both infected and uninfected females of both species are shown in Table 1. For both *balabacensis* and *maculatus*, uninfected females lived longer than infected females indicating that malaria infections in mosquitoes may have a detrimental effect on the vector. Infected females of *balabacensis* lived longer than uninfected *maculatus*. The longer life span of *balabacensis* might be partially responsible for this species being such an efficient vector in nature.

These studies have been completed.

Table 1. Comparison of longevity in *Plasmodium vivax* infected and uninfected anopheline mosquitoes.

	<u><i>Anopheles balabacensis</i></u>		<u><i>Anopheles maculatus</i></u>	
	Infected	Uninfected	Infected	Uninfected
Number of lots	22	19	20	13
Longevity in days				
Mean	21.3	23.5	16.9	21.0
Range	1-38	1-51	2-34	2-34

REFERENCES :

1. Boyd, M.F. 1940. On the correlation between the incidence of stomach and gland infection in *Anopheles quadrimaculatus* infected with *Plasmodium vivax*. Am. J. Trop. Med. 20: 129-31.
2. De Buck, A. and N.H. Swellengrebel. 1935. On the seasonal longevity of *Anopheles maculipennis* in Holland with reference to their ability to act as malarial vectors. Proc. R. Acad. Sci. (Amsterdam) 38: 335-43.
3. Ragab, H.A.A. 1958. Effect of *Plasmodium* on the transmitting mosquito host from the point of view of the longevity of the infected mosquito. J. Egypt. Med. Assoc. 41: 447-54.
4. Schiefer, B.A., R.A. Ward and B.F. Eldridge. 1977. *Plasmodium cynomolgi*. Effects of malaria infection on laboratory flight performance of *Anopheles stephensi* mosquitoes. Exp. Parasitol. 41: 397-404.