RISK FACTORS FOR MALARIA INFECTION AMONG ETHNIC MINORITIES IN BINH PHUOC, VIETNAM

Tomoko Abe¹, Sumihisa Honda², Shusuke Nakazawa³, Trinh Dinh Tuong⁴, Nguyen Quang Thieu⁴, Le Xuan Hung⁴, Le Khanh Thuan⁴, Kazuhiko Moji⁵, Masahiro Takagi¹ and Taro Yamamoto²

¹Department of Medical Entomology, ²Department of International Health, ³Department of Protozoology, Institute of Tropical Medicine, Nagasaki University, Japan; ⁴National Institute for Malariology, Parasitology, and Entomology, Hanoi, Vietnam; ⁵Research Institute for Humanity and Nature, Kyoto, Japan

Abstract. A cross-sectional study was undertaken to identify the prevalence of malaria infection, behavioral patterns of inhabitants and risk factors for malaria infection in a minority village in an area endemic for malaria in Vietnam. The target population were all the inhabitants of a village in Binh Phuoc Province, Vietnam. Interviews using a structured questionnaire and blood examination for malaria infection were conducted in house-to-house visits. Relationships between malaria infection and variables were examined using uni- and multivariate adjusted analysis. A total of 682 individuals from 159 households participated in both the interview and blood examinations. All households earned income through farming without forest activities at night, and the socio-economic status was generally not very low. The total prevalence of malaria infection was 6.2%, with a peak among 3- to 5-year-old children. Univariate analysis identified 3 - 5 year olds, a family size of \geq 5 people, sleeping with >3 people in a bed, and living in a wooden/bamboo house as factors associated with malaria infection. Multivariate adjusted analysis after variable selection identified age 3 - 5 years old, a family size of ≥5 people and living in a wooden/bamboo house were significantly related to malaria infection. Malaria in this area can be controlled by basic activities, such as early diagnosis and treatment and prevention using bednets, since risk factors for malaria infection did not include forest activities, but were young age, living in a wooden/bamboo house and belonging to a large family. Continuous and intensive expansion of existing malaria control activities are required.

INTRODUCTION

In Vietnam, malaria was highly endemic during the late 1980s and early 1990s (Hung *et al*, 2002). Since the introduction of a national malaria control program in 1992, malaria mortality and morbidity were reduced

Correspondence: Tomoko Abe, Institute of Tropical Medicine, Nagasaki University, 1-12-4, Sakamoto, Nagasaki 852-8523, Japan. Tel/Fax: ++81 95 8197808 E-mail: aamnabibi@gmail.com by 97.3% and 77%, respectively, from 1991 to 2003 (Hung *et al*, 2002; Erhart *et al*, 2004, 2005; Thuan, 2005). National malaria control activities, such as early diagnosis and treatment, bednet distribution, insecticide treated bednets, and indoor-residual spraying have led to great improvements in malaria control (Nam *et al*, 2005; Thuan, 2005; Hung, 2008). In spite of this successful record, malaria remains a threat to health in some regions, mainly mountainous, remote and forested areas (Erhart *et al*, 2004,2005; The Khanh Phu Malaria Research Project, 2004; Thang et al, 2008). The National Malaria Control Program has identified some problems to expanding control activities to such areas (Thuan, 2005). First, these endemic areas are geographically difficult to access, and health facilities and personnel are limited compared to the large and widespread population. Different living environments and behavioral patterns in the local populations represent another source of difficulties. Erhart et al (2004, 2005) described a strong association between malaria infection and forest activities of ethnic minorities, as one of the vector mosquitoes in this area, Anopheles dirus, is extremely anthropophilic and shows a distribution characterized by forested foothills, forests and forest fringes (Trung et al, 2004, 2005; Obsomer et al, 2007).

Binh Phuoc Province is one of the remaining areas endemic for malaria in Vietnam, with the third-highest number of confirmed malaria cases in the country in 2006 [National Institute of Malariology, Parasitology and Entomology (NIMPE), Vietnam, 2006, unpublished data)]. This province includes the mountainous area bordering Cambodia and 19% of the total population are from a minority background. In terms of nationwide development, economic and environmental situations have been changing (NIMPE, 1996; Thuan, 2005). First, rapid deforestation has occurred during the last two decades and forest density has declined (Meyfroidt et al, 2008). As Yasuoka et al (2007) reported, deforestation has had an impact on the ecology of anopheline mosquitoes, their density, and malaria epidemiology. Vector mosquito species have changed with large-scale ecological changes (Klinkenberg et al, 2004). In the areas endemic for malaria in Vietnam, forest activities at night have been reported as a clear risk factor for malaria infection (Erhart et al, 2004, 2005). However, in Binh Phuoc Province, along with

deforestation, some minority groups are becoming farmers not having forest activities. In spite of this, malaria remains endemic. Malaria threatens not only local populations, but also migrants from non-endemic areas. Furthermore, most endemic areas are located along the borders, therefore, malaria control in these areas is important for Vietnam and its neighboring countries.

We carried out a cross-sectional study to identify malaria infection prevalence, behavioral patterns of the inhabitants and risk factors for malaria infection in a minority village in a malaria-endemic area of Vietnam.

MATERIALS AND METHODS

Study site and study population

The study was conducted between June and August 2006, following a pilot study conducted from September to October 2005, in Phu Thuan Village, Phu Rieng Commune, Phuoc Long District, Binh Phuoc Province, Vietnam (Fig 1). The climate in this area is a combination of tropical monsoon and dry weather. The rainy season typically extends from June to October, with the dry season from November to May. The monthly mean temperature was 25-28°C with an annual mean of 24.5°C. The average monthly rain fall was 224 mm with humidity ranging between 70% and 90%. In central and southern Vietnam, the main malaria vector species are An. minimus and An. dirus (The Khanh Phu Malaria Research Project, 2004; Trung et al, 2004, 2005; Obsomer et al, 2007). At the site of the present study, An. dirus is considered to be the main vector because of a low density of An. minimus (unpublished data). The study village, Phu Thuan, is in a hillside area surrounded by small streams and covered with rubber trees and cashew nut orchards. Little natural forest remains and is 15 km from the village. The village is



Fig 1-Location of the study site.

mainly populated by Stieng, an ethnic minority group. Stieng people migrated to this village around 1985 and cleared the natural forest to establish new fields for farming. Dry rice was originally harvested using slash-and-burn methods, but after settlement they began planting cashew trees around 1990. According to the village elders, the village was extremely poor before 2000, but has become relatively prosperous thanks to cashew nut harvesting. Although the national malaria control program has spread throughout the country, the villagers could not access medical treatment due to financial and geographical difficulties, so many villagers, including small children, have died from "high fever". The construction of a bridge and wide road to the town in 2002 and continued economic progress have enabled immediate access to malaria treatment and the health condition of the inhabitants

has improved greatly.

Household questionnaire survey and observation

Interviews using structured questionnaires were administered by the research team to a representative person from each household, typically the head of the family or their spouse. Family members were confirmed with the village census book, in which individuals residing in the village for more than 3 months were registered.

The questionnaire was comprised of three levels: individual, bed, and household. Questions concerning age, sex, level of education, ethnicity, frequency of overnight stays in the field, activities after dinner, frequency of bednet use, and time of entering the bednet were determined at the individual level. The household level included information regarding socio-economic status, such as income, ownership of property, dwelling material, and number of family members. Type of a bed, possession of bednet, history of insecticide treatment of the bednet, total area of holes in bednet and number of individuals sleeping in a bed were included in the bed level.

The material used to construct dwelling walls (cement, brick, wood or bamboo), bed type (bed, mat or hammock) and condition of bednets were confirmed by direct observation with a structured checklist. Each bednet was inspected for size, material, and number and size of holes. All bednet holes were counted and measured for size. Holes repaired properly with rubber bands or threads were not counted.

Laboratory methods and case definition

Blood slides (thick film) were made by finger prick and examining the same day by microscopy by an expert from the provincial malaria control center. The microscopist was blinded to the interview results. Body temperatures were taken at the same time. The thick film was stained with 3% Giemsa solution for 30 minutes, then the number of parasites was counted. Slides were examined at 1,000x magnification, and diagnosed as negative when an examination of 100 thick film fields failed to reveal any parasites. All individuals with slides showing positive results for either *Plasmodium falciparum* or *P. vivax* were defined as malaria-infected cases. regardless of symptoms. Malaria-infected individuals were treated with an artemisininbased combination drug for *P. falciparum* and chloroquine and primaquine for *P. vivax* in accordance with Vietnamese national guidelines (The National Malaria Control Project, 2003).

Statistical analysis

Data were entered in the field and checked by the authors. Individuals with missing microscopic results or interview results were excluded from analysis. Associations between malaria infection and risk factors were analyzed using the chi-square test. Crude odds ratios (ORs) and 95% confidence intervals (95%CIs) were calculated. Simultaneous effects of risk factors on malaria infection were analyzed using a linear logistic model. Starting with a full model including all variables, the most appropriate model was selected on the basis of Akaike's Information Criterion (AIC). Variables that did not improve the fit of regression (as measured by AIC) were omitted. Adjusted ORs and *p*-values were calculated after determining the most appropriate model. STATA version 9 statistical software (STATA, College Station. Texas) was used for all calculations.

Ethical considerations

This study was approved by the ethics committees of the Institute of Tropical Medicine, Nagasaki University, Japan (Approval No. 02070502), and NIMPE, Vietnam. Interviews, observations and blood examinations were only conducted after the purpose of the study had been explained to participants, who were given the right to withdraw from the study at any time, without consequences. Written informed consent was obtained from each household representative. Free malaria treatment was provided to all slide-positive individuals.

RESULTS

Characteristics of the study population

All inhabitants in the village were recruited for this study. Of the 790 inhabitants, 108 were excluded: 78 refused the blood test and 30 could not be reached on three separate visits. In total, 682 individuals from 159 households who agreed with both blood examination and interview were included in the study (Table 1).

Ethnicity was Stieng in 80.9% of subjects, with the remaining 20% mainly being Kinh. The sex ratio was nearly 1:1. About half the adults had never attended school, but all could communicate in Vietnamese. All households earned their main income by farming cashew nuts. None of the subjects were regular forest workers who stayed or slept outside at night, because they had their fields near their residences. Most households were comprised a nuclear family, with relatives usually living as neighbors. The average family size was 4.8.

The socio-economic status was generally not particularly low, due to the success of the cashew nuts business. The average monthly income was 78 US dollars (USD), and 13.8% of households earned >150 USD/ month. Reflecting this economic situation, 41 houses (25.8%) were built from cement, while 98 houses (61.6%) were traditional wood or bamboo structures. Ninety percent of houses used iron sheet roofing; the eaves were widely opened. No public transportation was available, but 87 households (83.6%) had at least 1 motorbike, enabling

characteristics of the study po	pui		
Variables	n (%)		
Individual level (<i>N</i> =682)			
Age (years)			
0-2	64	(9.4)	
3-5	69	(10.1)	
6-9	59	(8.7)	
10-19	156	(22.9)	
20-29	130	(19.1)	
≥30	204	(29.9)	
Sex			
Male	327	(48.0)	
Female	355	(52.0)	
Ethnicity			
Stieng	552	(80.9)	
Kinh/Other	130	(19.1)	
School education (Age \geq 15 years,	n=39	8)	
No school education	190	(47.7)	
Some primary	142	(35.7)	
Complete primary/higher	66	(16.6)	
Sleep outside at night		(_ = = = =)	
Never	620	(90.9)	
Only in harvest season	62	(9.1)	
Regularly	0	(0,1)	
Activities after dinner	Ū	(0.0)	
Stav in the house	481	(70.5)	
Stay outside (around houses)	201	(70.0)	
Frequency of bednet use	201	(20.0)	
Fvery night	344	(50.4)	
Sometimes	80	(30.4)	
Only in rainy season	00 07	(11.7) (14.2)	
Not used	20	(14.2)	
No bodnot	129	(4.3)	
Time entering hednet	132	(19.4)	
Pofore 20:00	205	(56 5)	
20.00 ± 22.00	07	(30.3)	
20:00 to 22:00	97	(14.2)	
After 22:00	39	(0.7)	
Not used / no bednet	101	(23.6)	
Bed level (N=315)			
Material of Ded	0.0.1	(70.0)	
Wooden bed	221	(70.2)	
Bamboo bed	50	(15.9)	
Hammock/mat on floor	44	(14.0)	
Possession of bednet			
Yes	246	(78.1)	
No	69	(21.9)	

Table 1 Characteristics of the study population.

Table 1 (continued).

Variables	n	(%)
Number of sleepers in bed		
1-3	264	(83.8)
4-6	51	(16.2)
Insecticide treatment of bednet		
Within 6 months	0	(0.0)
6-12 months before	106	(33.7)
>12 months before	29	(9.2)
Never treated	97	(30.8)
Not used/no bednet	83	(26.3)
Total area of holes in bednet (cm ²	²)	
No holes	115	(36.5)
0.25-19.9	61	(19.4)
≥20	56	(17.8)
Not used/no bednet	83	(26.3)
Household level (<i>N</i> =159)		
Number of family members		
1-4	76	(47.8)
5-7	67	(42.1)
≥8	16	(10.1)
Monthly income (USD)		
<77	96	(60.4)
78-149	42	(26.4)
≥150	21	(13.2)
Property		
TV and motorbike	83	(52.2)
TV only	50	(31.5)
Motorbike only	4	(2.5)
Neither TV nor motorbike	22	(13.8)
Material of walls		
Cement	41	(25.8)
Brick	20	(12.6)
Wood/bamboo	98	(61.6)

access to the nearest town within 30 minutes. Although there were no health facilities in the village, malaria diagnosis and treatment was available free of charge at public health facilities, such as the communal health center in the nearest town. While adults usually treated fever and other symptoms with over-the-counter medications, children displaying fever were immediately taken to private clinics.

Evening activities and bednet use

In the evening, all adults and children returned home to have dinner at around 5:00 to 6:00 PM. After dinner, 481 subjects (70.5%) stayed inside the house, relaxing with family or neighbors, mainly watching TV or chatting. Another 29.5%, mostly older teenagers and males in their 20s, visited relatives or friends in the village until 10:00 or 11:00 PM.

The 315 beds examined included wooden and bamboo beds, mats on the floor and hammocks. Bednets were available for 246 beds (78.1%). A total of 113 bednets (45.9%) had been supplied by the government, and 133 (54.1%) had been bought in the market within the last 5 years. All were polyester bednets, manufactured in Vietnam. Long-lasting insecticide-treated nets (LLITNs) were not yet available in the province. An insecticide treatment service was provided by the government twice a year, but the participation rate in this village was low. Only 106 bednets (33.7%) had been treated using insecticide within the last year. Among the 246 identified bednets, 115 (36.5%) had no holes, 117 (37.1%) had holes or were torn. The total area of the holes was >20 cm² in 56 bednets (17.8%).

Despite high availability, the frequency of using bednets was not high. Among the 682 subjects, 344 (50.4%) used a bednet every night, 80 (11.7%) used a bednet sometimes and 97 (14.2%) only used a bednet during the rainy season because the dry season was considered too hot to sleep under bednets. In total, 161 subjects (23.7%) slept without bednets. More than half the subjects (56.5%) entered the bednet before 20:00, but 39 (5.7%), mainly young male adults, entered later than 22:00. In spite of the fact that 98% of identified bednets were double-sized (190 x 190 x 190 cm) and made for a maximum of 2 adults and 1 small child, 51 bednets (16.2%) were shared by 4 - 5 people.

Risk factors for malaria infection

Table 2 shows the results of univariate analysis by chi-square test of the relationship between the prevalence of malaria infection and risk factors. A total of 42 subjects (6.2%) displayed malaria infection, including 35 subjects with *P. falciparum* and 7 with *P. vivax*. No mixed infections or complicated cases were encountered. Among the 42 slidepositive cases, 39 (92.8%) had no fever, headaches or other symptoms.

The chi-square test suggested age, material of house walls, number of family members and number of persons sleeping together affected risk for malaria infection. Among 0- to 2-year-old children, the prevalence of malaria was 3.1%, but 3- to 5-yearold children had the highest prevalence of 21.7% (OR, 8.6; 95%CI: 1.76, 42.16). Only 1 case was detected among subjects >30 years old (OR, 0.15; 95%CI: 0.01, 1.74). Wooden or bamboo houses had a higher risk for malaria infection compared to cement houses (OR, 4.18: 95%CI 1.45, 12.10). A large family represented one of the strongest risk factors. Compared with a family of 1 - 4 members, the OR for a family of 5 - 7 members was 4.54 (95%CI: 1.55, 13.3), and that of the biggest family (≥8 members) was 5.16 (95%CI: 1.58, 16.9). The number of sleepers in a bed was also important, with a higher prevalence of malaria infection seen for beds with 4 - 6 people (OR, 2.11; 95%CI: 1.12, 3.98) compared to beds with 1 - 3 people.

The linear logistic model selected as the most appropriate description of the effects of risk factors on malaria infection included the following factors as covariates: age, number of family members, material of walls, and frequency of using a bednet. Table 3 shows the estimated ORs and *p*-values for each factor included in the model. Children age 3 - 5 years old had a substantially higher infection rate than those age 0 - 2 years old (OR, 7.74; p < 0.01). Families with 5 - 7 members

Variables	Case / n (%	%)	Crude OR (95%CI)
Total	42 / 682	(6.2)	
Age (years)			
0-2	2 / 64	(3.1)	1.00
3-5	15 / 69	(21.7)	8.61 (1.76, 42.16)
6-9	6 / 59	(10.2)	3.51 (0.66, 18.56)
10-19	10 / 156	(6.4)	2.12 (0.45, 10.04)
20-29	8 / 130	(6.2)	2.03 (0.42, 9.94)
≥30	1 / 204	(0.5)	0.15 (0.01, 1.74)
Sex			
Male	17 / 327	(5.2)	1.00
Female	25 / 355	(7.0)	1.38 (0.73, 2.61)
Ethnicity			
Stieng	38 / 552	(6.9)	1.00
Kinh/Other	4 / 130	(3.1)	0.43 (0.15, 1.23)
Activities after dinner			
Stay in the house	28 / 481	(5.8)	1.00
Stay outside	14 / 201	(7.0)	1.21 (0.62, 2.35)
Possession of bednet			
Yes	34 / 550	(6.2)	1.02 (0.46, 2.26)
No	8 / 132	(6.1)	1.00
Frequency of using bednet			
Not used / no bednet	10 / 161	(6.2)	1.00
Only in rainy season	7 / 97	(7.2)	1.17 (0.43, 3.20)
Sometimes	8 / 80	(10.0)	1.68 (0.63, 4.45)
Every night	17 / 344	(4.9)	0.79 (0.35, 1.76)
Time entering bednet			
Not used / no bednet	10 / 161	(6.2)	1.00
Before 20:00	24 / 385	(6.2)	1.00 (0.47, 2.15)
20:00 to 22:00	7 / 97	(7.2)	1.17 (0.43, 3.20)
After 22:00	1 / 39	(2.6)	0.40 (0.05, 3.23)
Number of sleepers in bed			
1-3	23 / 483	(4.8)	1.00
4-6	19 / 199	(9.5)	2.11 (1.12, 3.98)
Insecticide treatment of bednet			
Not used / no bednet	10 / 161	(6.2)	1.00
6-12 months before	14 / 246	(5.7)	0.91 (0.39, 2.11)
≥12 months before	4 / 61	(6.6)	1.06 (0.32, 3.52)
Never treated	14 / 214	(6.5)	1.06 (0.46, 2.45)
Total area of holes in bednet (cm ²)			
Not used / no bednet	10 / 161	(6.2)	1.00
No holes	15 / 271	(5.5)	0.88 (0.39, 2.02)
0.25-19.9	7 / 129	(5.4)	0.87 (0.32, 2.35)
≥20	10 / 121	(8.3)	1.36 (0.55, 3.39)

 Table 2

 Risk factor analysis for malaria infection - univariate analysis.

Variables	Case / n (%	6)	Crud	e OR (95%CI)
Number of family members				
1-4	4 / 216	(1.9)	1.00	
5-7	27 / 342	(7.9)	4.54	(1.55, 13.3)
≥8	11 / 124	(8.9)	5.16	(1.58, 16.90)
Monthly income (USD)				
<77	29 / 405	(7.2)	1.00	
78-149	11 / 189	(5.8)	0.80	(0.39, 1.64)
≥150	2 / 88	(2.3)	0.30	(0.07, 1.30)
Property				
TV and motorbike	19 / 377	(5.0)	1.00	
TV only	16 / 207	(7.7)	1.58	(0.79, 3.15)
Motorbike only	1 / 13	(7.7)	1.57	(0.19, 12.75)
No TV / motorbike	6 / 85	(7.1)	1.43	(0.55, 3.70)
Material of wall				
Cement	4 / 192	(2.1)	1.00	
Brick	4 / 74	(5.4)	2.69	(0.65, 11.12)
Wood/bamboo	34 / 416	(8.2)	4.18	(1.45, 12.10)

Table 2 (continued).

(OR, 4.70; p < 0.01) and 8 - 13 members (OR, 4.91; p = 0.01) displayed a higher risk for malaria infection than smaller families. Subjects living in traditional wooden/bamboo housing also had a higher risk for malaria infection (OR, 5.57; p < 0.01) compared with those living in cement housing. Although no significant difference was apparent, the risk for malaria infection in those using a bednet sometimes, only in the rainy season or never appeared 1.66-fold higher than in those using a bednet every night.

DISCUSSION

In the remaining areas endemic for malaria in Vietnam, clear relationships between forest activities and malaria infection have been described (The Khanh Phu Malaria Research Project, 2004; Erhart *et al*, 2004, 2005; Thang *et al*, 2008). The present study was conducted in a sub-mountainous area, and most of the study population were of minority background. However, malaria infection was not associated with forest activities, but instead with age, material used to construct walls and family size.

Due to rapid economic improvements and a changing natural environment, the malaria situation (including treatment and prevention behaviors and human-vector contact patterns) has also changed. In the study site, inhabitants had no forest activities and lived far from the natural forest, and were bitten by vector mosquitoes around and inside their houses at night. Poor housing using wood/bamboo materials represented one of the stronger risk factors, as reported in numerous studies (Konradsen et al, 2003; Lindsay et al, 2003; Ernst et al, 2006; Ye et al, 2006; Kirby et al, 2008; Thang et al, 2008). Residences in the village were located far from natural forest, but were dotted among cashew nut orchards and surrounded by numerous small streams. Wooden/bamboo houses were built directly on the earth and roofed with sheet-iron, but the eaves

Table 3
Risk factors for malaria infection -
adjusted multivariate analysis.

Variables	OR	<i>p</i> -value
Age (years)		
0-2	1.00	
3-5	7.74	0.01
6-9	2.45	0.29
10-19	1.83	0.45
20-29	2.40	0.29
≥30	0.14	0.11
Number of family members		
1-4	1.00	
5-7	4.70	< 0.01
≥8	4.91	0.01
Material of walls		
Cement	1.00	
Brick	3.26	0.12
Wood/bamboo	5.57	< 0.01
Frequency of using bednet		
Every night	1.00	
Sometimes/rainy season/ not used	1.66	0.16

were wide open. An. dirus mosquitoes were collected inside houses in light traps (unpublished data). Obsomer et al (2007) described An. dirus, the vector mosquito in this area, as strongly associated with deep forest for larval habitats and adult survival, but the species has been adapting to man-made habitats such as orchards and plantations. An. dirus is known as an extremely anthropophilic species (Dutta et al, 1996; The Khanh Phu Malaria Research Project, 2004; Trung et al, 2005; Obsomer et al, 2007), and usually bites individuals outside houses. However, very open houses in the jungle show no significant difference between indoors and outdoors (Tun-Lin et al. 1995: Dutta et al. 1996: Vythilingam et al, 2003). In some cases, biting is reportedly even more frequent indoors than outdoors (Obsomer et al. 2007). Iron roofing has protective effects against malaria (Ye *et al*, 2006), but risk for malaria infection remains high if a roof is present but the eaves are open (Lindsay *et al*, 2003). Such housing conditions decrease the outdoor/indoor difference and allow *An. dirus* access to bite indoors.

Bigger families show a higher prevalence for malaria infection than smaller families. This may be explained by a greater concentration of human attractants for mosquitoes, since the highly anthropophilic An. dirus is attracted to human odor (Takken et al, 1999). Increasing the number of persons in a house has been identified as an ecological risk factor for malaria infection in Kenya (Ernst et al, 2006). A house occupied by humans has been shown to attract more Anopheline mosquitoes than an unoccupied house (Takken et al, 1999; Kirby et al, 2008). The relationship between malaria infection and the number of persons sleeping together represents another explanatory factor. In our study, a bigger sleeping unit attracted more mosquitoes than a smaller one. Moreover, the protective efficacy of the bednet is reduced in a crowded sleeping unit, because part or all of the body can be pushed out from the bednet, or touch the bednet directly and be bitten by mosquitoes outside the bednet. This is an example of reducing the efficacy of the bednet with inappropriate use.

In the study area, bednets are protective against malaria infection because vector mosquitoes bite late at night and people regularly sleep inside the house at night. However, inappropriate use of the bednet reduces the protective effect. Bednets, particularly insecticide-treated nets (ITNs), are highly effective tools for reducing malaria mortality and morbidity (Binka *et al*, 1996; Abdulla *et al*, 2001; Lengeler, 2004; Muller *et al*, 2006). Even untreated, a bednet in relatively good condition can protect against malaria (Clarke *et al*, 2001; Mwangi *et al*, 2003). However, compliance with the proper use of the bednet is not always good. As Korenromp et al (2003) reported, bednet possession and use are not always correlated. In this population, nearly half of bednets were not used every night. Furthermore, sleeping under a bednet is not preferred during the hot season (Yohannes et al, 2000; Frey et al, 2006). Even if used every night, the protective efficacy is reduced if the bednet is not in good condition. Bednets are easily damaged in rural settings such as when washing the bednet in a stream, hanging on a tree to dry, or hanging on a bamboo bed. Moreover, participation in the insecticide treatment service was low among the study population, despite free provision of this service twice a year. As several studies reported, regular insecticide treatment is difficult due to socio-economic and delivery system problems (Snow et al, 1999; Brentlinger et al, 2007; Kazembe et al, 2007; Webster et al, 2007). The results of this study area show problems not only due to inadequate knowledge and low motivation for re-treatment, but also due to inconvenience of the service activity. Not only distribution, but also follow-up maintenance should be considered. Furthermore, as recommended by the World Health Organization (2007). introduction and distribution of LLITN is warranted.

The prevalence of malaria infection was significantly higher among 3 - 5 year olds than among 0 - 2 year olds. Treatment behaviors for infants may have affected the prevalence of malaria infection, as preventive behaviors, such as frequency of bednet use and time to enter bednet, did not differ greatly between these age groups. The expansion of national malaria control activities in remote areas and the rapid increase in cash income from the cashew nut business among this population have enabled prompt access to diagnosis and treatment of malaria in health facilities. The inhabitants recognize that high fever in infants is dangerous from their long experience living in areas endemic for malaria, so infants receive immediate attention in health facilities in the town when fever is identified. As infants get older, the symptoms get milder or even unnoticeable due to acquired immunity (Aponte *et al*, 2007). For 3- to 5-year-old children, mosquito bites are unlikely to be increased, but active treatment is decreased due to asymptomatic conditions. This shows as a clear increase in the prevalence of malaria infection among children 3 - 5 years old.

In conclusion, to control malaria in endemic areas with changing conditions, control strategies need to be designed to fit the local context, including cultural diversity. Malaria in the area investigated in this study could be controlled by basic activities, such as early diagnosis and treatment and prevention by bednet use, because risk factors for malaria infection were not forest activities, but young age, living in wooden/bamboo housing and belonging to a large family. Continuous and intensive expansion of existing malaria control activities is required.

ACKNOWLEDGEMENTS

We would like to extend our sincere thanks to all the inhabitants of Phu Thuan Village. We are grateful to the staff of the National Institute of Malariology, Parasitology and Entomology (NIMPE), Hanoi. Special thanks also to the staff of the Binh Phuoc Provincial Malaria Control Center for their technical support. Financial support for this study was partially provided by the Core University Program of the Japan Society for the Promotion of Science (JSPS), a Grant-in-Aid for Scientific Research from the Ministry of Education, Science, Sports and Culture (#17651132 for KM), and from Showa Shell Sekiyu KK (A201, 2004 for TA).

REFERENCES

- Abdulla S, Schellenberg JA, Nathan R, *et al.* Impact on malaria morbidity of a programme supplying insecticide treated nets in children aged under 2 years in Tanzania: community cross sectional study. *BMJ* 2001; 322: 270-3.
- Aponte JJ, Menendez C, Schellenberg D, *et al.* Age interactions in the development of naturally acquired immunity to *Plasmodium falciparum* and its clinical presentation. *PLoS Med* 2007; 4: e242.
- Binka FN, Kubaje A, Adjuik M, *et al.* Impact of permethrin impregnated bednets on child mortality in Kassena-Nankana district, Ghana: a randomized controlled trial. *Trop Med Int Health* 1996; 1: 147-54.
- Brentlinger PE, Correia MA, Chinhacata FS, Gimbel-Sherr KH, Stubbs B, Mercer MA. Lessons learned from bednet distribution in Central Mozambique. *Health Policy Plan* 2007; 22: 103-110.
- Clarke SE, Bogh C, Brown RC, Pinder M, Walraven GE, Lindsay SW. Do untreated bednets protect against malaria? *Trans R Soc Trop Med Hyg* 2001; 95: 457-62.
- Dutta P, Bhattacharyya DR, Khan SA, Sharma CK, Mahanta J. Feeding patterns of *Anopheles dirus*, the major vector of forest malaria in northeast India. *Southeast Asian J Trop Med Public Health* 1996; 27: 378-81.
- Erhart A, Ngo DT, Phan VK, *et al.* Epidemiology of forest malaria in central Vietnam: a large scale cross-sectional survey. *Malar J* 2005; 4.
- Erhart A, Thang ND, Hung NQ, *et al.* Forest malaria in Vietnam: a challenge for control. *Am J Trop Med Hyg* 2004; 70: 110-8.
- Ernst KC, Adoka SO, Kowuor DO, Wilson ML, John CC. Malaria hotspot areas in a highland Kenya site are consistent in epidemic and non-epidemic years and are associated with ecological factors. *Malar J* 2006; 5.
- Frey C, Traore C, De Allegri M, Kouyate B, Muller O. Compliance of young children with ITN protection in rural Burkina Faso. *Malar J* 2006; 5.

- Hung LQ, Vries PJ, Giao PT, *et al.* Control of malaria: a successful experience from Viet Nam. *Bull World Health Organ* 2002; 80: 660-6.
- Hung NM. Malaria control program in Vietnam and plan for 2008. Country epidemiological profile Vietnam 2008. ACT Malaria Org. [Cited 2008 Sep 20]. Available from: URL: http://www.actmalaria.org2008.
- Kazembe LN, Appleton CC, Kleinschmidt I. Geographical disparities in core population coverage indicators for roll back malaria in Malawi. *Int J Equity Health* 2007; 6: 5. Epub 2007 Jul 4. [Cited 2008 Sep 20]. Available from: URL: <u>http://www.pubmedcentral.</u> nih.gov/articlerender.fcgi?artid=193490
- Kirby MJ, Green C, Milligan PM, et al. Risk factors for house-entry by malaria vectors in a rural town and satellite villages in The Gambia. Malar J 2008; 7: 20. Epub 2008 Jan 7. [Cited 2008 Sep 20]. Available from: URL: http://www.malariajournal.com/content/7/ 1/2
- Klinkenberg E, Konradsen F, Herrel N, Mukhtar M, van der Hoek W, Amerasinghe FP. Malaria vectors in the changing environment of the southern Punjab, Pakistan. *Trans R Soc Trop Med Hyg* 2004; 98: 442-9.
- Konradsen F, Amerasinghe P, van der Hoek W, Amerasinghe F, Perera D, Piyaratne M. Strong association between house characteristics and malaria vectors in Sri Lanka. *Am J Trop Med Hyg* 2003; 68: 177-81.
- Korenromp EL, Miller J, Cibulskis RE, Kabir Cham M, Alnwick D, Dye C. Monitoring mosquito net coverage for malaria control in Africa: possession vs. use by children under 5 years. *Trop Med Int Health* 2003; 8: 693-703.
- Lengeler C. Insecticide-treated bed nets and curtains for preventing malaria. *Cochrane Database Syst Rev* 2004; 2: CD000363.
- Lindsay SW, Jawara M, Paine K, Pinder M, Walraven GE, Emerson PM. Changes in house design reduce exposure to malaria mosquitoes. *Trop Med Int Health* 2003; 8: 512-7.

- Meyfroidt P, Lambin EF. Forest transition in Vietnam and its environmental impacts. *Global Change Biol* 2008; 14: 1319-36.
- Muller O, Traore C, Kouyate B, et al. Effects of insecticide-treated bednets during early infancy in an African area of intense malaria transmission: a randomized controlled trial. Bull World Health Organ 2006; 84: 120-6.
- Mwangi TW, Ross A, Marsh K, Snow RW. The effects of untreated bednets on malaria infection and morbidity on the Kenyan coast. *Trans R Soc Trop Med Hyg* 2003; 97: 369-72.
- Nam NV, Vries PJ, Van LT, Nagelkerke N. Malaria control in Vietnam: the Binh Thuan experience. *Trop Med Int Health* 2005; 10: 357-65.
- National Institute of Malariology Parasitology and Entomology (NIMPE) V. The Khanh Phu Malaria Research Project: Review Meeting Report. Hanoi: NIMPE, 1996: 134-50.
- Obsomer V, Defourny P, Coosemans M. The Anopheles dirus complex: spatial distribution and environmental drivers. Malar J 2007; 6: 16. Epub 2007 Mar 6. [Cited 2008 Sep 20]. Available from: URL: <u>http://www.</u> malariajournal.com/content/6/1/26
- Snow RW, McCabe E, Mbogo CN, *et al.* The effect of delivery mechanisms on the uptake of bed net re-impregnation in Kilifi District, Kenya. *Health Policy Plan* 1999; 14: 18-25.
- Takken W, Knols BG. Odor-mediated behavior of Afrotropical malaria mosquitoes. *Annu Rev Entomol* 1999; 44: 131-57.
- Thang ND, Erhart A, Speybroeck N, *et al.* Malaria in central Vietnam: analysis of risk factors by multivariate analysis and classification tree models. *Malar J* 2008; 7.
- The Khanh Phu Malaria Research Project. Overview 10 years of malaria studies (1993-2003) in Khanh Phu. Hanoi, Vietnam. Hanoi: National Institute of Malariology, Parasitology and Emtomology (NIMPE), 2004.
- The National Malaria Control Project. Guidelines for malaria diagnosis and treatment in Vietnam. Hanoi: Ministry of Health, 2003.
- Thuan LK. Evaluation of the malaria control 2003

and plan of action for 2004 in Vietnam. Country epidemiological profile, Vietnam, 2005. ACT Malaria Org. [Cited 2008 Sep 20]. Available from: URL: <u>http://www.actmalaria.</u> <u>net2005</u>

- Trung HD, Bortel WV, Sochantha T, Keokenchanh K, Briet OJ, Coosemans M. Behavioural heterogeneity of *Anopheles* species in ecologically different localities in Southeast Asia: a challenge for vector control. *Trop Med Int Health* 2005; 10: 251-62.
- Trung HD, Van Bortel W, Sochantha T, *et al.* Malaria transmission and major malaria vectors in different geographical areas of Southeast Asia. *Trop Med Int Health* 2004; 9: 230-7.
- Tun-Lin W, Thu MM, Than SM, Mya MM. Hyperendemic malaria in a forested, hilly Myanmar village. *J Am Mosq Control Assoc* 1995; 11: 401-7.
- Vythilingam I, Phetsouvanh R, Keokenchanh K, et al. The prevalence of Anopheles (Diptera: Culicidae) mosquitoes in Sekong Province, Lao PDR in relation to malaria transmission. Trop Med Int Health 2003; 8: 525-35.
- Webster J, Hill J, Lines J, Hanson K. Delivery systems for insecticide treated and untreated mosquito nets in Africa: categorization and outcomes achieved. *Health Policy Plan* 2007; 22: 277-93.
- World Health Organization: Global Malaria Programme. Insecticide-treated mosquito nets: a WHO position statement. Geneva: WHO, 2007.
- Yasuoka J, Levins R. Impact of deforestation and agricultural development on anopheline ecology and malaria epidemiology. *Am J Trop Med Hyg* 2007; 76: 450-60.
- Ye Y, Hoshen M, Louis V, Seraphin S, Traore I, Sauerborn R. Housing conditions and *Plasmodium falciparum* infection: protective effect of iron-sheet roofed houses. *Malar J* 2006; 5.
- Yohannes K, Dulhunty JM, Kourleoutov C, *et al.* Malaria control in central Malaita, Solomon Islands. 1. The use of insecticide-impregnated bed nets. *Acta Trop* 2000; 75: 173-83.