MALARIA IN HONIARA, SOLOMON ISLANDS: VECTOR STUDIES

David Bell[1,2], Andrew Cameron[1], Mary Fernando[1], Khonesavanh Pholsyna[1], Desmond Foley[1], Bernard Bakote'e[3] and Joan H Bryan[1]

[1]Tropical Health Program, The University of Queensland Medical School, Herston Road, Herston, Australia Q 4006; [2]Present address: Katherine District Hospital, Katherine, Australia NT 0850; [3]Solomon Islands Medical Research and Training Institute, Honiara, Solomon Islands

Abstract. Adult and larval specimens of anopheline mosquitos were collected throughout eastern Honiara during a study into risk factors for malaria illness in adults. Species identification was by morphology, DNA probes and by PCR. Only *Anopheles farauti s.s.* were identified from part-night landing catches carried out from 1900 to 2200 hours. Most mosquitos attracted to humans were culicines. The majority of anophelines (85%) were captured between 1900 and 2000 hours. *An. farauti s.s.* larvae were most common but one *An. farauti* No. 7, and ten *An. punctulatus* larvae were also collected.

INTRODUCTION

Malaria is a serious public health problem in the Solomon Islands with cases being diagnosed throughout the year with peak incidence in 1994 between February and June. The annual incidence in Honiara, the capital of the Solomon Islands, has varied between 800/1,000 population to in excess of 1,000/1,000 population in the years 1992-1994 (Ministry of Health and Medical Services, 1995). Malaria control in Honiara is the responsibility of the Honiara Town Council. The control strategy aims to reduce morbidity and mortality by early diagnosis and treatment. Vector control to reduce transmission is by larviciding with temephos, by use of bednets impregnated with permethrin and the introduction of the mosquito-eating fish, *Gambusia affinis,* into water courses.

The vectors of malaria in the Solomon Islands are *Anopheles koliensis* Owen, *An. punctulatus* Dönitz and *An. farauti* Laveran *sensu lato (s.l.)* (Belkin, 1962). Following the classification proposed by Harbach (1994) these species all belong to the Punctulatus complex. Members of this complex can be differentiated from other species in the genus *Anopheles* which have been recorded from the Solomon Islands (*An. lungae* Belkin and Schlosser, *An. solomonis* Belkin, Knight and Rozeboom and *An. nataliae* Belkin) as the members of the Punctulatus complex are unique in having black scales on the knob of the halteres. *Anopheles farauti s.l.* consists of a number of morphologically indistinguishable species (Bryan, 1973; Mahon and Miethke, 1982; Foley *et al*, 1993, 1994, 1995) which have not yet been formally described but have been designated by number. *Anopheles farauti sensu stricto (s.s)*, formerly known as *An. farauti* No. 1, *An. farauti* No. 2 and No. 7 have been recorded from Guadalcanal (Foley *et al*, 1994). The revelation of additional species within the punctulatus complex and the variability of morphological features once considered diagnostic for individual species, have cast doubt on the previous species identifications in this group (Foley *et al*, 1993); the role of individual species in malaria transmission needs to be assessed.

The reasons for the high incidence of malaria in Honiara was investigated during a three month study from Januray 1995 (Bell *et al*, 1996). Patients from clinics in East Honiara from whom blood films were taken, were recruited into the study and interviewed to elucidate factors contributing to the risk of malaria parasitemia (Bell *et al*, 1996). Approximately half of these patients (120/309) were visited at home where factors which may influence their exposure to malaria were investigated. In this paper we report on the entomological findings of this study.

MATERIALS AND METHODS

The study site, the eastern half of Honiara, is illustrated in Fig 1. Larval searches were made throughout the study area and in particular in the vicinity of houses of patients recruited into the

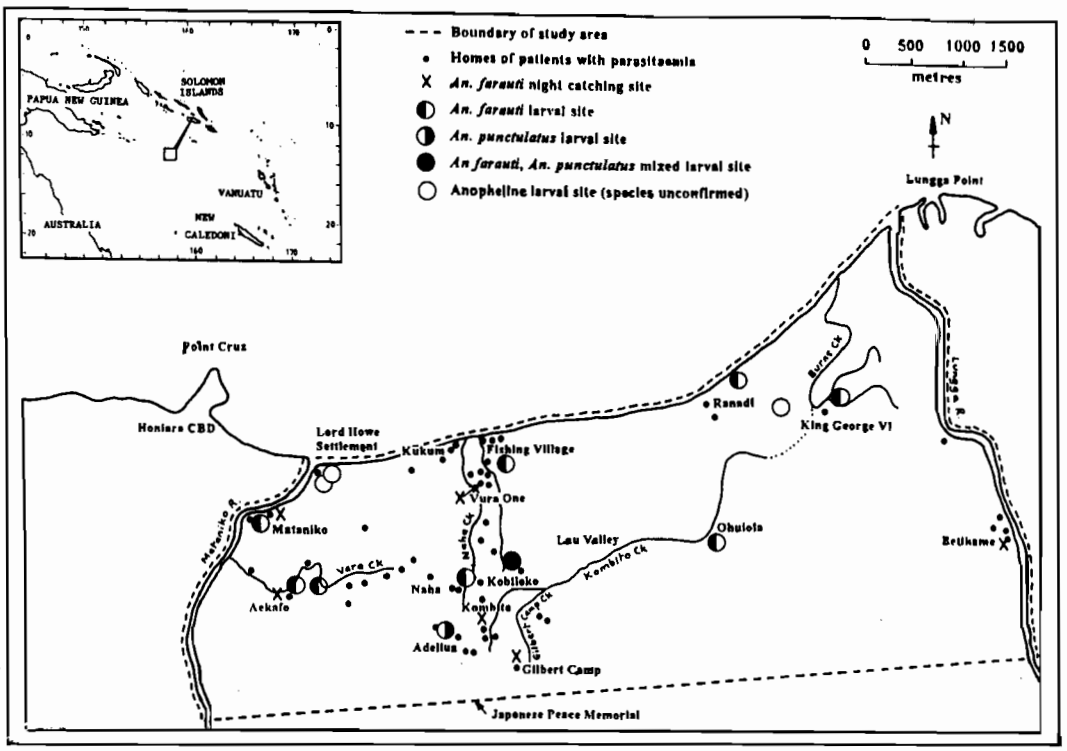

Fig 1–The study area, Honiara, Solomon Islands.

study. Anopheline larvae were collected and taken to an insectary where they were reared to the adult stage before being identified.

Part-night landing catches of mosquitos were carried out in areas with a high prevalence of malaria parasitemia cases. Two humans collected mosquitos attracted to themselves at 6 locations, Vura One School, Betikama School, Kombito Two, Gilbert Camp, Aekafo and Mataniko between 1900 (sunset) and 2230 hours. Larvae at Adeliua, developed into adults which conformed to the morphological description of *An. punctulatus*. At this site, night landing collections were made between 2200 and 2400 hours, as this is the peak biting period for this species (Lee *et al*, 1987).

A preliminary identification of adult mosquitos was made using the key in Belkin (1962). Specimens identified as *An. farauti s.l.* were preserved in 100% ethanol and identified in Brisbane using DNA probes (Beebe *et al*, 1994). For DNA probing, only the heads of specimens were squashed onto nylon filters and the remainder of the body was retained for future reference. Filters with heads were probed first for *An. farauti s.s.*. As this probe cross-reacts with *An. farauti* No. 7 (Beebe *et al*, 1996), filters were then stripped and probed for *An. farauti* No. 7. Specimens which were not identified by this process were probed for *An. farauti* No. 2. The remainder of specimens which reacted weakly or not at all with the probes were re-examined to confirm that they belonged to the *punctulatus* complex and then subjected to polymerase chain reaction (PCR) analysis of their internal transcribed spacer 2 (ITS-2) ribosomal DNA (rDNA). Digestion of the ITS-2 region of rDNA with the endonuclease MSP-1 gives species specific banding patterns. Details of this methods are given in Beebe and Saul (1995).

RESULTS

Mosquito collection sites are shown in Fig 1 and the results of mosquito collections in Tables 1 and 2.

All larval collections were within 2 km of the coast. *Anopheles punctulatus* was identified mor-

Table 1

Results of night landing catches undertaken in Honiara, Solomon Islands in March 1995.

Location (Date)	Species	Catches per time periods			*An. farauti s.s.*	
		1900-2000	2000-2100	2100-2200	By DNA probe	By PCR
Vura One	*Anopheles**	4	3	0	6	
(21.3.95)	culicines	79	15	13		
Betikama	*Anopheles*#*	63	4	6	65	4
(21.3.95)	culicines	68	14	9		
Gilbert Camp	*Anopheles*	5	1	1	7	
(27.3.95)	culicines	68	13	13		
Kombito Two	*Anopheles*	1	0	0	1	
(27.3.95)	culicines	14	7	13		
Aekafo	*Anopheles*	1	0	0	1	
(29.3.95)	culicines	4	4	1		
Mataniko	*Anopheles*	11	1	0	12	
(29.3.95)	culicines	25	26	13		
Totals	*Anopheles*	85	9	7	92	4
	culicines	228	79	62		

* 1 specimen not identified
\# 4 specimens not identified

Table 2

Identification of specimens collected as larvae in Honiara, January-March 1995.

Place	Identification		
	By DNA probe	By PCR	By morphology
Matariu	1♀ *An. farauti s.s.*	1♂ *An. farauti* No. 7	
	1♂ *An. farauti s.s.*		
Gwaimoa	2♀ *An. farauti s.s.*	1♂ *An. farauti s.s.*	
Rice Farm	40♀ *An. farauti s.s.*	1♀ *An. farauti s.s.*	
	32♂ *An. farauti s.s.*	1♂ *An. farauti s.s.*	
Ranadi (Brackish)	7♀ *An. farauti s.s.*		
	4♂ *An. farauti s.s.*		
SICHE* Kukum	12♀ *An. farauti s.s.*		
	4♂ *An. farauti s.s.*		
Naha	1♀ *An. farauti s.s.*	1♂ *An. punctulatus*	
	1♂ *An. farauti s.s.*		
Mataniko	1♀ *An. farauti s.s.*		
Adeliua		3♂ *An. punctulatus*	
Kombito	2♂ *An. farauti s.s.*		3♀ *An. punctulatus*
			4♂ *An. punctulatus*
Ohiuola	3♀ *An. farauti s.s.*		
	2♂ *An. farauti s.s.*		

* Solomon Island College of Higher Education

phologically from the 2 most inland sites, Mambulu Road (1.5 km from the coast) and Adeliua (2 km inland) and this identification was confirmed by PCR for one male specimen from Adeliua. One adult specimen reared from a larva collected at Naha was also identified by PCR as *An. punctulatus*. The Mambulu Road and Adeliua larval habitats were the most elevated at 55 m and 60 m above sea level respectively, exposed to sunlight throughout the day, and had clear water with emergent vegetation.

Anopheles farauti s.s. larval habitats were more varied, and included temporary pools, rice fields and backwaters of a creek. All habitats were predominantly sunlit; only Vara Creek had some shade. The Ranadi site was brackish, and the larvae in that site were darker than those from other sites. Similar dark larvae were collected from other brackish habitats outside the study area and these were also *An. farauti s.s.* (unpublished data). Vara Creek was the only creek where *G. affinis* were not seen, and the only creek in which anopheline larvae were found (at Gwaimaoa and Matariu). Apart from the Vara Creek and Ohuiola sites, all had clear water with emergent vegetation. The larvae at Ohuiola were in cattle hoof prints containing cloudy water contaminated with urine.

Anopheline mosquitos were not obtained in the collection at Adeliua from 2200 to 2400 hours. Two *An. farauti s.s.* were attracted to humans at Kukum at night.

All the anopheline mosquitos collected at human bait were *An. farauti s.l.* Of the 101 subjected to DNA probing, 92 were identified as *An. farauti s.s.*; an additional 4 specimens were identifed by PCR as *An. farauti s.s.* and 5 specimens could not be identified due to technical problems. The majority (57%, 86%, 71%, 92%, 100%) of the anophelines in the night landing collections were obtained in the first hour after sunset. At 4 sites, culicine mosquitos comprised 90% or more of the total catch. The highest proportion of anophelines was obtained at Betikama where they constituted 43.5% of the total collection.

DISCUSSION

Anopheles farauti s.s. larval habitats were present throughout the study area, extending from within a few metres of the coast to 1.5 km inland and from sea level to 55 m above sea level. Habitats of *An. punctulatus* were much more restricted being found in only three areas, all of which were more than 1 km from the coast and two sites were 50 m or more above sea level. This species has not been recorded in Honiara for a number of years and its re-appearance may be associated with the withdrawal of residual spraying with DDT. Since *An. punctulatus* is anthropophilic, endophilic and feeds during the middle of the night when most inhabitants have retired to bed, it is vulnerable to DDT spraying (Lee *et al*, 1987; Samawickrema *et al*, 1992) and impregnated bed nets (Bakote'e and Arabola, 1992) from entering houses to feed on its preferred host.

Anopheline larvae were not detected in either of the rivers in the study area or in a number of streams such as Vura, Gilbert Camp and Kombito Creeks. The mosquito fish, *G. affinis*, was abundant in these streams and may account for the absence of larvae. Most of the larval sites were temporary pools which would not support fish and would be difficult and expensive for the council to treat regularly with insecticide.

Larval habitats were similar to descriptions given by other workers (Belkin, 1962; Lee *et al*, 1987). Larvae were absent from heavily shaded areas and planting shade trees should reduce larval habitats. Contrarily, the often cited need to keep areas clean, leading in extreme cases to tree felling to reduce mosquitos, will encourage breeding of this species.

High densities of *An. farauti s.s.* larvae occurred in the newly established experimental rice farm in Honiara. Residential areas are sited within the flight range of this species (Charlwood *et al*, 1986). Future development of this potentially important economic activity needs to take the malaria risk into account.

The majority of anopheline specimens attracted to human bait in Honiara were *An. farauti s.s.* This is not unexpected as the majority of anopheline larvae in the study area where of this species. Although only a few landing collections were undertaken for part of the night, and the density of this species was not high, overall 84% of the specimens were obtained in the first hour after sunset. A biting pattern with the majority of specimens biting early in the evening has previously been described for the Solomon Islands (Taylor, 1975; Webber and

Southgate, 1981). This biting pattern is thought to be the result of DDT residual spraying; it allows mosquitos to feed without having to enter sprayed houses. Early evening biting enables *An. farauti* to feed readily on humans who are often out of doors at this time. Contact between exophilic vectors and people will not be inhibited by indoor spraying of residual insecticide or by the use of bednets impregnated with insecticides.

Because of difficulties in reducing contact between adult vectors and humans, control of larval habitats may be the only way of reducing malaria in this environment. As many of the habitats are small transient water collections, control will be beyond the capabilities of government alone and community members will need to be involved. Such participation could involve training key people to recognize anopheline larvae, allowing better targeting of small scale larviciding or drainage. The low flight range of *An. farauti s.l.* (Charlwood *et al*, 1986) and evidence of clustering of cases close to potential larval habitats in Honiara (Bell *et al*, in press) suggests that such a strategy may provide benefit to communities adopting such a strategy.

The majority (78.5%) of mosquitos attracted to humans were culicines. If communities are to be encouraged to destroy anopheline larval habitats as part of a program to involve residents in malaria control, they must be taught that anophelines could be eliminated without a perceivable reduction in mosquito biting nuisance.

ACKNOWLEDGEMENTS

This paper has been approved by the Solomon Islands Ministry of Health and Medical Services.

REFERENCES

Bakote'e B, Arabola A. Permethrin-treated mosquito nets: the Solomon Islands experience. Presented at the eleventh South-west Pacific Malaria Meeting. Port Vila, Vanuatu, 1992.

Beebe N, Foley DH, Saul A, Cooper L, Bryan JH, Burkot TR. DNA probes for identifying members of the *Anopheles punctulatus* complex in Papua New Guinea. *Am J Trop Med Hyg* 1994; 50 : 229-34.

Beebe NW, Foley DH, Cooper RD, Bryan JH, Saul A. DNA probes for the *Anopheles punctulatus* complex. *Am J Trop Med Hyg*, 1996; 54 : 395-8.

Beebe NW, Saul A. Discrimination of all members of the *Anopheles punctulatus* complex by polymerase chain reaction - restriction fragment length polymorphism analysis. *Am J Trop Med Hyg*, 1996; 53 : 478-81.

Belkin JN. The mosquitoes of the South Pacific (Diptera, Culicidae). Vols. 1 and 2. Berkeley and Los Angeles : University of California Press, 1962.

Bell D, Bryan JH, Cameron A, Fernando M, Le'afasia J, Pholsyna K. Malaria in Honiara, Solomon Islands : reasons for presentation and human and environmental factors influencing prevalence. *Southeast Asian J Trop Med Public Health*, 1996.

Bryan JH. Studies on the *Anopheles punctulatus* complex. Identification by proboscis morphological criteria and by cross-mating experiments. *Trans R Soc Trop Med Hyg* 1973; 67 : 64-9.

Charlwood JD, Graves PM, Birley M. Capture - recapture studies with mosquitoes of the group of *Anopheles punctulatus* Dönitz (Diptera : Culicidae) from Papua New Guinea. *Bull Ent Res* 1986; 76 : 211-27.

Foley DH, Paru R, Dagoro H, Bryan JH. Allozyme analysis reveals six species within the *Anopheles punctulatus* complex of mosquitoes in Papua New Guinea. *Med Vet Entomol* 1993; 7 : 37-48.

Foley DH, Meek SR, Bryan JH. The *Anopheles punctulatus* group of mosquitoes in the Solomon Islands and Vanuatu surveyed by allozyme electrophoresis. *Med Vet Entomol* 1994; 8 : 340-50.

Foley DH, Cooper RD, Bryan JH. A new species within the *Anopheles punctulatus* complex in Western Province, Papua New Guinea. *J Am Mosq Contr Assoc* 1995; 11 : 122-127.

Harbach RE. Review of the internal classification of the genus *Anopheles* (Diptera : Culicidae) : the foundation for comparative systematics and phylogenetic research. *Bull Ent Res* 1994; 84 : 331-342.

Lee DJ, Hicks MM, Griffiths M, *et al*. The Culicidae of the Australasian Region Vol 5. Canberra : Australian Government Publishing Service, 1987.

Mahon RJ, Miethke PM. *Anopheles farauti* No. 3, a hitherto unrecognized biological species of mosquito within the taxon *An. farauti* Laveran (Diptera : Culicidae). *Trans R Soc Trop Med Hyg* 1982; 76 : 8-12.

Ministry of Health and Medical Services. National malaria control program. Annual report 1994. Honiara: Ministry of Health and Medical Services, 1995.

Samawickrema WA, Parkinson AD, Kere N, Galo O. Seasonal abundance and biting behaviour of *Anopheles punctulatus* and *An. koliensis* in Malaita Province, Solomon Islands, and a trial of permethrin-impregnated bednets against malaria transmission. *Med Vet Entomol* 1992; 6 : 371-78.

Taylor B. Changes in the feeding behaviour of a malaria vector, *Anopheles farauti* Lav, following use of DDT as a residual spray in houses in the British Solomon Islands Protectorate. *Trans R Soc Trop Med Hyg* 1975; 127 : 277-92.

Webber RH, Southgate BA. The maximum density of anophelines that can be permitted in the absence of continuing transmission of filariasis. *Trans R Soc Trop Med Hyg* 1981; 75 : 499-506.