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Natural infections of *Wuchereria bancrofti* in *Aedes (Stegomyia) polynesiensis* and *Aedes (Finlaya) samoanus* in Samoa

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Abstract

Seven years after the 2nd mass treatment of the population with diethylcarbamazine, transmission of subperiodic *Wuchereria bancrofti* was studied in four villages in Samoa during one year by means of biting catches of *Aedes polynesiensis* and *A. samoanus*. 2 villages were coastal, one inland bush and the other an inland coconut plantation community. Overall infection and infective rates from 6702 *Ae. polynesiensis* were 0·84 and 0·27% respectively, and the infection rate from 2858 *Ae. samoanus*, collected in 10-minute catches from 24 sites, was 0·65%. No infective *Ae. samoanus* was found in these samples. 12-hour all-day catches in the 2 coastal villages confirmed active transmission by *Ae. polynesiensis*. 12-hour all-night catches in the same 2 villages recorded high transmission by *Ae. samoanus* although there was little evidence of local breeding. The annual transmission potential for *Ae. polynesiensis and Ae. samoanus* was high in one of the coastal villages and low in the otther. A total of 221 infected *Ae. polynesiensis* and 40 *Ae. samoanus* were recorded. Of the 72 infective *Ae. polynesiensis*, 59·1% contained 1 to 2 larvae each (median density 1·4); 70% of the 40 infected *Ae. samoanus* had 1 to 2 larvae (median density 1·1). From the proportion of infective *Ae. polynesiensis* the mean probability of survival was estimated as 0·917.

Introduuction

Filariasis due to diurnally subperiodic *Wuchereria bancrofti* in Samoa is transmitted mainly by 2 vectors, the day-biting *Aedes (Stegomyia) polynesiensis* (Marks) (BUXTON, 1928) and the night-biting *Ae. (Finlaya) samoanus* (Gruenberg) (RAMALINGAM & BELKIN, 1964). While *Ae. polynesiensis* is a container breeder, choosing a range of containers such as water storage drums, discarded receptacles and tree holes to oviposit, *Ae. samoanus* is a leaf axil breeder, restricting itself to axils of plants belonging to the family Pandanaceae, such as *Freycinetia*, a climber on forest trees, and *Pandanus*, the screwpine.

The first blood surveys of the country conducted in 1965 in 21 villages showed a microfilarial rate of 19·1%, which reduced to 1·62% after the first nationwide mass drug adminstration campaign using diethylcarbamazine citrate and to 0·24% after a 2nd treatment in 1971 (Muang & Penaia, 1976, unpublished WHO Report). In 1975, the microfilarial rate was 2·1%. Later, using the more sensitive membrane filtration method with 1 ml of venous blood, higher microfilarial rates were detected including several very low densitites of fewer than 10 microfilariae (mf) per ml. The finding of these residual low densities introduced new problems in planning control programmes. The present project commenced in 1977 to study further the epidemiology of filariasis and the ecology and bionomics of its vectors and to evaluate methods of control.

Materials and Methods

Study area

Four villages were selected on the island of Upolu within easy reach of the capital, Apia (Fig. 1). Two of the villages were coastal, Vailu'utai, 30 km to the west and Luatuanu'u, 15 km to the east of Apia. Vaipapa was an isolated community in an inland coconut plantation and Lalomauga was also an inland village, 35 km south-east of Apia, at the foot of a mountain. Coconut was grown as a cash crop and home gardens were planted with banana, breadfruit and cocoa.

Mosquito collections

(1) *Biting collections of ten minutes at each site.* Mosquitoes landing on man were collected for a period of 10 minutes at each of 12 indoor and 12 outdoor sites from 0830 to 1030 h and from 1600 to 1800 h for *Ae. polynesiensis*, and at 24 indoor sites from 1930 to 2130 h for *Ae. samoanus*. One morning catch and one afternoon catch were carried out each month in Vailu'utai and Vaipapa and one morning catch and one night catch were made in Luatuanu'u and Lalomauga from December 1977 to November 1978. Preliminary early night catches in Vailu'utai and Vaipapa yielded no *Ae. samoanus* and hence they were discontinued in these two villages.

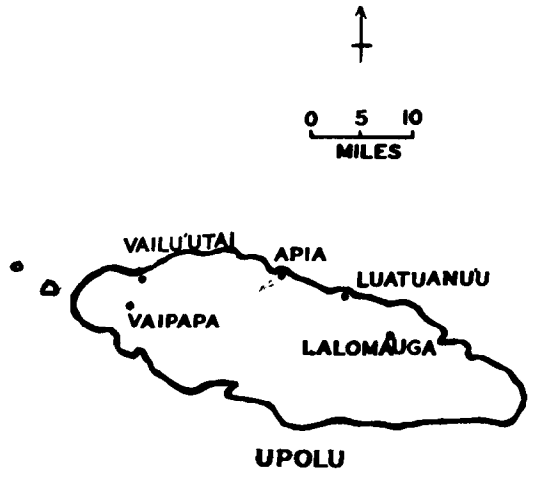

Fig. 1. Location of the four study villages in Upolu island in relation to Apia.

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(2) *Twelve hour day and night biting catch.* Catches of 12-hour duration from shortly before 0600 h to 1800 h (day) and from shortly before 1800 h to 0600 h (night) were made once a month at a single site in Vailu'utai from December 1977 to May 1979 and from December 1977 to December 1978 in Luatuanu'u.

Dissection

Only *Ae. polynesiensis* from the day collections and *Ae. samoanus, Ae. polynesiensis* and *Culex quinquefasciatus* from the night collections were dissected. Nearly all the mosquitoes collected were dissected except in the larger catches of *Ae. samoanus* at Luatuanu'u where only 20 to 50% were processed. The head, thorax and abdomen were placed in three drops of saline containing congo red and examined under the compound microscope for filarial infection. All stages of larvae were counted after identification by the morphological characteristics given by NELSON (1959) and RAMACHANDRAN (1970). About 5 to 20 specimens of the vector species from each of the 24 sites in the 10-minute catch series and from each hour in the 12-hour catch series were dissected by one of us (W.A.S.) for individual age grading by the Polovodova method (DETINOVA, 1962).

During the study, two seasons, a warm season from December to May and a mild cool season from June to November, occurred (SAMARAWICKREMA *et al.*, 1985b). The developmental period of mf in the vectors in laboratory experiments was shorter in the warm season.

Results

The total species composition of the samples of mosquitoes taken by the two methods will be described in another paper. Two species of filariae were identified in the mosquitoes dissected: *Wuchereria bancrofti* and *Dirofilaria immitis* (RAMALINGAM, 1968).

Transmission in the four villages

The results of dissections of *Ae. polynesiensis* from the four villages are given in Table 1. 6702 *Ae. polynesiensis*, 84·6% of the collection from the 4 villages over the year, were dissected; 56 were infected, including 18 with infective larvae, giving an overall infection rate of 0·84% and infective rate of 0·27%. Infected *Ae. polynesiensis* were collected both indoor and outdoor in all 4 villages. Infective *Ae. polynesiensis* were found in Vailu'utai, Vaipapa and

Table 1—Results of dissections of *Ae. polynesiensis* in catches (10 minutes per site) in the four villages

	Indoor			Outdoor			Total	
Village	No. dissected	No. infected	No. infective	No. dissected	No. infected	No. infective	Infection rate	Infective rate
Vailu'utai*	660	4	1	3053	31	12	0·94	0·35
Vaipapa*	448	3	0	591	5	2	0·77	0·19
Luatuanu'u	105	2	0	237	1	0	0·88	0·00
Lalomauga	510	2	0	1098	8	3	0·62	0·19

*The morning and afternoon catches in Vailu'utai and Vaipapa have been combined

Table 2—Results of dissections of *Ae. polynesiensis* and *Ae. samoanus* in 12-hour day and 12-hour night catches in Vailu'utai and Luatuanu'u

	Ae. polynesiensis								*Ae. samoanus*				
	Indoor			Outdoor			Total		Indoor				
Village	No. dissected	No. infected	No. infective	No. dissected	No. infected	No. infective	Infection rate	Infective rate	No. dissected	No. infected	No. infective	Infection rate	Infective rate
Vailu'utai	987	32	10	6007	129	44	2·30	0·77	791	14	11	1·77	1·39
Luatuanu'u	450	2	0	1077	2	0	0·26	0·00	7354	14	5	0·19	0·07

Table 3—Estimates of annual biting and infective biting rates and the annual transmission potential for *Ae. polynesiensis* and *Ae. samoanus* in 2 coastal villages in Samoa

	Vailu'utai		Luatuanu'u	
	Ae. polynesiensis	*Ae. samoanus*	*Ae. polynesiensis*	*Ae. samoanus*
Indoor				
Annual biting rate	25862	12145	14680	168667
Annual infective biting rate	173	172	0	125
Annual transmission potential	426	483	0	255
Outdoor				
Annual biting rate	150268	—*	35344	—
Annual infective biting rate	968	—	0	—
Annual transmission potential	3433	—	0	—

*—indicates no data obtained

Lalomauga. Transmission was most active in Vailu'utai, where *Ae. polynesiensis* with early stage larvae were found in every month of the year and no difference was observed between warm and cool seasons. Fewer infected mosquitoes were found in the other 3 villages.

The dissections of early night catches of *Ae. samoanus* in Luatuanu'u and Lalomauga showed low infection rates. In Luatuanu'u 748 *Ae. samoanus* were dissected during the year of which 4 (0·53%) were infected. In Lalomauga 1110 *Ae. samoanus* were dissected during the same period and 8 (0·72%) were infected. Overall infection rate was 0·65%. No infective *Ae. samoanus* was found in these collections.

Transmission at fixed sites in two villages

Results of dissections of mosquitoes taken at a single site in each of the coastal villages, Vailu'utai and Luatuanu'u, from 12-hour day and 12-hour night catches are given in Table 2. These values represent the infections over a period of 18 months in Vailu'utai and 12 months in Luatuanu'u at fixed sites. In Vailu'utai, infected *Ae. polynesiensis* were found every month, confirming that transmission occurred throughout the year. The low infection rate of *Ae. polynesiensis* in Luatuanu'u was confirmed in the 12 hour day catches over a year suggesting a low prevalence of microfilaraemia in the village. No mf prevalence survey was carried out here.

The few *Ae. samoanus* taken in Vailu'utai were all caught during the latter part of the night; high infection and infective rates were observed. Active transmission was also seen in the large *Ae. samoanus* population at Luatuan'u in 5 months of the year in both warm and cool season. The scanty plantations of *Pandanus* in the village, and the absence of *Freycinetia*, rendered it impossible for Luatuanu'u to support the breeding and emergence of such a large local population of *Ae. samoanus*. The mosquitoes probably invaded from breeding places outside the village, suggesting that at least some of the infections were introduced into the village in this way. Again, no difference in transmission was observed in the warm and cool seasons.

Annual tranmission potential

The 12-hour biting catches of *Ae. polynesiensis* and *Ae. samoanus* at Vailu'utai and Luatuanu'u were used to estimate the risk of infection from the number of bites per man per year (annual biting rate) and the number of infective larvae a man would be exposed to per year (annual transmission potential) (Table 3). The annual outdoor biting rate of *Ae. polynesiensis* at Vailu'utai was comparable to that of *Ae. samoanus* indoors at Luatuanu'u. As a result of high infective rates and high outdoor populations of *Ae. polynesiensis*, the estimated annual transmission potential at Vailu'utai was very high outdoors. Indoors the transmission of *Ae. polynesiensis* and *Ae. samoanus* was similar at Vailu'utai.

Intensity of transmission

221 infected *Ae. polynesiensis* were found in the 4 villages; 86 had sausage stage, 63 had stage II and 72 had infective larvae. Of the 72 infective mosquitoes, 36·1% had one larva each, 59·7% 1 to 2 larvae, 66·7% 1 to 3 larvae, 70·8% 1 to 4 larvae, 76·4% 1 to 5 larvae, 83·3% 1 to 6 larvae, 88·9% 1 to 7 larvae, 91·7% 1 to 8 larvae, 93·1% 1 to 9 larvae and 94·4% 1 to 10 larvae. The median density of the infective larvae was 1·4; those of stage II and sausage stage larvae were 1·9 and 2·2 respectively.

Of the 40 infected *Ae. samoanus* taken from 3 to 4 villages during the study, 47·5% had one larva each, 70·0% 1 to 2 larvae, 87·5% 1 to 4 larvae and 97·5% 1 to 9 larvae. The median density of all larvae was 1·1. These results from field material are consistent with the laboratory finding of higher microfilarial intake by *Ae. polynesiensis* than by *Ae. samoanus* when fed on bloods with similar microfilaraemia (SAMARAWICKREMA *et al.*, 1985a).

Age of infective Ae. polynesiensis

Of 56 infected and age-graded *Ae. polynesiensis*, 18 with sausage stage larvae were 1-parous, 8 with stage II larvae were 2-parous and 28 of 30 with infective larvae were 3-parous. This suggests that *Ae. polynesiensis* taking a blood meal after 3 ovipositions would be potentially infective. The number of infected *Ae. samoanus* age-graded was too small to observe a definite correlation.

Discussion

In entomological surveys in Samoa before the first mass drug administration, RAMALINGAM & BELKIN (1964) dissected 407 *Ae. polynesiensis* and found 12 (2·95%) infective and 380 *Ae. samoanus* with one (0·26%) infective, and MCCARTHY (1965, unpublished report) dissected 1120 *Ae. polynesiensis* and found 29 (2·60%) infective and 466 *Ae. samoanus* with 4 (0·86%) infective. SUZUKI & SONE (1975) dissected 4290 *Ae. polynesiensis* and 1148 *Ae. samoanus* from December 1966 to December 1970 and found 3 (0·07%) infective *Ae. polynesiensis* and no infective *Ae. samoanus*. These results showed that transmission had been reduced considerably by treatment with diethylcarbamazine citrate (DEC-C).

Subsequently, DESOWITZ & SOUTHGATE (1973) used the membrane filtration technique in Samoa on 61 originally infected adults treated with the full course of 72 mg/kg of DEC-C during the mass drug administration. They found 14 (23%) still positive, mostly with low densities undetectable by a 60mm^3 thick blood film. The situation in Samoa was the same when Shibuya (1977, unpublished WHO report) conducted a pilot survey using the Nuclepore filtration technique in 6 villages shortly before the commencement of the present project; in Vailu'utai he found a microfilaria prevalence rate of 5·7% and 11 of the 34 positive cases (32·4%) had counts of 10 mf/ml or less. These carriers were distributed evenly in the village and were awaiting treatment during the period of the present study.

The present records of vector infections are the first obtained 7 years after the second mass drug administration in 1971. 59·7% of the 62 infective *Ae. polynesiensis* from Vailu'utai had 1 to 2 larvae. This was consistent with the finding of 1 to 2 infective larvae in laboratory bred and wild caught *Ae. polynesiensis* infected by feeding on carriers with a low density range of 7 to 37 mf/ml (SAMARAWICKREMA *et al.*, 1985a). The natural infections with 1 to 2 larvae could have originated from feeding on the low density carriers in Vailu'utai. These results are at variance

with the findings of BRYAN & SOUTHGATE (1976) in Samoa who reported that wild caught *Ae. polynesiensis* when fed on carriers with very low microfilaraemias produced infections in greater proportion.

BURNETT (1960) estimated the mean daily mortality of *Ae. pseudoscutellaris, Ae. fijiensis* and *Culex quinquefasciatus* from the proportion of infected mosquitoes that survived to the infective stage. LAURENCE (1963) made similar estimates for *Cx quinquefasciatus* in Vellore, India. In the present study, out of 221 infected *Ae. polynesiensis*, 135 survived to stage II, a period of 8 to 10 days, and 72 survived to the infective stage, 12 to 14 days (SAMARAWICKREMA *et al.*, 1985b). The probability of survival, p, when $p^8 = 135/221$ and $p^{10} = 135/221$, was 0·940 and 0·952 with a mean value of 0·946. Similarly p, when $p^{12} = 72/221$ and $p^{14} = 72/221$, was 0·911 and 0·923 with a mean value of 0·917.

The original estimates of risk of infection were made by WHARTON (1962) on the intensity of transmission of *Brugia malayi* by *Mansonia* in Malaysia. More recent estimates for vectors such as *Cx quinquefasciatus* and *An. gambiae* in several endemic areas include those given by HAIRSTON & DE MEILLON (1968), ROZEBOOM *et al.* (1968), GUBLER & BHATTACHARYA (1974), WIJERS (1977), SELF *et al.* (1978), BUSHROD (1979) and MCMAHON *et al.* (1981). Comparison of these estimates can be meaningful if the sampling methods of vectors are standardized; such estimates are extremely difficult to determine accurately as infective mosquitoes are very few and larvae can be missed in dissection.

The infective biting rate and the transmission potential depend on the number of mosquitoes biting man, their longevity and the size and availability of the reservoir of microfilariae. Thus, in our study, Vailu'utai was a risk area with a very high transmission potential by *Ae. polynesiensis*. The corresponding transmission potential for Luatuanu'u was zero due to the absence of infective larvae from the samples of mosquitoes dissected. The index for *Ae. samoanus* in Luatuanu'u showed, however, that transmission occurred during the year. The high degree of transmission in Vailu'utai was also reflected in the transmission potential of the small population of *Ae. samoanus*. Only the indoor populations of *Ae. samoanus* were measured. Outdoor transmission by *Ae. samoanus* was considered of little significance as few people would be exposed to its bites in the bush at night. When measuring the risk of exposure to subperiodic *W. bancrofti* in Samoa throughout 24 h it is essential to combine results for daytime transmission by *Ae. polynesiensis* and for night-time transmission by *Ae. samoanus*.

Baseline data for the annual infective biting rate and the transmission potential were obtained for only the two coastal villages. 12-hour catches were not carried out in the inland villages. The results enable entomological evaluation of the control measures and it would be interesting to do this during and after treatment of the mf carriers and control of *Ae. polynesiensis* by selective larvicides, source reduction and environmental management using community participation.

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