

## DIURNAL PATTERN OF HUMAN-BITING ACTIVITY AND TRANSMISSION OF SUBPERIODIC *WUCHERERIA BANCROFTI* (FILARIIDEA: DIPETALONEMATIDAE) BY *OCHLEROTATUS NIVEUS* (DIPTERA: CULICIDAE) ON THE ANDAMAN AND NICOBAR ISLANDS OF INDIA

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**Abstract.** We monitored diel-landing periodicity (biting activity/cycle) of *Ochlerotatus niveus* and the infection/infectivity pattern through human-landing collections on Teresa Island, which is remotely located in the Nicobar district of the Andaman and Nicobar group of Islands of India, for a period of one year. The biting activity was seen throughout the day, exhibiting a bimodal peak, the first at dawn (4:00–6:00 AM) and the other towards dusk (5:00–6:00 PM). This pattern was similar during all the seasons of the year. Peak biting hours of *Oc. niveus* coincides with the peak appearance of microfilariae. Overall infection and infectivity rates were 2.65% and 0.5%, respectively. Perennial transmission is evident from the records of vectors with parasites (infection), including infective larvae in all months of the year, although no infective mosquitoes were recorded at a few points. The risk of transmission of filariasis based on parity status of *Oc. niveus* was maximal at dusk (5:00–6:00 PM) in this region. The issue of control with respect to reducing human-vector contact is discussed.

### INTRODUCTION

In India, diurnally subperiodic *Wuchereria bancrofti* is prevalent only in the Nicobar group of islands, where it is reported to be a major public health problem in the remotely located Nancowry group of Islands. Microfilaremia prevalence ranging between 9.4% and 12% has been reported in this group of islands<sup>1–4</sup> for the past five decades. This form of filariasis on these islands is transmitted by *Aedes (Finlaya) niveus*.<sup>4</sup> Consequent to the elevation of subgenus *Ochlerotatus* to the genus rank and the reclassification of the other subgenera,<sup>5</sup> the species *niveus* is now placed in the genus *Ochlerotatus* and is currently known as *Ochlerotatus (Finlaya) niveus*.

The intensity of transmission of filarial infection in an area depends on the degree of human-vector contact. In addition, variations in the biting rhythm of different age groups of the mosquito vector also influences the transmission of filariasis.<sup>6</sup> Comparison of peak appearance of microfilariae and peak biting period would provide information on the relationship between the appearance and density of microfilariae in the peripheral blood of humans and the biting rhythm of the vector population, which will influence the dynamics of transmission of filariasis.<sup>7</sup> These are essential to understand the transmission potential of the vector species through time. In addition, information on biting periodicity will be useful in designing personal protection measures. *Aedes polynesiensis*, *Ae. pseudoscutellaris*, *Ae. vigilax*, and *Oc. niveus* are the principal vectors of the diurnally subperiodic form of *W. bancrofti*. Information on the biting periodicity is known only for *Ae. polynesiensis* and *Ae. pseudoscutellaris*.<sup>8–10</sup>

Previous studies in the Nicobar group of islands<sup>1,2,4</sup> were intended to assess the endemicity status from time to time. However, no published data on the biting periodicity and peak biting activity of *Oc. niveus* in the Nicobar group of islands are available. Therefore, we studied the biting periodicity of *Oc. niveus* and infection and infectivity status in the vector population. The results are presented in this report.

### MATERIALS AND METHODS

**Study site.** Approximately 500 islands are grouped into two districts, the Andaman and Nicobar Islands, which include the northern and southern islands, respectively. The Nancowry group of islands of the Nicobar district (8.5–9.5°N, 93–94°E) is a small region composed of seven remotely located islands (Bompoka, Chowra, Kamorta, Katchal, Nancowry, Teresa, and Trinket) with a population of approximately 25,000 people, mainly Nicobarese tribes who are at the risk of acquiring *W. bancrofti* filarial infection. The present study was carried out on Teresa Island (8°20'N, 93°7'–93°15'E) in Bay of Bengal. This island has an area of 87.04 km<sup>2</sup> and a population of 1,935 Nicobarese residing in 11 villages. The native inhabitants depend mainly on pigs for food, which they rear. For their livelihood, the people collect forest produce.

Temperatures on this island range between 24°C and 31°C and the humidity is high. Rainfall is heavy from May to November, and is influenced by both the southwest and northeast monsoons. In other months, rainfall is generally low, with February being the driest month. The mean annual rainfall has ranged between 100 and 273 mm over the last seven years. The soil is porous coral sand, which quickly absorbs rainwater, leaving hardly any water stagnant. Tree holes are the major breeding habitats of *Oc. niveus* in the Nancowry group of islands.<sup>4</sup> All villages are surrounded by forest interspersed with coconut and arecanut groves. The tribal population lives in typical huts raised on wooden stilts. This island also has densely forested tropical jungles. The villages are characteristic of a forest ecosystem, and are located and scattered within the forest area. Thus, villages and forests are inseparable.

**Mosquito collection.** Human-landing collections were carried out in 5 of 11 randomly selected villages. Before the start of the study, 24-hour human-landing collections were carried out in the selected villages. Since there were no *Oc. niveus* biting between 7:00 PM and 4:00 AM, human-landing collections were conducted between 4:00 AM and 7:00 PM. The collections were carried out once in a month in fixed catching

stations in selected villages for a one-year period. A human volunteer of the village acted as bait, and was made to sit on a raised wooden platform that forms the part of the Nicobarese hut, adjoining the forest fringe, while wearing his normal clothing. His exposed body surface was searched and the mosquitoes that were attempting to bite were collected using oral aspirators by an insect collector. During human-landing collections, insect collectors worked in shifts, but the same person acted as bait. The hourly collections of mosquitoes were kept separate and brought to the field laboratory alive, anesthetized with ether, and identified. The mosquitoes collected were dissected for determination of physiologic age<sup>11</sup> and filarial infection.<sup>12</sup> Mosquitoes positive for any filarial stage were considered as infected and those with infective stage larvae (L3) as infective. Because of the inherent logistic difficulties, human-landing collections were not carried out inside the forest.

**Statistical analysis.** The biting rhythm was computed as William's mean ( $M_w$ ),<sup>13</sup> which gives the best measure of central tendency. The  $M_w$  is a modified geometric mean that will compensate for many zero values, if any, in the data and is calculated using the following formula  $M_w = \text{anti log} [\text{sum} (x + 1) / N] - 1$ , where  $M_w$  = the modified geometric mean,  $X$  = the number of mosquitoes caught during different hours, and  $N$  = the number of observations.

The numbers of mosquitoes collected during daily human-landing collections over a one-year period were used for computation of  $M_w$  during different hours of the day. The  $M_w$  was also computed for different seasons of the year.

**Ethical aspects.** The study protocol was reviewed and approved by the Scientific Advisory Committee of Regional Medical Research Center (RMRC) in Port Blair. The study was also reviewed and approved by the Institutional Ethical Committee of the RMRC in Port Blair. While conducting the human-landing collections throughout the study period, adequate care was taken that the mosquitoes did not bite the human bait.

## RESULTS

**Species composition.** Overall, 4,039 mosquitoes belonging to 12 species were collected during the study period. *Ochlerotatus niveus* predominated (89.7%) among all the mosqui-

toes collected followed by *Ae. malayensis* (3.4%) and *Culex sp* (2.7%). Of 3,625 *Oc. niveus* mosquitoes collected during the study period, 2,767 (76.33%) were nulliparous. Among the parous mosquitoes, 659 (18.17%) had evidence of one contact with the host, 172 (4.74%) had two contacts, and 27(0.74%) had three contacts.

**Infection and infectivity status.** The dissection results of various species of mosquitoes are shown in Table 1. Among the 12 species of mosquitoes dissected, only *Oc. niveus* was naturally infected with *W. bancrofti* larvae. Of 3,625 *Oc. niveus* dissected, 96 were positive for filarial infection, with an infection rate (number of mosquitoes with filarial larva of any stage of the total dissected) of 2.65% and 18 were positive for L3, with an infectivity rate (number of mosquitoes with infective stage larvae of the total dissected) of 0.5%.

The overall infection and infectivity pattern observed in *Oc. niveus* during the study period is shown in Figure 1. Infected mosquitoes were found throughout the year, and infection was highest in January. Although there were some fluctuations in other months, the overall trend was a gradual increase after February up to April, then a steady decrease until June, after which there was a rapid decrease in July–August, and a gradual increase thereafter. Infectivity was observed to be highest in January with a decrease in February and a trend toward zero during March. Thereafter, it gradually increased to attain a small peak in the month of May, and then decreased from June to zero levels in July and August. It then gradually increased from September onward.

**Biting periodicity.** The biting periodicity, expressed as William's mean, is shown in Figure 2. The data indicate that the biting activity was seen throughout the day, with a major peak at dusk (5:00–6:00 PM) and a minor peak at dawn (4:00–6:00 AM). Seasonal analysis showed that the biting periodicity remained qualitatively similar in all seasons. However, in the summer months the peak during the dusk hours was more pronounced (Figure 3).

Both parous and nulliparous mosquitoes were found to bite throughout the day. The biting activity of parous mosquitoes with one host contact was found to exhibit fluctuations during the day without a noticeable peak. The biting pattern of mosquitoes with two or three host contacts also showed two peaks, one at dawn (4:00–6:00 AM) and the other towards dusk (5:00–6:00 PM) (Figure 4).

TABLE 1  
Species composition and dissection results of mosquito species\*

No.	Species	No. collected (%)	No. dissected	No. infected	No. infective	Infection rate (%)	Infectivity rate (%)
1	<i>Oc. niveus</i>	3,625 (89.7)	3,625	96	18	2.65	0.50
2	<i>Ae. malayensis</i>	138 (3.4)	138	0	0	0.00	0.00
3	<i>Cx. spp.</i>	110 (2.7)	110	0	0	0.00	0.00
4	<i>Cx. quinquefasciatus</i>	69 (1.7)	69	0	0	0.00	0.00
5	<i>Ar. subalbatus</i>	32 (0.8)	32	0	0	0.00	0.00
6	<i>An. sondaicus</i>	29 (0.7)	29	0	0	0.00	0.00
7	<i>Ae. albopictus</i>	17 (0.4)	17	0	0	0.00	0.00
8	<i>An. maculatus</i>	7 (0.2)	7	0	0	0.00	0.00
9	<i>An. theobaldi</i>	4 (0.1)	4	0	0	0.00	0.00
10	<i>Ae. fumidus</i>	4 (0.1)	4	0	0	0.00	0.00
11	<i>An. tessellatus</i>	3 (0.1)	3	0	0	0.00	0.00
12	<i>Ae. jamesi</i>	1 (0.0)	1	0	0	0.00	0.00

\* *Oc.* = *Ochlerotatus*; *Ae.* = *Aedes*; *Cx.* = *Culex*; *Ar.* = *Armigeres*; *An.* = *Anopheles*.

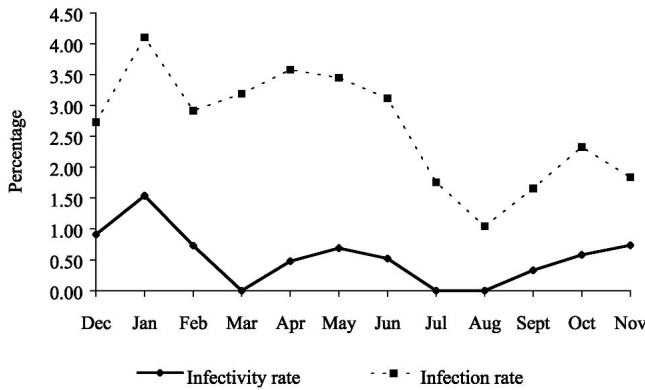


FIGURE 1. Overall infection and infectivity pattern in *Ochlerotatus niveus* during the study period.

## DISCUSSION

Previously, investigators in the Andaman and Nicobar Islands could not conduct intensive entomological studies on filariasis because of poor accessibility of an area, inadequate infrastructure, and the inherent hardships involved. Kalra<sup>1</sup> and Russel and others<sup>2</sup> could not incriminate the vector species because only few mosquitoes could be collected in the dry season. In addition, since these two studies were only point surveys, the biting periodicity of *Oc. niveus* and the infection and infectivity patterns over time could not be determined. However, in 1995,<sup>4</sup> *Ae. niveus* (*Oc. niveus*) was incriminated as the vector of diurnally subperiodic *W. bancrofti* in the Nancowry group of islands.

This is the first study in the Andaman and Nicobar Islands that covered all seasons. We determined the mosquito species composition, biting periodicity of *Oc. niveus*, and infection and infectivity status in *Oc. niveus*. *Ochlerotatus niveus* predominated among the mosquitoes collected. In the present study, the density of *Cx. quinquefasciatus* was very low, and the only species found naturally infected was *Oc. niveus*. The study also shows that only *Oc. niveus* is involved in the transmission of filariasis among the 12 mosquito species found to bite humans in the peri-domestic location of this island. At least five mosquito species/species groups, namely *Ae. (Finlaya) niveus*, *Ae. scutellaris* group, *Cx. quinquefasciatus*, *Mansonia (Mansoniodes) dives*, and *Anopheles sundaicus*, bite the

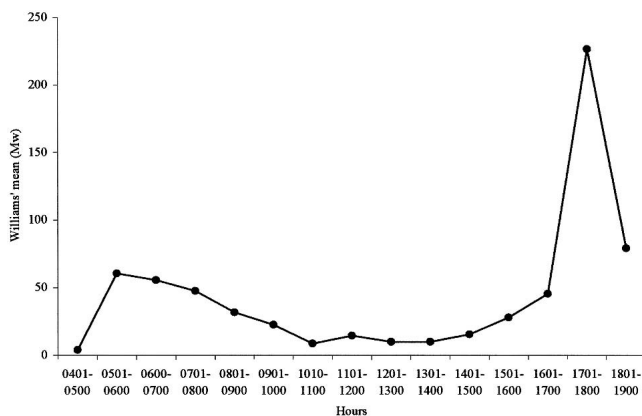


FIGURE 2. Biting periodicity of *Ochlerotatus niveus*.

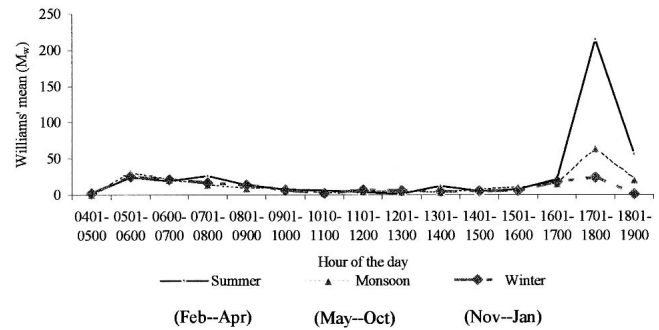


FIGURE 3. Seasonal biting periodicity of *Ochlerotatus niveus*.

native population.<sup>1,2,4</sup> In the present study, *Oc. niveus* was predominant among the mosquitoes collected in the human-landing catches. The documentation of 12 species biting the native population is probably due to the one-year observation period.

The biting behavior of any mosquito species is a biologic characteristic and could vary among species. The biting activity of *Oc. niveus* was seen throughout the day, thus showing that it is a diurnal species with bimodal peaks, a minor one at dawn (4:00–6:00 AM) and a major one at dusk (5:00–6:00 PM). The biting pattern was similar in all seasons, although the peak observed during the dusk hours in the monsoon season and cooler months is less pronounced. This could be due to the flushing of larvae from the tree holes because of heavy rains during these seasons. *Aedes polynesiensis* is the principal vector of subperiodic *W. bancrofti* over a wide area of the south Pacific islands. Studies carried out by Rakai and others,<sup>8</sup> Jachowski,<sup>9</sup> and Ramalingam<sup>10</sup> reported *Ae. polynesiensis* as being diurnally active with two peaks of activity, a lesser one in the morning (6:00–10:00 AM) and a greater one in afternoon (3:00–6:00 PM). Although the biting activity of *Ae. polynesiensis* was predominantly diurnal, some females were found to bite at night. A similar peak biting activity was observed in *Ae. pseudoscutellaris*.<sup>8</sup> The study of Ramalingam<sup>10</sup> also reported the biting activity of *Ae. (Stegomyia) tabu*, a member of the *scutellaris* group, which is a secondary vector of subperiodic *W. bancrofti* in Tongatabu, Samoa. Peak biting

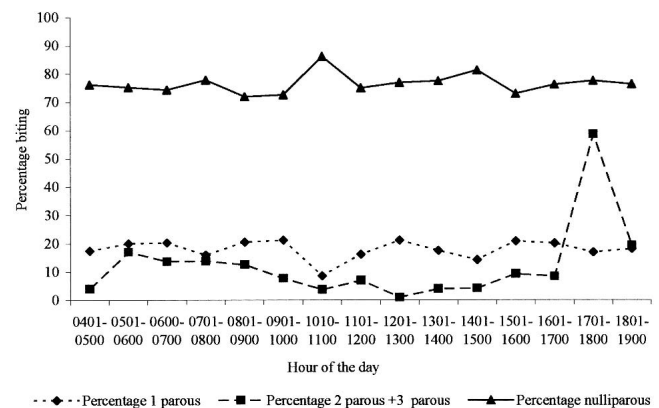


FIGURE 4. Biting periodicity of nulliparous, parous with one host contact (1 parous), and a combination of parous with two host contacts (2-parous) and parous with three host contacts (3-parous) *Ochlerotatus niveus* from day human-landing collections.

was observed between 10:00 AM and noon, with no biting before sunrise or after sunset, which showed that this mosquito is strictly a diurnal and crepuscular species. In the present study, a single collection carried out initially in the study villages to determine the biting pattern over a 24-hour period showed no *Oc. niveus* mosquitoes between 6:00 PM and 4:00 AM, suggesting that *Oc. niveus* is a diurnal species. Subsequent biting collections were carried out between dawn and dusk only. The biting pattern of *Oc. niveus* observed in the present study corresponds in general with the results of previous studies of *Ae. polynesiensis*<sup>9,10</sup>

Prior to the taxonomic revision, there were 25 species in the *niveus* group of mosquitoes. Subsequently, all of these species have been shifted to *Ochlerotatus* (*Fin.*). Information on the biting cycle/periodicity for other mosquitoes in the *niveus* group is not known, probably because it was not abundant. However, mosquitoes of the *Ae* (*Fin.*) *niveus* complex, namely, *Ae* (*Fin.*) *harinasutai*, have been implicated in the transmission of diurnally subperiodic *W. bancrofti*. Mosquitoes of this group are reported to be diurnally active species biting humans in the forested regions of southeast Asia.<sup>14</sup>

Analysis of microfilarial periodicity<sup>4</sup> showed that the density of microfilariae in the peripheral blood of humans was highly variable, with a peak at 6:00 PM and a trough between 3:00 AM and 6:00 AM. The present study shows that dusk was the peak biting hours of *Oc. niveus*, which coincides with that of the peak appearance of microfilariae. This facilitates the optimum infection of the vector population.

Several factors influence the number and periodicity of mosquitoes biting humans. Haddow<sup>15</sup> commenting on the biting activity of African mosquitoes, stated that there are different groups within the same species that bite in successive waves and that factors such as wind, impulse to bite, and proximity of breeding places influence the numbers biting at a given time. In the present study, the parous mosquitoes with one host contact were found to bite throughout the day, whereas, parous mosquitoes with two or three host contacts showed two peaks of biting activity corresponding to the overall biting cycle. The duration required for the development of microfilariae and L1 into L3 stage is 10–12 and 8–10 days, respectively,<sup>6</sup> and the potential for transmission comes from the female mosquitoes that survive a minimum of two gonotrophic cycles. The risk of transmission of filariasis due to *Oc. niveus* based on parity status was found to be in the dawn (4:00–6:00 AM) and dusk (5:00–6:00 PM) hours in this region.

Controlling *Oc. niveus* is ecologically and logistically challenging. The options for the control of *Oc. niveus* are very limited. Human-vector contact could be reduced by a time (peak biting period) and space (while working in the jungles) targeted approach of adopting personal protective measures such as protective clothing and repellants. These measures need to be encouraged among the native population. If one considers the enormous control of the tribal chieftains and the pastors have over the tribal community, the native population could be sensitized and motivated by the tribal chieftains and pastors to adopt such measures. In addition, these measures could be popularized through radio programs in their native language. The key to implementation of these personal protective measures are health education and socioeconomic development. Protective clothing and repellants would reduce

the annual infective biting rate and reduce transmission potentials.

In light of the findings of this study, we conclude that the biting periodicity of *Oc. niveus* is diurnal, with bimodal peaks. The peak biting activity of this mosquito coincides with the peak appearance of microfilariae. Infection and infectivity observed in the vector population indicate perennial transmission in a typical sylvatic ecosystem. The risk of acquiring an infection based on parity status could be elucidated towards the dawn and dusk hours, as shown by the bimodal peaks of biting activity of mosquitoes, namely, a minor peak at dawn and a major peak at dusk. Human-vector contact could be reduced by a time and space targeted approach of adopting personal protective measures such as protective clothing and repellants.

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