

EFFECT OF HOUSING FACTORS ON INFESTATION BY *Aedes aegypti* (L.) AND *Aedes albopictus* SKUSE IN URBAN HANOI CITY, VIETNAM

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Abstract. To determine the effect of housing factors on infestation with *Aedes aegypti* (L.) and *Aedes albopictus* Skuse we conducted an entomological survey and inspection of 267 urban houses in Hanoi City, Vietnam. Two hundred ten pupae and 194 adult *Ae. aegypti* were collected from 19 and 88 houses, respectively. One hundred eighty-one pupae and 24 adult *Ae. albopictus* were collected from 21 and 14 houses, respectively. The presence of a private well was associated with increasing infestation with *Ae. aegypti* adults ($p = 0.01$) and increased the risk of *Ae. aegypti* and *Ae. albopictus* pupal presence ($p = 0.04$ for *Ae. aegypti*, $p = 0.03$ for *Ae. albopictus*). The presence of an outdoor space in the household premises was associated with a higher risk of *Ae. albopictus* pupal presence ($p = 0.004$) and a higher risk of high levels of *Ae. albopictus* adults ($p = 0.01$); however, it had no association with infestation with *Ae. aegypti*. The presence of an air-conditioning unit ($p = 0.03$) and four or more rooms in the residence ($p = 0.02$) were negatively and positively associated with the risk for *Ae. albopictus* presence, respectively.

Keyword: *Aedes aegypti*, *Aedes albopictus*, housing factor, infestation, Vietnam

INTRODUCTION

Dengue outbreaks are caused by interactions among ecological and biological factors of the pathogen (dengue virus), host (human) and vector (*Aedes* mosquitoes) (Guzmán and Kourí, 2002; Halstead, 2007). The dengue virus is transmitted to

humans by the bite of an infected vector mosquito. Effective vector control strategies are important to reduce the burden of dengue.

Hanoi, the capital city of Vietnam, is located in the north of the country. It is characterized by a warm, humid, subtropical climate with plentiful precipitation. Periodic outbreaks of dengue are common in the city, but a large number of cases were reported in 2009, with more than 20,000 individuals being hospitalized (Cuong *et al*, 2011). The *Aedes aegypti* mosquito is the primary dengue vector inhabiting tropical and subtropical regions, whereas

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Aedes albopictus is considered a secondary vector in tropical and temperate areas (Higa, 2011). These two mosquito species co-exist in Hanoi (Phong and Nam, 1999).

The effect of various housing factors on larval and pupal infestation by *Ae. aegypti* and *Ae. albopictus* has been reported in several studies (Takagi *et al*, 1995; Higa *et al*, 2001; Howell and Chadee, 2007; Tsuzuki *et al*, 2009a); however, specific housing factors affecting infestation by both species in an area have not been investigated in Vietnam. The effect of housing factors on adults *Aedes* mosquitoes is important since only adult mosquitoes are able to transmit dengue viruses. Since both *Aedes* species are present in Hanoi, vector control problems need to be aware of specific housing factors effecting the adults of both these species. To determine the housing features that influence infestation by the pupae and adults of *Ae. aegypti* and *Ae. albopictus*, we conducted house-to-house visits and determined vector infestation in urban residences of Hanoi.

MATERIALS AND METHODS

Hanoi is located in the north of Vietnam and has a sub-tropical climate with a hot, rainy season from April to October and a cool, dry season between November and March. The city has a population of approximately 6 million and is divided into 10 urban districts (2 million people) and 19 suburban districts (4 million people). This study was conducted in three neighboring central urban districts (Hai Ba Trung, Dong Da, and Hoan Mai) where the greatest number of dengue cases was reported in 2009.

Thirty non-adjacent residential neighborhoods were selected within the three districts for investigation. Each neighborhood is comprised of 100-372 residences.

A household was defined as a basic residential unit in which economic production, consumption, inheritance, child rearing and/or shelter were organized or carried out. An identification number was assigned to each household based on a list maintained by the local health office. Households could consist of members living in a solitary house with one or more buildings, a row house (consisting of two or more home buildings connected to each other) an apartment or a dormitory room. Using computer-generated random numbers, 10 households were selected from each of 28 neighborhoods (plus 15 households from each of two neighborhoods) for a total of 310 households. Household visits were conducted between 9 and 27 August 2010 since most dengue cases are reported between August and December.

We collected adult mosquitoes from the household premises, including all indoor rooms and outdoor areas, such as front yards, verandas and top floors. A backpack aspirator is usually a standard *Aedes* adult mosquito collection device (WHO, 2003); but the dwellings in the study area were crowded, and movement in the narrow alleys and small rooms would have been difficult considering the size and weight of the backpack aspirator. Two trained investigators carefully collected resting and flying mosquitoes by sweeping back and forth with a handnet. A torch light was used to detect mosquitoes in indoor spaces. We spent 20-30 minutes per household in collecting mosquitoes. Collected mosquitoes were stored in glass tubes and identified at local laboratories. During household visits, mosquito pupae were also collected according to previously described standardized procedures and breeding sites were similarly categorized as bonsai (a planting art form using miniature trees grown in

containers), discarded outdoor containers (all types of artificial containers discarded around the premises), or concrete basins (built-in containers that hold water for cleaning dishes and/or clothes, generally located in the kitchen) (Tsuzuki *et al*, 2009b). For households with private wells, standardized funnel traps (made from plastic containers, funnels, and sinkers purchased at a local supermarket) were used. Each funnel trap was left in the well for 16-18 hours overnight and the number of pupae in the wells was estimated based on the number of pupae collected in the trap, as described previously (Kay *et al*, 1992). We defined key container types (important immature breeding sites) as those having pupal productivity (number of pupae collected from the container type \times 100/total number of pupae collected) of 10.0% or more, and efficiency (pupal productivity/prevalence of the container type) of 1.0 or more based on the definition of Tsuzuki *et al* (2009c).

Housing variables related to the use of water (*ie*, presence of a tap or private well) and the number of household members, were recorded, since these have been reported to influence *Ae. aegypti* infestation in residences in Vietnam (Tsuzuki *et al*, 2009a). Structural aspects of residences (residential type, number of rooms, presence of an outdoor space, presence of an air conditioning unit) were noted as potential factors affecting infestation with the two species. Since the number of pupae directly affects the number of adult mosquitoes in a household, the number of pupae collected for each species was included as a continuous explanatory variable on the adult mosquito analysis.

To account for differences in mosquito infestation levels between neighborhoods, a generalized linear mixed model (GLMM) with the 26 neighborhoods as a

random effect was applied to determine the effect of housing variables on the presence of pupae and adults. Neighborhood was considered as a random intercept, and the modeling allowed for a random slope to find optimal random errors. For analysis of the adult data, a GLMM using the package "glmmADMB" in the R version 0.7.2.12 were applied. Initially, Poisson regression was applied to optimize the statistical model since the response variable (number of adults) was the count. A negative binomial regression was applied when the data were overdispersed. When a dataset included excess zeros (absence of adults), a zero-inflated regression model was used. An optimal statistical model was selected using Akaike Information Criterion (AIC). The significance of each explanatory variable remaining in the optimal model was estimated using a log likelihood ratio test. A GLMM using binomial distribution with the package "lme4" in R version 0.999999-0 was applied to analyze the pupal data and binomial regression was used to optimize the statistical model. The number of pupae collected per house was influenced not only by housing variables but also by the presence of productive breeding containers in the households. The productivity of individual containers could not be evaluated solely on its nominal category (flower vase, water jar) but could be affected by other factors such as its location, volume of water, presence and use of a lid and frequency of water use (Focks *et al*, 1993). Since we did not collect information about these attributes for each container, we simply evaluated the effect of the presence of each container type in addition to the other housing variables on the presence (or absence) of pupae in the household.

Housing variables used for the analysis were: residential type (1=isolated or

row house, 2=apartment or dormitory room), number of rooms except bathroom/toilet (1=1 to 3 rooms, 2=4 or more rooms), outdoor space, such as a back-/front-yard (1= no outdoor space, 2= with outdoor space), number of household members (1=1 to 4 residents, 2=5 or more residents), presence of an air-conditioner (1= no air-conditioner, 2=1 or more air-conditioners) and presence of a private well (1= no well, 2= well).

This study was reviewed and approved by the ethics committee that oversees research at the Institute of Tropical Medicine, Nagasaki University, Nagasaki, Japan and the bioethics committee of the National Institute of Hygiene and Epidemiology, Hanoi, Vietnam.

RESULTS

Of the 310 households initially chosen in the 30 neighborhoods, the survey was conducted in 267 households (43 households were excluded due to refusal or non-occupation). Among the studied households, the majority of occupants (79.8%) lived in isolated or row houses while others lived in apartments or dormitory rooms. Approximately half of the premises (49.9%) were comprised of three or less rooms, with the exception of the kitchen, toilet and bathroom, whereas the other premises had more rooms (4-9 rooms). Fewer than half the households (37.1%) had a formal outdoor space, such as a front-yard. More than half of the households (61.0%) had four or fewer members; whereas the others were comprised of between 5 and 10 members. Most households (67.0%) had at least one air-conditioner. All the households had piped tap-water, but five households also owned a private well for household chores.

In the 267 households studied 633

water-holding containers were inspected (Table 1). We collected more *Ae. aegypti* pupae than *Ae. albopictus* pupae and more than half the *Ae. aegypti* pupae (56.2%) occurred in a well. Bonsai containers and wells were key breeding sites for *Ae. aegypti*, producing more than 10% of *Ae. aegypti* pupae (*ie*, pupal productivity: 12.4 and 56.2), and the efficacy of these containers (*ie*, pupal productivity/prevalence per container) was greater than 1.0 (2.1 and 71.1). Outdoor discarded containers were key sites for finding *Ae. albopictus* and contained the majority (118/181) of pupae collected in the survey.

Both mosquito species were collected in some households (34.8% for *Ae. aegypti*, 10.5% for *Ae. albopictus*) (Fig 1, Fig 2). The number of households positive for *Ae. aegypti* adults was four times more than the number of households positive for *Ae. aegypti* pupae. However, for *Ae. albopictus*, more households were positive for pupae than adults (Table 2). There were eight times more *Ae. aegypti* adults than *Ae. albopictus* adults (194 *Ae. aegypti* vs 24 *Ae. albopictus*); but the number of pupae were nearly equal (210 *Ae. aegypti* vs 181 *Ae. albopictus*). In the 93 *Ae. aegypti*-infested households, adults were found 95% of the time and pupae were found 20% of the time. However, in the 28 *Ae. albopictus*-infested households, adults were found 50% of the time and pupae were found 75% of the time (Fig 2).

The GLMM with negative binomial distribution had the lowest AIC values explaining the relationship between housing variables and the number of adult mosquitoes collected for both species. For *Ae. aegypti* adults, the optimal model indicated residency in solitary or row houses ($p=0.01$) and the presence of a well ($p<0.001$) were positively associated with the number of adults collected. For *Ae. albo-*

Table 1
Presence of *Aedes aegypti* (AEG) and *Aedes albopictus* (ALB) in breeding containers from 267 residential houses.

Container type	Number of containers	Prevalence per container ^a	Container with pupae		Number of pupae		Pupal productivity ^b		Pupal efficiency ^c	
			AEG	ALB	AEG	ALB	AEG	ALB	AEG	ALB
Flower vase	172	27.2	3	3	8	9	3.8	5.0	0.1	0.2
Plastic bucket	151	23.9	2	10	10	37	4.8	20.4	0.2	0.9
Discarded outdoor container	114	18.0	6	16	19	118	9.0	65.2	0.5	3.6
Concrete basin	83	13.1	4	2	16	2	7.6	1.1	0.6	0.1
Bonsai pot	37	5.8	4	1	26	3	12.4	1.7	2.1	0.3
Jar	19	3.0	2	2	2	4	1.0	2.2	0.3	0.7
Well	5	0.8	1	0	118	0	56.2	0.0	71.1	0.0
Metal drum	2	0.3	1	0	1	0	0.5	0.0	1.5	0.0
Others	50	7.9	2	3	10	8	4.8	4.4	0.6	0.6
Total	633	100.0	25	37	210	181	100.0	100.0	1.0	1.0

^aPrevalence per container = no. per container x 100/all containers; ^bProductivity = pupae no. x 100/all pupae. ^cEfficiency = productivity/prevalence per container.

Table 2
The number of households positive for *Aedes aegypti* (AEG) and *Aedes albopictus* (ALB) pupae and adults in the 267 residences.

	Number of households	
	AEG positive	ALB positive
Adults	88	14
Pupae	19	21

pictus adults, the presence of an outdoor space ($p=0.08$) and the number of *Ae. albopictus* pupae ($p=0.003$) were factors associated with greater number of adults.

In 225 households (excepting 42 which had no water-holding containers), the GLMM with binomial distribution revealed that the presence of a private well was positively associated with the presence of *Ae. aegypti* pupae ($p=0.04$) but

other variables were not significant. Having a well ($p=0.03$), four or more rooms ($p=0.02$) and an outdoor space on the premises ($p=0.004$) were all significantly associated with the presence of *Ae. albopictus* pupae. The presence of an air-conditioning unit was negatively associated with the presence of *Ae. albopictus* pupae ($p=0.03$).

DISCUSSION

The presence of a private well was significantly associated with greater number of *Ae. aegypti* adults in urban residential Hanoi. Wells are usually located indoors or under the eaves. The conditions inside wells are suitable for *Ae. aegypti* adults since they prefer dimly lit, humid indoor areas (Rickard, 2009). Private wells increased the risk for *Ae. aegypti* and *Ae. albopictus* pupae. Water from the wells is not used for drinking but for other purposes, such as washing dishes

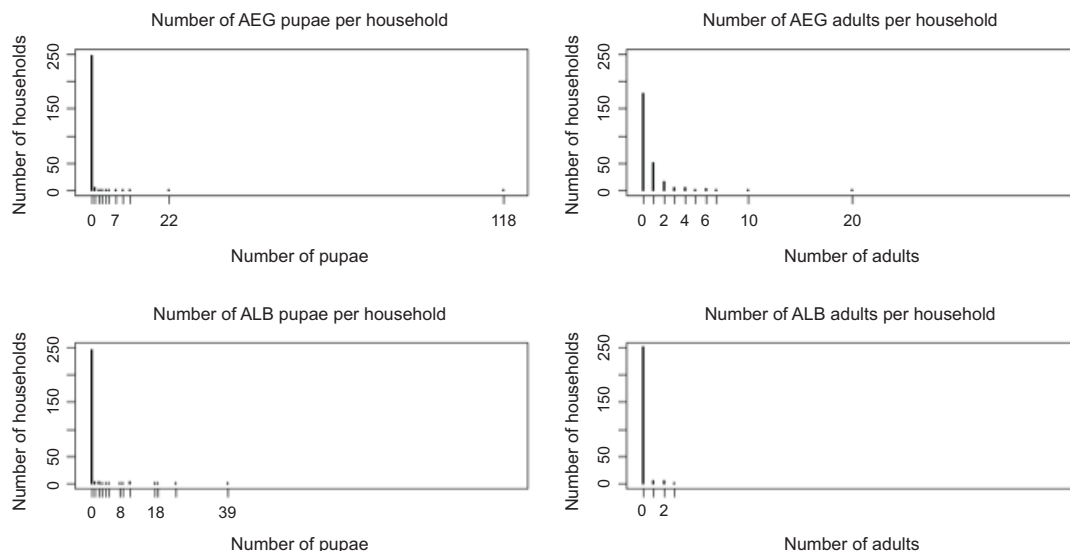


Fig 1–The number of *Aedes aegypti* (AEG) and *Aedes albopictus* (ALB) pupae and adults collected from 267 residences.

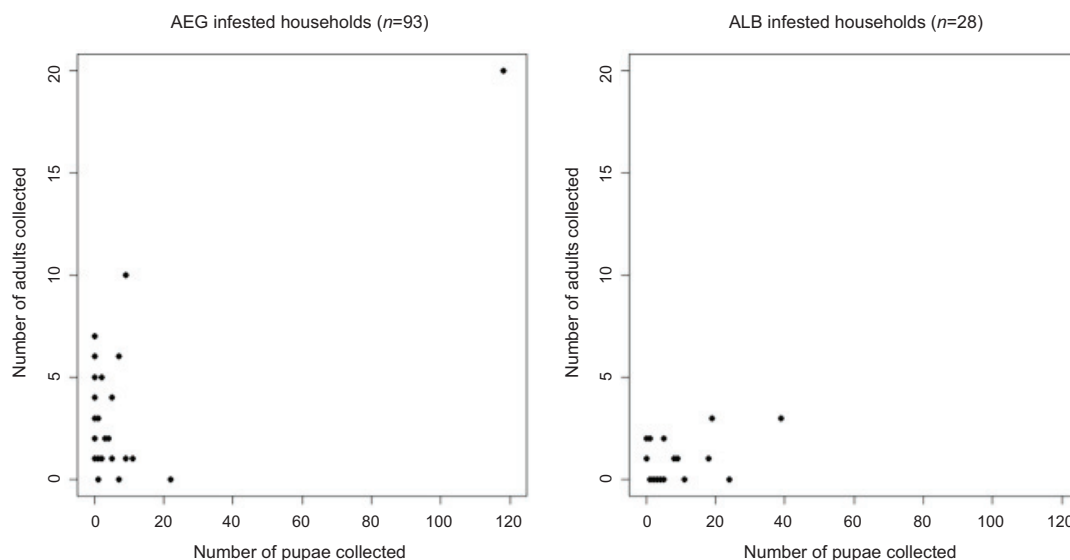


Fig 2–Relationship between the number of pupae and adult *Aedes aegypti* (AEG) and *Aedes albopictus* (ALB) collected.

and clothes, toilet flushing, cleaning and watering plants. Storing well water in containers may increase the risk for infestation compared to stored tapwater, since both species prefer to lay eggs in water containing organic matter (Bentley and Day, 1989). One household with a private

well had 118 *Ae. aegypti* pupae (56.2% of the total collected in our study). The well was located in a small, dim hut used as a storage room for kitchen utensils. All the pupae were found in the well itself and most of the adults were collected from the hut. A study from southern Vietnam

found the presence of a private well in urban premises was an important risk factor for *Ae. aegypti* pupae infestation (Tsuzuki *et al*, 2009b). Therefore, containers on the premises with wells are appropriate primary targets for source reduction of immature *Aedes* in urban Hanoi. The infestation level with *Ae. aegypti* adults was high in isolated and row houses. This is probably because these types of houses have more interior resting places for *Ae. aegypti* adults compared to apartments and dormitory rooms. The number of *Ae. albopictus* adults collected was positively associated with the number of pupae collected. This is due to the adults emerging from pupae.

The presence of an outdoor space was associated with *Ae. albopictus* adults and pupae, but was not associated with the number of *Ae. aegypti*; pupae and adults. Several studies from Vietnam have found immature *Ae. aegypti* in urban residences use various types of water-holding containers in and around houses as breeding sites since this is where they deposit their eggs; however, immature *Ae. albopictus* preferred outdoor discarded containers (Phong and Nam, 1999; Tsuzuki *et al*, 2009a,b) similar to the present study (Table 1). In our study, most households positive for *Ae. albopictus* pupae were negative for *Ae. albopictus* adults and most households positive for *Ae. aegypti* adults were negative for *Ae. aegypti* pupae (Fig 2). These results probably indicate most *Ae. albopictus* adults disperse accidentally (*ie*, by wind) or intentionally (*ie*, seeking a source of blood) soon after emerging from outdoor discarded containers, since they feed on humans and domestic/companion animals (*ie*, dogs, cats and chickens) in urban areas (Gratz, 2004; Ponlawat and Harrington, 2005). However, *Ae. aegypti* adults remain near their original breed-

ing sites since they feed exclusively on human blood and have no need to seek out other sources of food (Ponlawat and Harrington, 2005; Suwonkerd *et al*, 2006). The daily survival rate of *Ae. albopictus* adults was lower than *Ae. aegypti* adults, since *Ae. aegypti* adults rest indoors before and after blood feeding and lay eggs in artificial containers in and around homes. In contrast, *Ae. albopictus* adults prefer to rest outdoors near vegetation before and after feeding and lay eggs in outdoor discarded containers (Takagi *et al*, 1995; Higa *et al*, 2001) and are more vulnerable to the effects of fluctuating weather conditions (temperature, humidity, rainfall) and natural enemies, such as bats and spiders. Hence, households with outdoor spaces may have more outdoor discarded containers for pupae and outdoor resting places for adults of *Ae. albopictus*.

The existence of a private well was the only factor increasing the risk for *Ae. aegypti* pupae in our study. For *Ae. albopictus*, other housing factors influenced the risk for having pupae. A house with four or more rooms was at higher risk of having *Ae. albopictus* pupae. This may be because residences with more rooms usually have larger eaves and verandas increasing the likelihood of having outdoor containers. Having an air-conditioner on the premises reduced the risk of having *Ae. albopictus* pupae. Houses with air-conditioning usually close windows to maintain efficacy. This might give *Ae. albopictus* adults, which prefer outdoor conditions (Higa *et al*, 2001), less opportunity for blood feeding of residents compared to house that have open windows for ventilation, thus reducing the presence *Ae. albopictus* pupae.

Immature *Ae. aegypti* inhabit a variety of water-holding containers in and around residences in urban areas (Phong and

Nam, 1999; Tsuzuki *et al*, 2009a,b) making it difficult to identify target containers for control. However, targeting private wells and household premises with wells might reduce the workload of vector control activities in urban areas since this factor increases the risk of *Ae. aegypti* and *Ae. albopictus* pupae and *Ae. aegypti* adults. The use of larvicides is legally restricted in Vietnam; therefore, the use of natural enemies, such as larvivorous fish (Seng *et al*, 2008) and copepods (Kay and Vu, 2005) and insecticide-treated covers (Vanlerberghe *et al*, 2011) appear to be effective measures for protecting wells and surrounding premises from *Ae. aegypti*.

Although we carefully investigated indoor and outdoor areas around residences, we collected only a few *Ae. albopictus* adults (24 *Ae. albopictus* vs 194 *Ae. aegypti*). The importance of *Ae. albopictus* as a dengue vector is probably low in our study area; however, *Ae. albopictus* have the potential to cause outbreaks of dengue in areas with high mosquito density (Effler *et al*, 2005; Wu *et al*, 2010), as well as outbreaks of chikungunya (Pialoux *et al*, 2007). Control of *Ae. albopictus* should include control of areas with many outdoor discarded containers, such as vacant lots, construction areas, temples and premises with large outdoor spaces.

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