ABSENCE OF *AEDES AEGYPTI* (L.) ON AN ECOLOGICAL ISLAND: COMPETITIVE EXCLUSION?

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Abstract. Ovitrap surveillance was conducted in 2012 and 2006 in Malay and Aboriginal Villages on Carey Island. In each village, standard ovitraps were placed indoors and outdoors at randomly selected houses/locations. All L3 larvae recovered were identified up to species level. Results demonstrated that only larvae of *Aedes albopictus* were found in all the positive ovitraps placed indoors and outdoors. In 2012, a high ovitrap index (OI) of 66.7% indoor and 84.0% outdoor in the Malay Village; and 62.5% indoor and 88.0% outdoor in Aboriginal Village with an apparent absence of *Aedes aegypti*. In 2006, a 100% OI was recorded in all ovitraps set indoors and outdoors in both villages.

Keywords: *Aedes aegypti*, *Aedes albopictus*, ovitrap index, Carey Island, Malaysia

INTRODUCTION

Dengue and dengue hemorrhagic fever are the two most frequently reported *Aedes*-borne diseases in Malaysia. The first classical dengue and dengue hemorrhagic fever (DHF) cases were reported in Penang by Skae in 1902 and Rudnick *et al* in 1962, respectively (Skae, 1902; Rudnick *et al*, 1965). *Aedes* (*Stegomyia*) *aegypti* (Linnaeus) and *Aedes* (*Stegomyia*) *albopictus* (Skuse) are the incriminated vectors for dengue (Smith, 1956; Rudnick, 1967; Hammond, 1966). In addition to dengue infection, the latter is also responsible for the first reported chikungunya outbreak in Klang, Selangor between 1998 and February 1999 (Lam *et al*, 2001). In 2006 and 2008, the re-emergence of chikungunya disease occurred in Bagan Panchor, Perak, and Johor State, which then spread to other states and federal territories in Malaysia (AbuBakar *et al*, 2007; Sam *et al*, 2009). The ability of both species to adapt to both urban and sub-urban environments enables them to spread rapidly wherever the suitable breeding conditions are available (Knudsen, 1995).

Study has shown that the abundance of *Aedes* sp, together with the combination of several other factors such as rainfall, temperature, and relative humidity are directly correlated with the abundance of dengue cases (Okogun *et al*, 2003). A general mosquito survey in Peninsular Malaysia was conducted by Rudnick (1967) from 1965-1969 to determine the roles that arboviruses play in producing infection and/or disease in Malaysia, with a special reference to their ecology. Intensive mosquito sampling trips were conducted covering numerous habitats in

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Peninsular Malaysia, ranging from urban villages and town, coastal fishing villages, remote primary forest areas, mangroves swamp forests, high mountain forest, plantations, and aboriginal villages.

One of the study sites established in his study was Carey Island, which is located approximately 16 km south of Port Klang. The island's area totals 16,187.45 ha, separated from the Selangor coast by the Langat River. Unlike many other islands, Carey Island is located in close proximity to the mainland, and the river that separates it from the mainland is practically a stream. In his study, Rudnick (1986) reported that only a small numbers of *Aedes* (Stegomyia) mosquitoes were present, other than *Ae. albopictus* with a complete absence of *Ae. aegypti* mosquitoes.

Most parts of the island are under cultivation with large interspersed field of rubber and oil palm. In a review paper by MacDonald (1956), in 1913, the seaports and coastal areas were populated by *Ae. aegypti*, which replaced *Ae. albopictus* as a common *Aedes* species in the towns. Considering the location of Carey Island, which is only approximately 16 km away from Port Klang, it is interesting to note the complete absence of *Ae. aegypti* on Carey Island. Since the last survey in 1968, another survey was conducted in 2006 by the Medical Entomology Unit, Institute for Medical Research, Kuala Lumpur via ovitrapping, and again, *Ae. albopictus* dominated the area with an apparent absence of *Ae. aegypti*.

In 2012, we decided to conduct ovitrapping on Carey Island in order to update our information on the abundance and dispersal of the *Aedes* spp, specifically the two major vectors for dengue; *Ae. aegypti* and *Ae. albopictus*, as well as to determine whether the invasion of *Ae. aegypti* population has occurred in that place, given that there are a lot of incoming and outgoing traffic in the island by the local people due to the presence of a paved road running through its middle in 2008.

The logistical and transportation needs have been further improved with the construction of the South Klang Valley Expressway (SKVE) project that connects main destinations in the Klang Valley to Carey Island and Indah Island (Fig 1). The growing numbers of factories, as well the development of the golf-course resorts, has made the movement of people less restricted. Therefore, the present study aims to investigate the distribution of the two main *Aedes* spp, particularly *Ae. albopictus* and *Ae. aegypti*, on Carey Island to determine whether there is a change in the abundance of these two species of mosquitoes.

MATERIALS AND METHODS

Study area

The study was carried out in the only Malay Village (latitude: 2° 49′ 11″; longitude: 101° 22′ 17″) and Aboriginal Village (latitude: 2° 50′ 26″; longitude: E101° 22′ 32″) on Carey Island in the Kuala Langat District of Selangor (Fig 2). During the course of the study, a mean ambient temperature of 28°C with 65% relative humidity was recorded in the study area. The monthly mean for rainfall intensity was recorded at 180 mm.

Mosquito collection

Aedes ovitrapping was conducted in July 2012, following the guidelines of Ministry of Health on ovitrap deployment (Ministry of Health, 1997). The ovitrap used consisted of a black plastic container

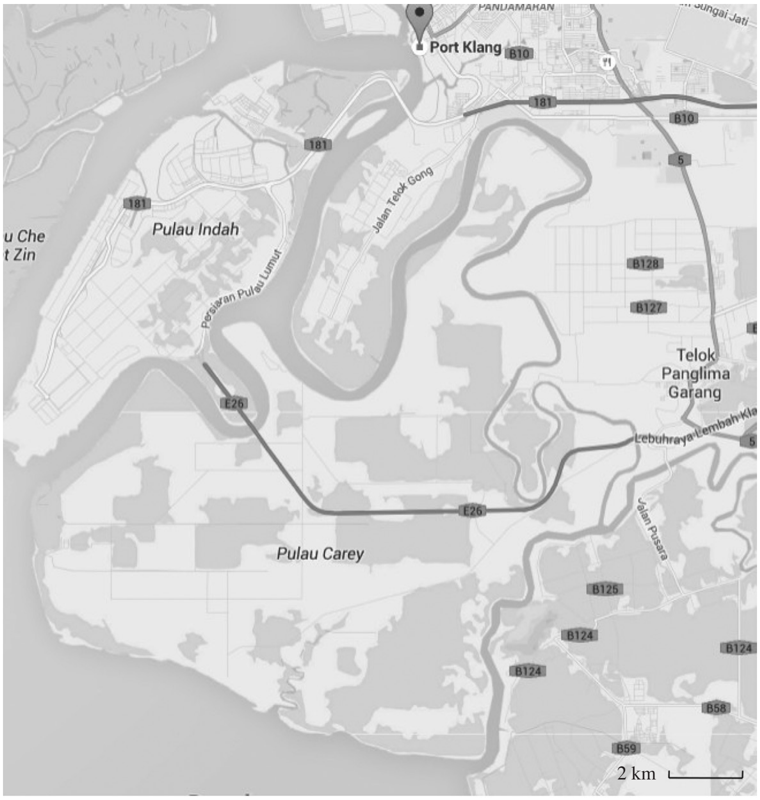

Fig 1–Carey Island is located approximately 16 km from Port Klang.

of 300-ml volume (base diameter 6.5 cm, opening diameter 7.8 cm and height 9.0 cm). A hard-board measuring 10 cm x 2.5 cm x 0.3 cm was placed in the ovitrap container with the rough surface upward to serve as an oviposition paddle. Clean water was added to a level of 5.5 cm. In each village, a total of 50 standard ovitraps were deployed, whereby 25 ovitraps were randomly set indoors and outdoors, respectively. Each ovitrap was located indoors and outdoors in randomly selected house. In this study, indoor is referred to the interior of the house and outdoor is outside of the house but confined to the immediate vicinity of the house particularly in the shaded area to avoid from direct sunlight and heavy rain that may cause water spillage. After 5 days of exposure, a total of 98 ovitraps was recovered from both villages and brought back to the Medical Entomology Unit laboratory and sorted accordingly.

In 2006, similar ovitrapping method was conducted in Malay and Aboriginal Villages, with a reduced number of ovitrap; a total of 20 in each village at which 10 ovitraps were deployed indoors and 10 ovitraps were set outdoors.

Identification of larvae

The contents of each ovitrap were poured into respective plastic containers, together with the paddles. The eggs on the paddle were allowed to hatch and larvae identification was conducted at L3 stage. Larvae were routinely fed on liver powder and liver chunk diet. All larvae (L3) recovered were identified to species level according to the key by Mahadevan and Cheong, 1974 and the numbers of larvae in each positive ovitrap were recorded.

Analysis of data

Data obtained were analyzed as follow:

Ovitrap index (OI) = (Number of positive traps / Number of recovered traps) x 100%

Mean larvae per trap = Total number of larvae / Number of recovered ovitraps

Independent *t*-test analysis was performed using SPSS® (version 19.0; IBM, Armonk, NY).

Fig 2–The sampling sites: the Malay Village and Aboriginal Village.

RESULTS

Only larvae of *Ae. albopictus* were found in all of the positive indoor and outdoor ovitraps. The ovitrap index was very high, 66.7% indoor and 84.0% outdoor in the Malay Village, and 62.5% indoor and 88.0% outdoor in the Aboriginal Village. A higher mean number of *Ae. albopictus* larvae per recovered indoor ovitrap was recorded in the Malay Village than in the Aboriginal Village (25.75±5.40 *vs* 20.46±5.00), while more larvae were recovered from outdoor ovitraps in the Aboriginal Village than the Malay Village (43.40±5.90 *vs* 31.08±6.40). Overall, the outdoor population of *Ae. albopictus* was significantly higher ($p<0.05$) than the indoor population (Table 1). By comparison, the 2006 ovitrap survey indicated a higher ovitrap index of 100% in all ovitraps placed indoors and outdoors in both villages.

In 2012, 1,109 and 1,862 *Ae. albopictus* larvae were recovered from positive ovitrap set indoors and outdoors in both villages, respectively, with an indoor:outdoor ratio of 1:1.7. By contrast, during 2006 survey, a ratio of 1:0.52 was recorded.

DISCUSSION

In the previous mosquito survey done on Carey Island by Rudnick in 1968, the most salient observation was the complete absence of *Ae. aegypti*. Re-surveys were done in 2006 and 2012 to determine whether the invasion of *Ae. aegypti* had occurred on Carey Island, given that Carey Island now is no longer considered a remote area due to the rapid development occurring throughout the island region. The increment in the numbers of oil palm factories and the advancement in the basic facilities of the island region–including a concrete bridge and an SKVE connection between Carey Island and the mainland–have no longer restricted the movement of people. The facilities have drastically increased the mobility of humans and transportation into and out of the island

Table 1
Ovitrap index (OI), mean *n* of larvae per recovered ovitrap.

Locality	Year	Ovitrap placement	Ovitrap collected (*n*)	Ovitrap recovered (*n*)	Positive ovitrap (*n*)	Ovitrap index (OI) (%)	Larvae per recovered ovitrap (Mean *n*)	
							Ae. albopictus	*Ae. aegypti*
Malay Village	2012	Indoor	25	24	16	66.7	25.75±5.40	0
		Outdoor	25	25	21	84.0	31.08±6.40	0
Aboriginal Village		Indoor	25	24	15	62.5	20.46±5.00	0
		Outdoor	25	25	22	88.0	43.40±5.90	0
Malay Village	2006	Indoor	20	20	20	100.0	N/A	0
		Outdoor	20	20	20	100.0	N/A	0
Aboriginal Village		Indoor	20	20	20	100.0	N/A	0
		Outdoor	20	20	20	100.0	N/A	0

as well as creating many man made opportunities for *Aedes* mosquito breeding.

For *Aedes* surveillance purposes, we decided to use ovitrapping technique for collecting *Aedes* spp, as ovitrap surveillance has been reported as an effective and sensitive technique when compared to larvae survey, especially when the *Aedes* infestation rates are very low (Lee, 1992; Arunachalam *et al*, 1999). Again, many years after the last survey in 1968, the re-survey results in 2006 and 2012 indicated the complete absence of *Ae. aegypti* on Carey Island. Interestingly, despite the high abundance of the *Ae. albopictus*, which is one of the important vectors for chikungunya virus (CHIKV) and dengue in Malaysia, no chikungunya cases have been reported so far at this particular study site (Ministry of Health, Personal communication, 2012). By contrast, relatively low dengue cases were reported by the District Health Office, with 14 cases for a period of 10 years, averaging about one case annually.

Many ovitrap surveillance studies have reported that *Ae. aegypti* is an indoor breeder, while *Ae. albopictus* is the dominant outdoor breeder (Hawley, 1988; Sota *et al*, 1992; Lim *et al*, 2010). In 2006, the surveillance on Carey Island indicated that the ratio of indoor:outdoor for *Ae. albopictus* was 1:0.52 contradicting the finding that *Ae. albopictus* is the outdoor container breeders. In fact, there are a few studies reporting that *Ae. albopictus* are also found to breed indoors (Chen *et al*, 2006; Preechaporn *et al*, 2006; Lim *et al*, 2010). As this mosquito typically shows outdoor breeding behavior with the persistence of its larval and pupal stages in indoor containers over a long period, this suggests that *Ae. albopictus* is adapting to the indoor environment (Dieng *et al*, 2010). The change in the oviposition preference may be due to the complete absence of *Ae. aegypti*, or *Ae. albopictus* is able to oviposit freely in whatever containers available. The latter seemed to be the case when *Ae. albopictus* was found to oviposit more in outdoor containers than indoors in 2012.

The complete absence of *Ae. aegypti* from Carey Island is difficult to be explained, as the island is close to Port Klang, where *Ae. aegypti* was reportedly first found in 1908 and believed to be an introduced species (Cheong, 1968; Rudnick, 1986). All nearby areas in Port Klang have since reported the presence of *Ae. aegypti*, which had successfully established itself; with the exception of Carey Island. There could also be several possibilities: one could be that the *Ae. albopictus* population is so dominant that it will not allow small number of other *Aedes* to be established on this Island, as the spread and establishment of *Ae. albopictus* was associated with a reduction in the abundance and range of *Ae. aegypti* (Hobbs *et al*, 1991; Mekuria and Hyatt, 1995). In several regions of Malaysia, studies reported mixed infestation between the two species in the ovitraps deployed (Chen *et al*, 2006; Dieng *et al*, 2010; Lim *et al*, 2010).

Currently, Carey Island is still mainly palm oil plantation land, mainly owned by Sime Darby Plantations Sdn Bhd. According to the report of this major oil palm company on Carey Island, most of the agricultural pesticides used were weedicide (Glyphosate, Ally and Garlton) and the only insecticide used belonged to the organophosphate class of insecticides (Methamidophos) for control of an outbreak of bagworm (Ross, 2011). Perhaps, it could be possible that *Ae. aegypti* cannot tolerate these insecticides, because this species is generally known to resist pyrethroids. Nonetheless, the evolutionary, ecological, environmental, and epidemiological factors responsible for the long-term absence of *Ae. aegypti* on Carey Island remains to be elucidated. Factor(s) responsible for the complete absence of *Ae. aegypti* on Carey Island may provide clues for effective elimination of this species.

In conclusion, after many years after the first published work on mosquito sampling done on Carey Island in 1968, there has been no sign of *Ae. aegypti* invasion on the island. Therefore, information compiled from this survey will be useful for future study (that is, mark-release-recapture), which requires as a pre-requisite a potential site with a high population of *Ae. albopictus* and the absence of *Ae. aegypti*.

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