Medical and Veterinary Entomology (2002) **16**, 116–119

SHORT COMMUNICATION

Impact of untreated bednets on prevalence of *Wuchereria bancrofti* transmitted by *Anopheles farauti* in Papua New Guinea

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Abstract. Despite the growing evidence that insecticide-treated mosquito nets reduce malaria morbidity and mortality in a variety of epidemiological conditions, their value against lymphatic filariasis infection and disease is yet to be established. The impact of untreated bednets on the prevalence of *Wuchereria bancrofti* (Cobbold) (Nematoda: Filarioidea) infection and disease was investigated on Bagabag island in Papua New Guinea, where both malaria and filariasis are transmitted by the same vector mosquitoes of the *Anopheles punctulatus* Dönitz group (Diptera: Culicidae). Community-wide surveys were conducted recording demographic characteristics including bednet usage. Physical examinations for hydrocoele and lymphoedema were performed and blood samples assessed for filarial and malaria parasites. Mosquitoes were sampled using the all-night landing catch method and individually dissected to determine *W. bancrofti* infection and infective rates. Bednet usage among residents was 61% and the mean age of users (25.6 years) was similar to non-users (22.5 years). *Anopheles farauti* Laveran was the only species were found to contain filarial larvae: 2.7% infected (all stages), 0.5% infective (L3). The overall *W. bancrofti* microfilaraemia and antigenaemia rates were 28.5% and 53.1%, respectively. Bednet users had lower prevalence of *W. bancrofti* microfilaraemia, antigenaemia and hydrocoele rates than non-users. In comparison, untreated bednets had no effect on the prevalence and intensity of *Plasmodium falciparum* and *P. vivax* infections. The impact of bednet usage on rates of microfilaraemia and antigenaemia remained significant even when confounding factors such as age, location and sex were taken into account, suggesting that untreated bednets protect against *W. bancrofti* infection.

Key words. *Anopheles farauti*, *An. punctulatus*, *Culex annulirostris*, *Cx. quinquefasciatus*, *Plasmodium falciparum*, *P. vivax*, *Wuchereria bancrofti*, bednets, filariasis control, filariasis vectors, malaria, personal protection, vector control, Papua New Guinea.

The discovery by Manson (1884) that certain species of mosquitoes are obligate vectors of *Wuchereria bancrofti*, causing lymphatic filariasis, was the first time that any insect had been implicated in the active transmission of human or animal disease (Service, 1978). This gave rise to hopes for new, possibly easy, ways of controlling

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mosquito-borne diseases by reducing human–vector contact. Filariasis vector control is particularly attractive because transmission of the parasite is very inefficient. There is no multiplication of the parasite in the mosquito vector and only continuous exposure to bites of many infected mosquitoes maintains the infection in humans. Among the available vector control measures, protection from mosquito bites by bednets has received a lot of attention because it is already a familiar concept that is effective and simple to implement. Despite the growing evidence that insecticide-treated mosquito nets reduce malaria morbidity and mortality in various epidemiological conditions (Choi *et al*., 1995), their value against lymphatic filariasis infection and disease is yet to be established (Ottesen *et al*., 1997).

Malaria and filariasis are the most common mosquito-borne parasitic infections in Papua New Guinea (PNG) and both diseases are transmitted mainly by members of the *Anopheles punctulatus* group of mosquitoes. Recent analysis of the impact of untreated bednets in PNG showed that protracted use can result in profound changes in malaria endemicity, even when coverage is incomplete (Smith *et al*., 2001). One would expect reduction in mosquito–human contact to be more effective against the transmission of lymphatic filariasis than malaria, as more infective bites are required to produce a patent infection of *W. bancrofti* (Southgate, 1992). Whereas malaria transmission usually results from one infective mosquito bite, Hairston & De Meillon (1968) estimated that approximately 15 500 infective bites/person are required to produce a single patent infection with *W. bancrofti*. The present study assesses the impact of untreated bednets on prevalence of *W. bancrofti* infection and disease in the human population of Bagabag Island, PNG, where people might have benefited from their habitual use of bednets reducing transmission rates of *W. bancrofti*.

Community-wide surveys were conducted on Bagabag Island (4°47′ S, 146°14′ E), a relatively isolated island with a population of about 1500 people living in three villages: Badilu, Matiu 1 and Matiu 2. Bagabag island is situated in the Pacific Ocean, about 65 km NE from Madang town on the coast of PNG Madang Province. Epidemiological surveys were conducted in all households on the island, recording demographic characteristics, including bednet usage for the previous night. Bednets had been used habitually by residents of Bagabag Island for several years prior to this study. Individuals who had been interviewed during the day were observed by field staff during the night, to assess the discrepancy between reported and true usage of bednets.

Venous blood (5 ml) samples were collected from all consenting individuals and assessed for filarial and malaria parasites. Microfilaraemia of nocturnally periodic *W. bancrofti* was quantified from 1 ml blood samples obtained between 22.00 and 02.00 hours. Nuclepore® filtration and counting of microfilariae (mff) were performed as described by Desowitz & Hitchcock (1974) and results were expressed as mff/ml of blood. ELISAs were performed to determine prevalence and intensity of antigenaemia as instructed by the manufacturer of the Og4C3 ELISA kit (TropBio, JCU Tropical Biotechnology Pty Ltd, Townsville, Queensland, Australia). Physical examinations for hydrocoele and lymphoedema of the lower extremities was performed according to protocols described by the World Health Organization (WHO, 1984). Non-pitting lymphoedema that is not reversible upon elevation of the leg (grade II) was distinguished from gross increase in leg volume with dermatosclerosis and papillomatous changes (grade III). Lymphoedema of the upper extremities was not observed. Malaria blood slides were stained with 4% Giemsa and examined for 100 microscopic thick film fields under oil immersion prior to being declared negative. Densities were recorded as the number of parasites/200 white blood cells (WBC). These densities were then converted to the number of parasites/μl blood, assuming 8000 WBC/μl blood. Human-biting female mosquitoes were sampled by all-night landing catch, as described elsewhere (Bockarie *et al*., 1996). Mosquitoes were stained and individually dissected to determine *W. bancrofti* infection and infective rates following the procedures of Nelson (1958).

Bednet usage

A total of 1073 individuals consented to participate in the survey and were interviewed: 646 (60.2%) reported that they had used a bednet the previous night. The number of years of bednet usage by users ranged from one 1.5–17 (mean = 7 years). Ten people with bednets not long enough to be tucked under the mattress, badly damaged nets or those with five or more finger-size holes/net were excluded from the analysis. There was no discrepancy between the bednet usage observed by the survey team and that reported by the subjects themselves. Of those interviewed, 1041 were classified as residents who had always lived on the island. Bednet usage reported by residents was 60.6% overall, varying between the three villages: 38.3%, 66.8% and 74.1% for Badilu, Matiu 1 and Matiu 2, respectively. The mean age of users (25.6 years) was similar to non-users (22.5 years). Bednet usage among males (46.1%) and females (53.9%) was significantly different ($\chi 2 = 7.6$, d.f. = 1, $P = 0.006$).

Vectors of filariasis

Entomological surveys undertaken at the time of the survey yielded a total of 996 anthropophilic mosquitoes, including 400 (40%) *Anopheles farauti*, 14 (1.4%) *An. punctulatus*, 4 (0.4%) *Culex annulirostris* and 578 (58%) *Cx. quinquefasciatus*. *Anopheles farauti* (a member of the *An. punctulatus* group) was the only species found to contain filarial larvae, with infection and infective rates of 2.7% and 0.5%, respectively.

Bednets and human infection

Overall *W. bancrofti* microfilaraemia and antigenaemia rates were 28.5% and 53.1%, respectively. Bednet users had a significantly lower microfilaria rate, antigen positivity

Table 1. Impact of untreated bednets on the prevalence of *Wuchereria bancrofti* microfilaraemia (Mf) and disease, and *Plasmodium falciparum* (PF) and *P. vivax* (PV) infection rates on Bagabag Island, Madang Province, Papua New Guinea.

	Bednet users	Non-users	Odds Ratio (95% C.I)	*P*
Infection rates				
Proportion (%) with Mf	154/620 (24.8)	138/406 (34.0)	0.68 (0.50–0.92)	0.013
Proportion (%) antigen positive	296/622 (47.6)	251/408 (61.5)	0.61 (0.46–0.81)	0.001
Proportion (%) with PF	127/635 (20.0)	81/425 (19.1)	1.10 (0.78–1.54)	0.575
Proportion (%) with PV	112/635 (17.6)	69/425 (16.2)	1.08 (0.76–1.55)	0.655
Disease rates (20 + years)				
Proportion (%) with lymphoedema	8/309 (2.6)	5/234 (2.1)	1.23 (0.38–4.02)	0.721
Proportion (%) with hydrocoele	3/140 (2.1)	9/131 (6.9)	0.36 (0.09–1.39)	0.138

rate and intensity of antigenaemia than non-users (Table 1). These effects remained significant when adjustments were made for age, sex and village using a logistic regression analysis (see Table 1). Geometric mean intensity of microfilaraemia was lower for bednet users (4.1 mf/ml) than non-users (7.7 mf/ml), although this difference was not significant ($P = 0.11$). In contrast, the prevalence and intensity of infection with malaria parasites, *Plasmodium falciparum* and *P. vivax*, were similar for bednet users and non-users.

Prevalence of filarial disease in the form of hydrocoele, and lymphoedema of the extremities was assessed for people aged 20 years and older. The proportion of people with lymphoedema was similar for bednet users (2.6%) and non-users (2.1%). The hydrocoele rate for non-users (6.9% of 131) was three times higher than users (2.1% of 140), but this difference was not statistically significant (Table 1).

Thus, we found that bednet users on Bagabag Island had lower rates of *W. bancrofti* microfilaraemia, antigenaemia and hydrocoele than non-users. The impact of bednet usage on filarial infection remained significant even when confounding factors such as age, location and sex were taken into account, suggesting that untreated bednets protect against *W. bancrofti* infection. Untreated bednets, however, had no effect on the prevalence and intensity of *P. falciparum* and *P. vivax* infections on the island. It has been shown previously that when filariasis and malaria transmission depend upon the same *Anopheles* species, vector control often results in a significant reduction in filariasis transmission intensity even if malaria transmission continues to be a problem (Webber, 1975, 1977, 1979). Although observational studies in the Gambia (Campbell *et al*., 1987; Clarke *et al*., 2001) and Papua New Guinea (Genton *et al*., 1994) have suggested that untreated bednets also protect against *P. falciparum* infection, two randomised control trials found no significant effects on malaria incidence (Snow *et al*., 1988; Maxwell *et al*., 1999).

In a previous study, the introduction of untreated bednets diverted *An. punctulatus* females away from humans in a PNG village, significantly reducing vector–human contact and the human blood index (HBI), with concomitant five-fold rise in the rate of mosquitoes feeding on dogs (Burkot *et al*., 1990). The drop in HBI led to a significant decrease in the infection rates for *P. falciparum* and *W. bancrofti*; however, the drop in the *P. falciparum* sporozoite rate was insufficient to affect parasite rates in the human population. Analysis of the impact of untreated bednets on malaria in the nearby East Sepik Province suggest that if coverage is high, long-term use of untreated bednets may have substantial impact on disease prevalence, more because of effects on vector bionomics than of individual protection (Genton *et al*., 1994; Hii *et al*., 2001). Also, Smith *et al*. (2001) determined that protracted use of untreated bednets in PNG reduced the endemicity of malaria transmitted by the *An. punctulatus* complex, even when coverage is incomplete.

Treating bednets with residual pyrethroid insecticides can significantly reduce vector density and survival rates. The entomological impact of using insecticide-treated bednets on vectors of malaria and filariasis has been studied also in Africa. In Kenya, permethrin-impregnated bednets reduced the number of indoor-resting *Anopheles* mosquitoes by over 94% but there was no change in the number of *Cx. quinquefasciatus* (Bøgh *et al*., 1998). In Tanzania, permethrin-impregnated bednets reduced the entomological inoculation rate by 90% (Magesa *et al*., 1991). In previous studies in Madang villages of PNG, permethrin-impregnated bednets resulted in a significant 67% drop in sporozoite rate for *An. punctulatus*, with a significant reduction in *P. falciparum* incidence in young children (Graves *et al*., 1987).

Studies comparing the impact of permethrin-impregnated bednets and DDT house-spraying against malaria transmission in the Solomon Islands have shown the former to be more effective (Hii *et al*., 1993; Kere *et al*., 1996). This suggests that treated bednets may be more effective against filariasis than malaria in PNG, where both infections are transmitted by the *An. punctulatus* group of mosquitoes. In the Solomon Islands, DDT house-spraying operations of insufficient efficiency to interrupt transmission of malaria led to the eradication of lymphatic filariasis transmitted by the *An. punctulatus* group of mosquitoes (Webber, 1975, 1977, 1979). Likewise, the use of bednets, even if not treated with insecticides, apparently has relatively more impact on filariasis than malaria infections rates where *Anopheles* vectors transmit both *W. bancrofti* and *Plasmodium*.

Acknowledgements

We thank the Bagabag people who generously agreed to participate in this study, the staff of the Lutheran Health Centre on Bagabag Island, and the field and laboratory

staff of the Papua New Guinea Institute of Medical Research for their dedicated support in this project. The project was funded by WHO/CTD grant no. CTD/FIL/F3/181/145.

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Accepted 12 December 2001