

BITING AND RESTING BEHAVIOR OF *ANOPHELES DARLINGI* IN THE SURINAME RAINFOREST

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ABSTRACT. An experimental hut study was conducted on the biting and house resting behavior of *Anopheles darlingi* in the rainforest of Suriname. Of all anophelines collected on human bait at indoor and peridomestic sites, *An. darlingi* comprised 100% and 98%, respectively. A single peak in biting activity occurred around 2300 h. With hourly exit trap collections, a peak of exit from the hut was observed at sunrise. Of these mosquitoes, 91% were freshly fed. The indoor resting period was calculated to be about 4 h. When the human bait in the hut was protected by a mosquito net, only 18% of the exiting mosquitoes were fed and the resting period was about 3 h.

INTRODUCTION

Several patterns of biting behavior have been reported for *Anopheles darlingi* Root. In Suriname, Hudson (1984) found a peak in biting activity between 2230–2330 h² at Aselikamp, Lawa River. Peaks near midnight were reported in Amazonas, Brazil, by Charlwood and Hayes (1978) and in Colombia by Elliott (1972). Contrasting with these single peaks near midnight, Pajot et al. (1977) found a biting activity with peaks at dusk, in the middle of the night and at dawn in French Guyana. In southern Brazil (Forattini 1987), in Mato Grosso, Brazil (Charlwood and Wilkes 1979), and along the Ituxi River, Amazonas, Brazil (Roberts et al. 1987), peaks of biting were found at dawn and dusk but not at midnight.

Daytime resting *An. darlingi* were found in houses in the coastal regions of Venezuela and Guyana (Giglioli 1956). In those areas the strongly endophilic behavior of *An. darlingi* was the main reason for the successes of the DDT campaign in the decade following 1945. According to de Bustamente (1959), *An. darlingi* was found resting in houses at daytime in the Mato Grosso region of Brazil. Also, in Brazil, along the Ituxi River, Amazonas, Roberts et al. (1987) found a small portion of the *An. darlingi* population resting indoors through the gonotrophic cycle. However, also in the Amazon region (Deane et al. 1948, Charlwood and Wilkes 1979), in Colombia (Elliott 1972) and in Suriname (Hudson 1984; G. A. Fleming, PAHO-Entomologist in several unpublished reports, 1962–65, to the head of the antimalaria campaign), *An. darlingi* was only found resting at night but never by day.

The post-bite resting period in houses was estimated by Elliott (1972) at 150 min. Charlwood (1980) and Hudson (1984) estimated the pre-bite resting period to be 10 and 7.7 min, respectively. Roberts et al. (1987) observed a peak of entry at sunset and a peak of exit at sunrise. Regional differences in biting cycles and house resting behavior clearly exist. To determine local characteristics, the behavior of the geographically different populations should be observed in each area. In 1984, about 92% of all *Plasmodium falciparum* cases in Suriname originated from the Marowijne River area, which was selected as our study site.

Herein we will present data on man-biting activity and on the indoor resting period of fed and unfed mosquitoes.

Historically, studies on the behavior of *An. darlingi* in Suriname have been hampered by low *An. darlingi* population densities. Fleming (loc. cit.) constructed an experimental hut in Pokigron along the Upper-Suriname River without ever collecting a single specimen of *An. darlingi*. It was later confirmed by Rosendaal (1987) that the density of *An. darlingi* in this area is particularly low. Therefore, special attention was given to the selection of a site with population densities suitable for behavioral studies in a specifically constructed experimental hut.

MATERIALS AND METHODS

Selection of experimental station: In the most malarious area of Suriname, inhabited by the Djuka bush negro tribe, the basin of the Upper-Marowijne River with its tributaries, the Lawa and Tapanahony rivers, studies were performed on the biology of man biting anophelines in 1983–84 (Rozendaal 1987). One of the seven sampling stations, Abetredjoeka (Fig. 1), was selected for the present study because of its accessibility and relatively stable *An. darlingi* population densities. Abetredjoeka is a small bush negro settlement on one of the many is-

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² In Suriname sunset is at approximately 1900 h and sunrise at approximately 0600 h.

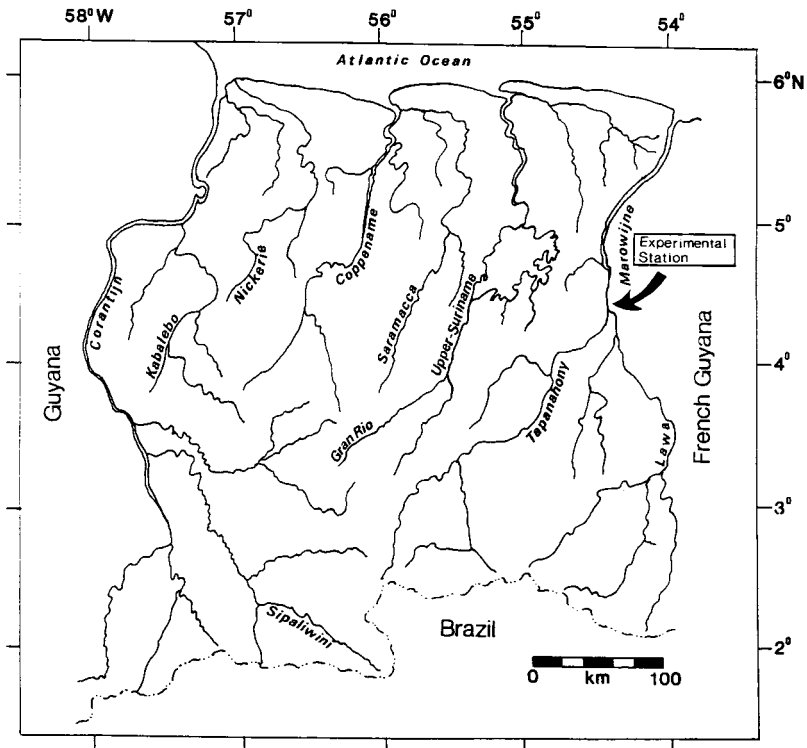


Fig. 1. Map of Suriname with location of the experimental station Abetredjoeka.

lands in the river. The population, usually between 20 and 30 people, practices slash and burn cultivation in the rainforest.

Indoor and peridomestic all night collections: Before using the experimental hut described in the following section mosquitoes were collected simultaneously indoors, in a bush negro hut, and peridomestically, at a site located between several houses. Collections from 1830 to 0730 h were separated into hourly catches. Six catchers used aspirators and flashlights to collect mosquitoes from their exposed legs. To reduce bias due to individual differences in skill and attraction, catchers were rotated between shifts. Mosquitoes were dried, preserved on naphthaline paper and sent to the laboratory for subsequent identification using the key of Faran and Linthicum (1981).

Experimental hut procedures for study of resting period: The traditional experimental hut with inlet louvers, inlet and exit traps, as described by the World Health Organization (WHO, 1975) was adapted for local use with due consideration for the low mosquito density, an average of 1.9/manhour during 1983–84. Therefore, exit traps and inlet louvers were constructed, but no entry traps were installed because a reduction in number of mosquitoes expected to enter the hut was considered

undesirable. The hut was constructed in local style with wooden walls and thatched roof (Fig. 2). The walls were made mosquito-proof to prevent uncontrolled escape, but the slits and gaps which occur in the walls of most houses were constructed with an inlet louver placed low along the long sides of the hut. The eve openings also were provided with baffles to permit entry but prevent escape. One exit trap was placed on each end wall. The exit traps were emptied each hour during the night and specimens were identified and abdominal status (Sella's stage) was determined. Biting activity was monitored by a man sitting on a chair in the hut who counted all mosquitoes feeding on his bared legs on an hourly basis. A flashlight with a red filter was used to observe the mosquitoes as they fed to repletion. The entry time and the time of biting were considered to be similar since the pre-bite resting period was reported to be very short (Hudson 1984, Charlwood 1980). The observation hours, from 1830 to 0730 h were equally divided by three of a six-man team every night (Fig. 3). To prevent bias due to individual differences, all observation periods were rotated among all the members of the field team. At 0800 h each morning, the hut was searched for resting mosquitoes.

Calculation of the resting period: According to

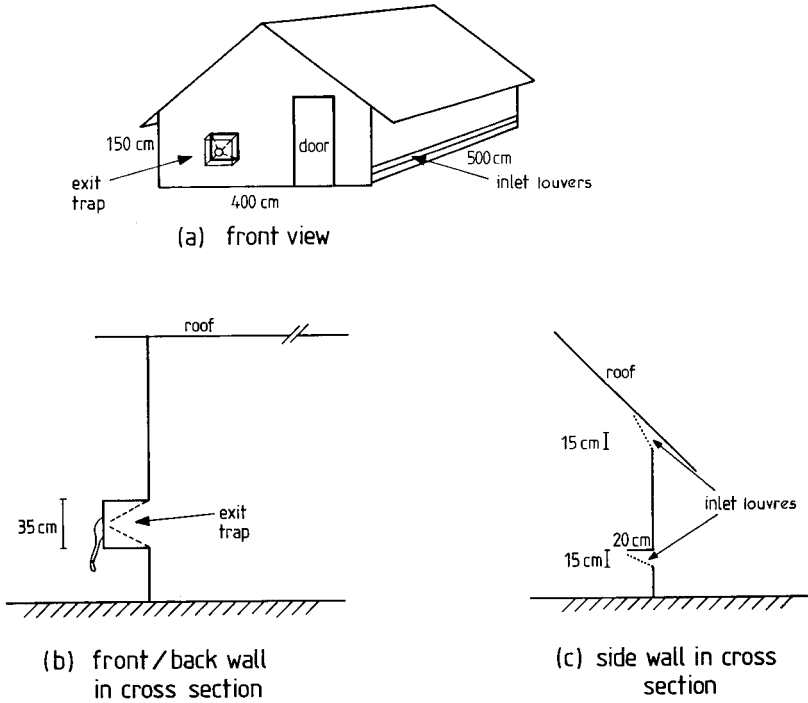


Fig. 2. Experimental hut. (a) front view; (b) front or back wall in cross section; (c) side wall in cross section.

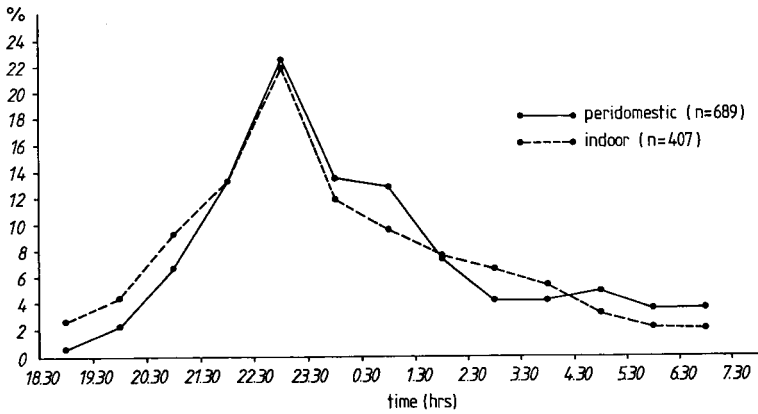


Fig. 3. Hourly percentage of total catch of *Anopheles darlingi* at indoor and peridomestic sites based on 26 nights of human bait collections from 1830-0730 h.

Elliott (1972) the frequency distributions of entry/exit times were normal. The normal distribution enabled him to equate the average indoor resting period with the time interval between the period 50% of the entering mosquitoes had arrived in the hut and the period 50% of the departing mosquitoes had left the hut. The data of this study, however, do not follow the normal distribution, and a more complicated model would be required to calculate the exact resting period. Assuming no difference in time between: a) entering the house and biting, b) time of

departure and time of collecting in the exit traps; and assuming that the percentiles of mosquitoes biting and the percentiles of mosquitoes exiting via the traps represent the same mosquitoes, we can calculate Surface III (Fig. 4), which represents the total number of percent hours for the interval after biting and before leaving. The average resting time is then approximately $1/100 \times$ Surface III. This is equal to $1,300 - (\text{Surface I} + \text{Surface II})$ (Fig. 4), in which Surface I = the total number of percent hours before biting and Surface II = the total number of

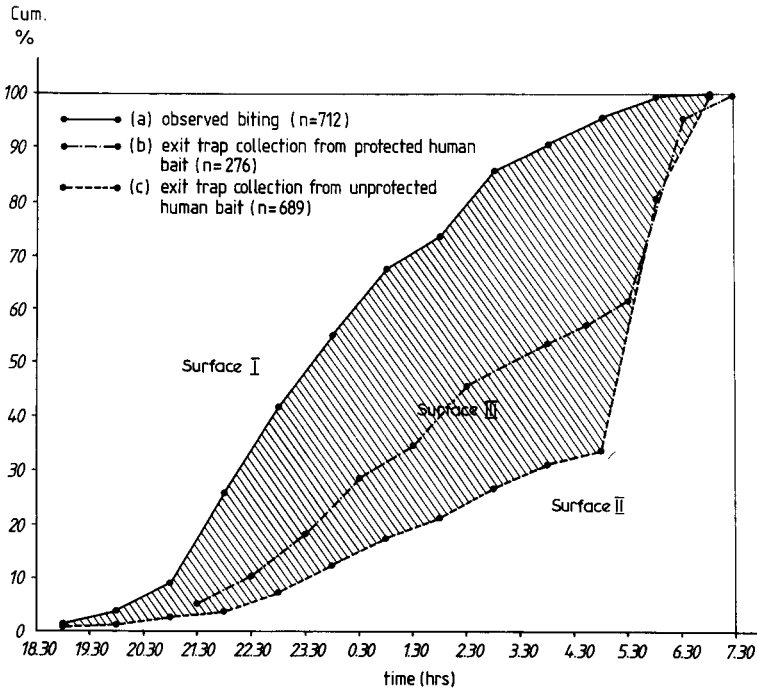


Fig. 4. Hourly cumulative percentage of total *Anopheles darlingi*: (a) observed biting human bait per hour from 1830-0730 over 57 nights; (b) collected per hour in exit traps from 2100-0800 h during 74 nights, human bait within the hut was protected with a mosquito net; (c) collected per hour in exit traps from 1830-0730 h during 57 nights, human bait within the hut was not protected with a mosquito net. Also indicated are: Surface I, the total number of percent hours before biting; Surface II, the total number of percent hours after departure; Surface III, the total number of percent hours after biting and before departure (striped).

percent hours after departure. In formula:

$$R \approx 1/100 [1300(\text{Surface I} + \text{Surface II})]$$

$$R \approx 1/100 \left[\sum_{i=1}^i O_i - \sum_{i=1}^i T_i \right]$$

R = Average indoor resting period.

$\sum_{i=1}^i O_i$ = Cumulative percentage of all mosquitoes observed biting until observation period i.

$\sum_{i=1}^i T_i$ = Cumulative percentage of all mosquitoes collected in the exit traps until period i.

To calculate the difference in time (D) of biting with the all night observations versus the all night collections, the same formula is used; but R = D and $T_i = C_i$, and C_i = percentage of all mosquitoes collected while biting on human bait during period i.

Experimental hut with human bait protected by a mosquito net. To more closely simulate the local situation in which bush negroes sleep with mosquito "nets" of closely woven cotton cloth, a second experimental hut was used in which the bait slept in a hammock surrounded by a mosquito net. The time of entry/biting could not be

determined but was expected to be similar to the time of entry/biting on the unprotected human bait-observer. Following local customs, the human bait retired to sleep at 2100 h. The exit traps were emptied every hour between 2100 and 0800 h and a difference of 30 min in collection times of protected and unprotected bait was needed because several experimental huts were operated simultaneously and only one man was available to empty all exit traps.

Period of study: Due to limited manpower, the three components of the study were conducted sequentially and not simultaneously. Thus, the indoor-peridomestic biting cycle was studied from July to September 1984, the house-resting period with an unprotected bait from December 1984 to June 1985, and the house-resting period with the protected human bait from August to December 1985.

RESULTS

Indoor and peridomestic all night collections: In Table 1 and Fig. 3, data are presented from both indoor and peridomestic catches. Although peridomestic collections yielded more mosquitoes, peak biting density was between 2230 and 2330 h in the two types of catches. *Anopheles*

darlingi was the only anopheline species collected indoors, with 407 specimens collected in 26 nights. Peridomestic collections yielded 689 *An. darlingi* and, in addition, 12 *An. nuneztovari* Gabaldon, one *An. oswaldoi* (Peryassu) and one *An. mediopunctatus* (Theobald).

House resting time of fed and unfed An. darlingi females: Data obtained during 57 nights of observations in the experimental hut with the unprotected human-bait observer are presented in Table 1 and Figs. 4-6. A total of 712 anophelines, or an average of 12 to 13 per night, were observed biting. A peak was observed between 2200 and 0100 h, and the mean time of biting is 29 minutes later than the mean time of biting when the mosquitoes were collected, as calculated with formula D.

A total of 689 *An. darlingi* were collected in exit traps ($\bar{x} = 12/\text{night}$); of these, 91.0% were freshly fed and the remainder unfed. More than 50% were captured exiting the hut after 0530 h. The morning search for resting mosquitoes yielded only 31 freshly fed *An. darlingi*, all resting on the inlet gauze baffle, presumably trying to leave the hut. The house resting period post-feeding (R) is calculated as 4.07 h.

A strong association between the numbers observed biting on a human bait indoors and the numbers collected in the exit traps is demonstrated by calculating the correlation coefficient (*r*) between the nightly totals of both parameters. The correlation appears to be highly significant ($r = 0.90, n = 57, P > 0.005$).

Results from the experimental hut with the

Table 1. Human bait collections at indoor and peridomestic sites, and experimental hut study with an unprotected human bait counting per hour all biting mosquitoes and hourly exit trap collections.*

| Observation period (i) | Time | Human bait collections (26 nights) | | | | | | Experimental hut study with unprotected human bait (57 nights) | | | | | |
|--------------------------|-----------|------------------------------------|------------|-----------------|--------------|------------|-----------------|--|------------|-----------------|-----------------|------------|-----------------|
| | | Indoor | | | Peridomestic | | | Observed biting | | | Exit trap coll. | | |
| | | No. coll. | % of total | Cum. % of total | No. obs. | % of total | Cum. % of total | No. obs. | % of total | Cum. % of total | No. coll. | % of total | Cum. % of total |
| 1 | 1830-1930 | 11 | 2.7 | 2.7 | 4 | 0.6 | 0.6 | 10 | 1.4 | 1.4 | 5 | 0.7 | 0.7 |
| 2 | 1930-2030 | 18 | 4.4 | 7.1 | 16 | 2.3 | 2.9 | 18 | 2.5 | 3.9 | 4 | 0.6 | 1.3 |
| 3 | 2030-2130 | 38 | 9.3 | 16.5 | 47 | 6.8 | 9.7 | 38 | 5.3 | 9.3 | 9 | 1.3 | 2.6 |
| 4 | 2130-2230 | 54 | 13.3 | 29.7 | 92 | 13.3 | 23.1 | 119 | 16.7 | 26.0 | 9 | 1.3 | 3.9 |
| 5 | 2230-2330 | 89 | 21.9 | 51.7 | 155 | 22.5 | 45.6 | 114 | 16.0 | 42.0 | 23 | 3.3 | 7.2 |
| 6 | 2330-0030 | 48 | 11.8 | 63.4 | 93 | 13.5 | 59.1 | 94 | 13.2 | 55.2 | 34 | 4.9 | 12.2 |
| 7 | 0030-0130 | 39 | 9.6 | 73.0 | 89 | 12.9 | 72.0 | 89 | 12.5 | 67.7 | 37 | 5.4 | 17.6 |
| 8 | 0130-0230 | 31 | 7.6 | 80.6 | 51 | 7.4 | 79.4 | 44 | 6.2 | 73.9 | 26 | 3.8 | 21.3 |
| 9 | 0230-0330 | 27 | 6.6 | 87.2 | 29 | 4.2 | 83.6 | 84 | 11.8 | 85.7 | 38 | 5.5 | 26.9 |
| 10 | 0330-0430 | 22 | 5.4 | 92.6 | 29 | 4.2 | 87.8 | 36 | 5.1 | 90.7 | 29 | 4.2 | 31.1 |
| 11 | 0430-0530 | 13 | 3.2 | 95.8 | 34 | 4.9 | 92.7 | 33 | 4.6 | 95.4 | 19 | 2.7 | 33.8 |
| 12 | 0530-0630 | 9 | 2.2 | 98.0 | 25 | 3.6 | 96.4 | 27 | 3.8 | 99.2 | 323 | 46.9 | 80.7 |
| 13 | 0630-0730 | 8 | 2.0 | 100.0 | 25 | 3.6 | 100.0 | 6 | 0.8 | 100.0 | 133 | 19.3 | 100.0 |
| Total/ $\sum_{i=1}^{13}$ | | 407 | 100.0 | 789.3 | 689 | 100.0 | 752.9 | 712 | 100.0 | 750.4 | 689 | 100.0 | 100.0 |

91.0% fed

* Indicated are per hour: the total number of *Anopheles darlingi* collected or observed, the hourly percentage of the total number collected or observed, the cumulative percentage of the total number collected or observed and the percentage fed.

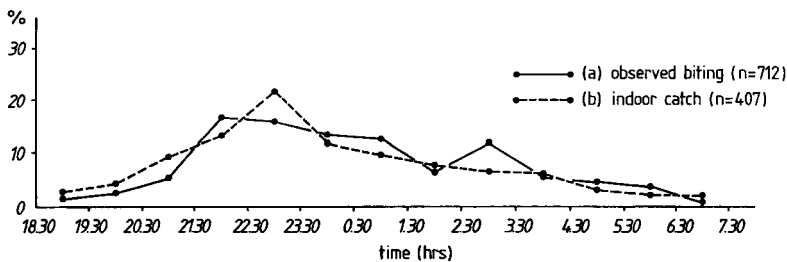


Fig. 5. Hourly percentage of total (a) observed biting per hour from 1830-0730 h over 57 nights and (b) collected indoors on human bait from 1830-0730 h over 36 nights.

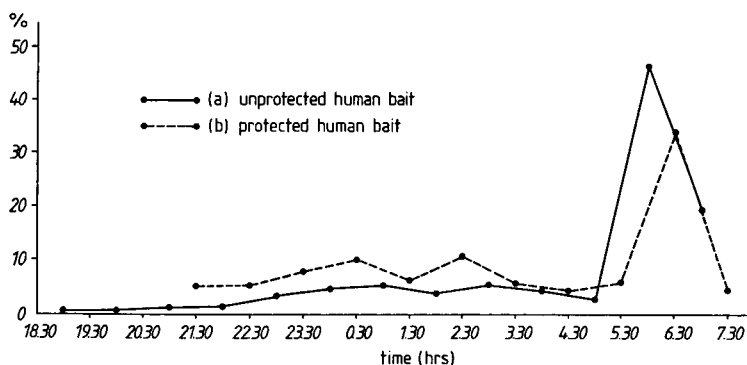


Fig. 6. Hourly percentage of total collected in exit traps (a) from 1830–0730 h, human bait within the hut was not protected with a mosquito net and (b) from 2100–0800 h, human bait within the hut was protected with a mosquito net.

Table 2. Hourly exit trap collections of *Anopheles darlingi* from an experimental hut with a human bait protected by a mosquito net (74 nights).*

| Observation period (i) | Time | Exit trap coll. with in hut protected human bait (74 nights) | | |
|------------------------|--------------------------|--|------------|-----------------|
| | | No. coll. | % of total | Cum. % of total |
| 1 | 2100–2200 | 14 | 5.1 | 5.1 |
| 2 | 2200–2300 | 15 | 5.4 | 10.5 |
| 3 | 2300–2400 | 22 | 8.0 | 18.5 |
| 4 | 2400–0100 | 28 | 10.1 | 28.6 |
| 5 | 0100–0200 | 17 | 6.2 | 34.8 |
| 6 | 0200–0300 | 30 | 10.9 | 45.7 |
| 7 | 0300–0400 | 16 | 5.8 | 51.5 |
| 8 | 0400–0500 | 12 | 4.3 | 55.8 |
| 9 | 0500–0600 | 16 | 5.8 | 61.9 |
| 10 | 0600–0700 | 93 | 33.7 | 95.3 |
| 11 | 0700–0800 | 13 | 4.7 | 100.0 |
| | Total/ $\sum_{i=1}^{11}$ | 276 | 100.0 | 507.2 |
| | | 18.1% fed | | |

* Indicated are per hour: the number of *An. darlingi* collected, the percentage of the total number collected, and the cumulative percentage of the total number collected.

protected human bait are reported in Table 2 and Figs. 4 and 6. During 74 nights of operation, an average of almost four per night, or 276 *An. darlingi*, were collected in the exit traps, and a majority were unfed (18.7% engorged). The formula for R was used to calculate the resting period. To compensate for the additional 30 min of exit trap collections in this hut, the same period was added to R. The resting period was calculated to be 2.56 h, but is probably a few minutes less since no collections were made between 1830 and 2100 h.

DISCUSSION

Biting activity and transmission: We have documented a biting pattern that peaks just before midnight. This pattern for *An. darlingi* activity is consistent with earlier observations by Hudson (1984), Elliott (1972) in Colombia and Charlwood and Hayes (1978) in Brazil.

In paired, indoor/outside, collections, the ready accessibility of outdoor collectors for blood-seeking mosquitoes probably accounts for the higher numbers encountered outdoors. However, under normal circumstances the human population is to be found indoors after 2100–2130 h. The tendency of *An. darlingi* to seek hosts inside human settlements rather than in forests (Rozendaal 1987) may be interpreted as an anthropophilic behavior. Coincidentally, *An. darlingi* was the only anopheline biting indoors at our study site. This and the observation that *An. darlingi* was the predominant species biting man outdoors in this study and at seven other sampling stations near Abetredjoeka (Rozendaal 1987) constitutes ample ecological evidence that *An. darlingi* is the principal malaria vector in the area. The late peak in biting activity occurs when most people are asleep in their homes; therefore, transmission apparently takes place indoors.

Accuracy and effectiveness of the experimental hut system: A total of 712 *An. darlingi* was observed biting indoors in the experimental hut. The numbers collected on the inlet baffles and those from the exit trap totaled 657 specimens. The small difference between the two numbers indicates a high capture rate. The discrepancy could be due to: (1) mosquitoes escaping through the inlet louvers, (2) some mosquitoes feeding more than once before repletion, and (3) mosquitoes biting but not feeding due to disturbance. The reliability of using the numbers of

mosquitoes observed biting indoors on a human bait as an indicator for the numbers of mosquitoes which entered the hut is demonstrated by the highly significant correlation between this parameter and the numbers collected daily in the exit traps.

The patterns of biting activity recorded from catches versus observations in the experimental huts were similar. The interval of 29 min between the means might be due to the study methods or to the differences in season and biting activity between studies as was found by Elliott (1968) in Colombia who compared biting activities during wet and dry seasons.

Fewer mosquitoes were collected from exit traps when the human bait was protected by a mosquito net than when the human bait was unprotected (4/night vs. 12/night). Either a masking effect of attractive odors by the net or the differences in study period and location of the experimental huts could be the cause of this reduction in collection size. Under the conditions prevailing in the study area, with low biting densities the experimental hut design proved to be effective for observing the indoor biting and resting habits of *An. darlingi*.

The indoor resting time of An. darlingi: Although the entomological assistants used their nets as carefully as possible during the study; nevertheless, 18.1% of the mosquitoes succeeded in taking a blood meal upon protected human bait. Local people are less careful in their use of nets, and this affects the feeding success and house resting time of the entering mosquitoes. Thus, under local conditions the indoor resting period is expected to be between 3 and 4 h.

In Colombia, Elliott (1972) found an interval of only 150 min whereas the resting time found by Roberts et al. (1987) in Brazil was much longer as peak entry occurred just after sunset and peak exodus was at sunrise. Additionally, small numbers of females rested in the house during the day. These differences in behavior of *An. darlingi* females probably reflect geographical variations between populations of this species.

The short indoor resting periods described in this report could reduce the effect of DDT residual house-spraying considerably, but factors such as DDT susceptibility, degree of avoidance/repellency, surface material, and duration of vector-insecticide contact are also of importance. Local studies on these subjects will be reported elsewhere (Rozendaal et al. 1989a, 1989b).

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