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Microfilarial periodicity of *Wuchereria bancrofti* in Vanuatu

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Abstract

A study on the relationship between the microfilarial periodicity of *Wuchereria bancrofti* and vector biting activity was carried out in Penama province, Vanuatu from February to April 1999, to enable the design of a more efficient strategy to control filariasis transmission. The microfilarial periodicities of 22 *W. bancrofti* antigen-positive volunteers were studied. Microfilariae (mf) were counted every hour for 24 h for 6 volunteers and every hour for 12 h (from 18:00 to 06:00) for 16 volunteers. At the same time as the preparation of mf test slides, indoor human landing catches of the vector mosquito, *Anopheles farauti*, were conducted to assess the vector biting activity. The time of peak microfilaraemia was 01:32 and the microfilarial periodicity index was 112.3, confirming the nocturnal periodicity of *Wuchereria bancrofti* in Vanuatu. Nearly all (98.5%) of the mf appeared during the time periods when *A. farauti* were collected. The timing of vector biting activity corresponded to the time of mf circulation.

Keywords: filariasis, *Wuchereria bancrofti*, *Anopheles farauti*, transmission, microfilarial periodicity, Vanuatu

Introduction

Following the global filariasis elimination programme (GPELF) formulated by the WHO, the Government of Vanuatu integrated a filariasis elimination programme as part of the Pacific Programme for the Elimination of Lymphatic Filariasis (PacELF). In June 2000, the first nationwide mass drug administration (MDA) for filariasis using a combination drug regimen of albendazole (400 mg/person) and diethylcarbamazine citrate (DEC; 6 mg/kg bodyweight) was launched in Vanuatu. Prior to the first MDA, a national filariasis screening survey, using a statistically stratified sampling strategy, was conducted from 1997 to 1998. The overall prevalence of microfilariae (mf) was 2.77% and mf density was 14.82/60 μL. The highest prevalence and transmission potential were found in Penama province (K. Ichimori, unpublished data).

Filariasis in Vanuatu is caused by nocturnally periodic *Wuchereria bancrofti*, transmitted by *Anopheles farauti* (Sasa, 1976). However, there is no further information on microfilarial periodicity and vector mosquitoes in Vanuatu. Detailed knowledge of microfilarial periodicity in relation to vector biting activity will enable the design of vector control strategies as adjunct control measures to the PacELF- administered MDA. Such a multidisciplinary control approach should enable the design of a more efficient strategy to control filarial transmission in Vanuatu.

Materials and Methods

This study was carried out from February to April 1999. Villages were selected randomly on 3 islands in Penama province. Nasawa, Sakau, and Wanur villages were identified as highly endemic filariasis villages through the national filariasis survey. Baitora and Lolovatali villages, located near Nasawa and Sakau villages, respectively, were also included in the study. All residents participating in this study were native Melanesians.

Antigen test

A rapid test for *W. bancrofti* antigen (ICT Diagnostics©, AMRAD ICT, NSW, Australia) was used to identify antigen-positive individuals. Children aged < 15 years were excluded from the antigen test. One hundred microlitres of whole blood were collected in a capillary tube by finger-prick from each individual and added to the test card. The results were recorded within 15 min.

Microfilariae count

Using a haemoglobin pipette, 60 μL of finger-prick blood were smeared in 3 lines, dried and Giemsa-stained. The total number of mf in the 3 lines was counted and recorded for each slide.

To assess microfilarial periodicity, mf slide tests were carried out 24 h or 12 h prior to enrolment in the study. Volunteers were educated on the procedures to be used as well as the risks and benefits to them and their community of their participation in this study. Volunteers were then tested for microfilarial periodicity every hour for 24 h in Wanur village or every hour for 12 h at night (from 18:00 to 06:00) in the other villages.

Mosquito collection

Anopheles farauti were collected by indoor human landing catches, using an aspirator, by volunteers from each village, for 15 min every hour with hourly collections of mosquitoes stored separately. Mosquitoes were identified using the keys of Belkin (1963) and numbers of females were recorded. The use of volunteer mosquito collectors was approved by the Government and their communities. Mosquito collectors were provided with a single dose of DEC tablets (6 mg/kg bodyweight).

Mosquito collections were conducted every hour for 24 h in Wanur village or every hour for 12 h at night (from 18:00–19:00 to 06:00–07:00) in the other villages. A total of 22 collections was carried out.

Microfilarial periodicity

The microfilarial periodicity index and the peak hour as defined by Sasa and Tanaka (1974), (S–T method) and Aikat and Das (1977) (A–D method) were calculated using the data sampled every hour for 24 h from the 6 volunteers from Wanur village.

Results

Microfilarial periodicity

The 22 study volunteers positive for *W. bancrofti* antigen from the 5 villages ranged in age from 16 to 67 years (Table).

The mf counts of the 6 volunteers from Wanur village were conducted every hour throughout a 24 h cycle; 97.2% of positive mf counts were detected from 18:00 to 06:00 and 98.5% were detected from 18:00 to 07:00. Microfilariae were found during daylight in 2 high-density mf carriers but the mf densities were lower than those observed in the same individuals at night.

Based on the S–T calculation method, the theoretical microfilarial periodicity and the mf counts of the 6 volunteers from Wanur village for 24 h are shown in

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Table. Characteristics of *Wuchereria bancrofti* antigen-positive volunteers by island and village of residence in Penama province, Vanuatu, February–April 1999

Subject no.	Age (years)	Gender	Mf slide test (h)
Pentecost			
Wanur			
1	31	F	24
2	38	M	24
3	64	M	24
4	67	M	24
5	27	F	24
6	42	F	24
Ambae			
Sakau			
7	37	M	12
8	34	F	12
Lolovatali			
9	65	M	12
10	55	M	12
11	49	M	12
12	16	F	12
Maewo			
Nasawa			
13	45	F	12
14	58	M	12
15	55	F	12
Baitora			
16	42	M	12
17	38	F	12
18	23	F	12
19	21	M	12
20	27	M	12
21	18	F	12
22	17	F	12

Fig. 1. The periodicity index was 112.3 and the peak hour was 1.54, corresponding to 01:32. The actual mf counts had 2 clear peaks at 22:00 and 04:00 and were considerably different from the theoretical microfilarial periodicity calculated.

To investigate the possible significance of the 2 peaks in mf counts, the mf counts of the 16 volunteers from the other 4 villages were counted each hour for 12 h. All 22 volunteers were grouped according to mf density: mf count < 50 per 60 µL blood in group 1, > 50 mf to 500 mf per 60 µL blood in group 2, and > 500 mf per 60 µL blood in group 3. The mean ratios of mf counts of each group are shown in Fig. 2. Two peaks of mf density were not found in any of these groups suggesting that the bimodal pattern seen in the mf densities of the 6 individuals from Wanur village was due to the small sample size.

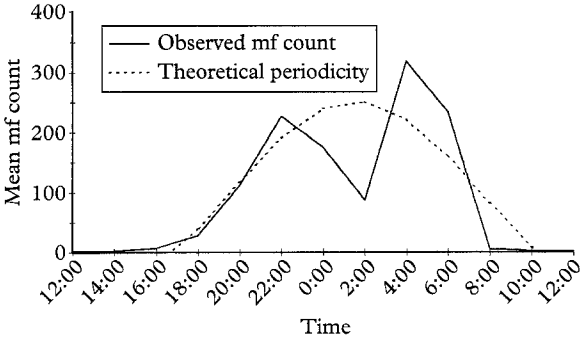

Fig. 1. Mean microfilariae (mf) count and theoretical microfilarial periodicity index during a 24 h period in six *Wuchereria bancrofti* antigen-positive volunteers from Wanur village, Penama province, Vanuatu, February–April 1999. Theoretical periodicity: $Y = 100 + 1.347D\cos 15(h - k)$ where $D = 112.30$ (microfilarial periodicity index) and $k = 1.54$ (peak hour).

Mosquito biting activity

A total of 159 *A. farauti* females was collected by indoor human biting catches during the 22-night study period, with 88 *A. farauti* collected from 7 volunteers in Baitora. All female *A. farauti* were caught between 18:00 and 07:00. The biting activity time corresponded to the time of mf appearance in circulating blood (Fig. 3). The peak *A. farauti* biting activity period was 22:00–23:00 when 19.9 females per person per hour were collected. After midnight a small peak was found again at 04:00–05:00 when 12.7 females per person per hour were collected.

The cumulative ratio of collected females at 21:00 was 25.0%, at 00:00 it was 60.1%, and at 03:00 it was 77.2%.

Discussion

Microfilarial periodicity

Wuchereria bancrofti in Vanuatu was nocturnally periodic, similar to the peak biting times in other geographical areas — 00:56 in Kenya (Gatika *et al.*, 1994), 01:11 in Sri Lanka (Weerasooriya *et al.*, 1998), 01:32 in Malaysia (Hii *et al.*, 1985), and 01:52 in Tanzania (Simonsen *et al.*, 1997). The microfilarial periodicity index calculated from this study was similar to those found in other studies — 117.5 in Tanzania (Simonsen

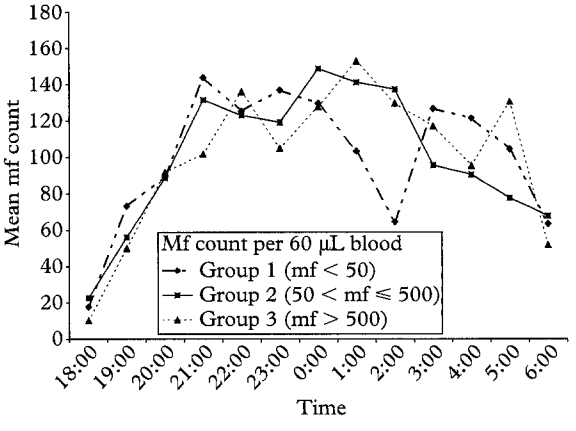

Fig. 2. Hourly observed microfilariae (mf) count in 22 *Wuchereria bancrofti* antigen-positive volunteers from Penama province, Vanuatu, February–April 1999.

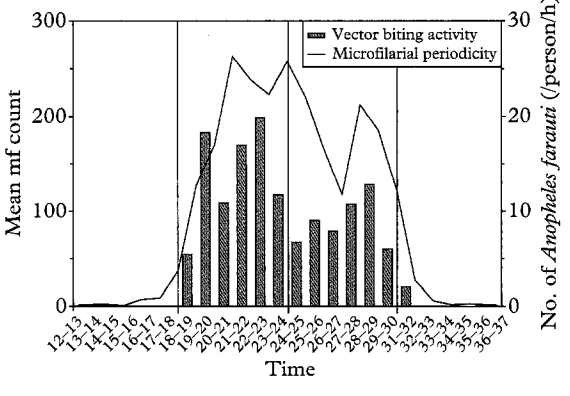

Fig. 3. Relationship between vector biting activity and observed microfilarial periodicity during a 24-h period, Penama province, Vanuatu, February–April 1999. Total number of mosquitoes collected = 159.

et al., 1997), 117.8 in Kenya, 139.2 in Sri Lanka, and 137.2 in Malaysia (Hii *et al.*, 1985).

Mosquito biting activity and microfilarial periodicity

Vector biting activity was investigated from 18:00 to 07:00 and the results of human landing collections from another study in Vanuatu were compared with this study. Ree *et al.* (1991) carried out a human bait collection of *A. farauti* on Santo Island and landing catches showed that the percentage of total *A. farauti* females collected was 21.3% at 21:00, 65.0% by 00:00, and 80.1% by 03:00 which is similar to the results of this study. The appearance of mf corresponded to the times of *A. farauti* biting activity; 98.5% of mf appeared in peripheral blood from 18:00 to 07:00, when all *A. farauti* were collected.

As some mf were found in blood samples in the daytime, very limited transmission of *W. bancrofti* by daytime biting species of mosquito, such as *Aedes scuttellaris* in Polynesian countries (Ramalingam, 1968), is possible.

In Vanuatu, *A. farauti* is the only malaria vector. If a barrier exists between the *A. farauti* and humans during the time of *A. farauti* blood-feeding, transmission of both malarial and filarial parasites can be prevented. One of the best barriers is an insecticide-treated mosquito net. Most people go to bed by 21:00 in rural villages in Vanuatu (Ree *et al.*, 1991). According to the results of mosquito landing catches in this study, up to 75% of transmission by *A. farauti* might be prevented if all the villagers were protected by a mosquito net by 21:00.

Mosquito nets are the main strategy of the national vector-borne diseases control programme for malaria control in Vanuatu. Since 1988, when permethrin-impregnated mosquito nets were introduced, a marked decrease in the incidence of malaria has been seen. It is also assumed that widespread mosquito net usage can contribute to interruption of filarial transmission. However, the distribution of these 2 diseases is not uniform over the country and there is no significant correlation between the geographical distribution of filarial antigenaemia and malaria incidence in Vanuatu (K. Ichimori *et al.*, unpublished data). Therefore nationwide measures, like mosquito nets, are needed to prevent malaria and filariasis. Mosquito control using mosquito nets should be encouraged to enhance the impact of MDA.

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Announcement

ROYAL SOCIETY OF TROPICAL MEDICINE AND HYGIENE
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