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Surveillance of *Aedes* mosquitoes in a university campus in Kuala Lumpur, Malaysia

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Abstract. Ovitrap surveillance was initiated for eight continuous weeks to determine the distribution and abundance of *Aedes* sp. mosquitoes in the University of Malaya campus, Kuala Lumpur, and the impact of meteorological conditions on the *Aedes* populations. Two study areas within the campus were selected: Varsity Lake and Seventh Residential College. The abundance of *Aedes* populations in Varsity Lake was indicated by ovitrap index (OI) which ranged from 60.00% – 90.00%. The mean number of larvae per ovitrap of *Aedes albopictus* in Varsity Lake ranged from 11.23 ± 2.42 – 43.80 ± 6.22. On the other hand, the outdoor OI for Seventh Residential College ranged from 73.33% – 93.33%, respectively, while the mean number larvae per ovitrap for this area ranged from 19.33 ± 4.55 – 35.27 ± 5.46, respectively. In addition, the indoor OI of Seventh Residential College ranged from 0.00% - 30.00%, while the mean number of larvae per ovitrap for *Ae. albopictus* ranged from 0 – 5.90 ± 3.55. There was no significant difference ($p > 0.05$) of *Ae. albopictus* population between Varsity Lake and Seventh Residential College. The studies showed a correlation between OI and mean number of larvae per ovitrap for outdoor *Ae. albopictus* populations in Varsity Lake and Seventh Residential College ($r = 0.794$). There was also a correlation between the mean larvae number per ovitrap of *Ae. albopictus* obtained from eight weeks indoor ovitrap surveillance in Seventh Residential College with rainfall ($r = 0.584$). However, there was no correlation between the mean larvae number per ovitrap of *Ae. albopictus* in both study areas with temperature and relative humidity. *Aedes aegypti* mosquitoes were found neither indoor nor outdoor in both study areas. This study indicated that the principal dengue vector in the university campus was most likely *Ae. albopictus*.

INTRODUCTION

Dengue fever (DF) and dengue haemorrhagic fever (DHF) are the most important vector-borne diseases in tropical, subtropical and temperate regions of the world (Gubler *et al.*, 1998). WHO (2002) estimated that around 50 million DF and DHF infections occur in tropical and subtropical regions each year. The first major national DF and DHF outbreak in Malaysia occurred in 1973 (Lee, 1994). Since then, both diseases continued to be endemic in Malaysia.

Aedes aegypti (Linnaeus) and *Aedes albopictus* Skuse have been incriminated as the vectors involved in these infections (Rebecca, 1987; Lam, 1993; Lee & Inder, 1993; Nogueira *et al.*, 1999; Chen *et al.*, 2005). *Ae. aegypti* is an urban mosquito that has adapted to utilizing man-made containers (flower pots, small cisterns, discarded tyres and cans) for breeding. It feeds primarily on humans (Christophers, 1960; Cheong, 1967; Lee & Cheong, 1987). According to Perich *et al.* (2000), *Ae. aegypti* rests in secluded locations inside homes such as under beds, in closets and on curtains. In contrast, *Ae. albopictus* which breeds in both man-made containers such as cans, tires and water jars; as well as in natural containers such as bamboo, bromeliads, coconut shells is more

cosmopolitan in its feeding habitats and rests both inside and outside homes, making control difficult. The distribution of *Ae. aegypti* and *Ae. albopictus* in Malaysia overlaps (Yap, 1975; Sulaiman *et al.*, 1991). Both species are adapting to urban and suburban areas (Chen *et al.*, 2006).

Ovitrap surveillance is the most common sampling method to monitor *Ae. aegypti* and *Ae. albopictus* populations through their egg-laying activities (Service, 1992). Ovitrap surveillance has been claimed to be a more effective and sensitive technique compared to the conventional larval surveys, especially when the *Aedes* infestation rates were very low (Lee, 1992a).

The main objective of this study was to determine the distribution and abundance of *Aedes* sp. mosquitoes in the campus of University of Malaya, Kuala Lumpur. In addition, the impact of meteorological conditions on the *Aedes* populations and the efficacy of ovitraps in monitoring *Aedes* populations as dengue vectors were also determined.

MATERIALS AND METHODS

Study areas

Ovitrap surveillances was conducted in 2 sites: Varsity Lake and Seventh Residential College, which are located in the campus of University of Malaya, Kuala Lumpur. University of Malaya is situated on a 750 acre of land, located at the southwest of Kuala Lumpur City Centre. The ecological description of the study sites is given in Table 1.

Ovitrap surveillance

Eight continuous ovitrap surveillance weeks were conducted in Varsity Lake and Seventh Residential College. Ovitrap as described by Lee (1992a) was used in this study. The ovitrap consists of a 300 ml plastic container with straight, slight tapered sides. The opening measures 7.8 cm in diameter, the base diameter is 6.5 cm, and the container is 9.0 cm in height. The outer wall of the container is coated with a layer of black oil paint. An oviposition paddle made from hardboard (10.0 cm x 2.5 cm x 0.3 cm) was placed diagonally into each ovitrap. Each ovitrap was filled with tap water to the level of 5.5 cm.

A total of 30 ovitraps was placed randomly around the Varsity Lake. However, a total of 30 ovitraps were placed outdoors and 10 indoors in the Seventh Residential College. In this study, "outdoor" refers to the outside of the building but confined to the immediate vicinity of the house, while "indoor" refers to those parts of the house under its roof (Lee, 1992b). Ovitraps in all sites were collected after five days and fresh ovitraps and paddles were replaced weekly.

Outdoor ovitrap surveillance in Varsity Lake and Seventh Residential College was conducted simultaneously, while indoor ovitrap surveillance in Seventh Residential College was conducted sequentially.

Identification of larvae

The collected ovitraps were brought back to the laboratory and the contents were poured into a plastic container, together with the paddle. Fresh water was added into the container and the larvae were allowed to

Table 1. Ecological description of study sites

Study site	Ecological description
Varsity Lake	• Located at the south of the university campus • Many student activities were conducted around the lake, such as jogging, canoeing, sports etc. • Trees and shrubs found in this site • The environment is generally clean
Seventh Residential College	• Located at the west of the university campus • Student hostel • 800 students living in 4 blocks of 4 storey flat • Trees, ornamental plants and shrubs found in the college • The college surrounded by vegetations • The environment is generally clean

hatch and colonize in the laboratory for another 9 days. The container was kept covered. A small piece (10 mm) of fresh cow liver was added into each container as larval food. The hatched larvae were subsequently counted and identified at 3rd instar. The larval numbers were recorded individually for each positive ovitrap.

Only hatched larvae were counted in this study, as the larval stage is closely associated with the actual field mosquito populations since not all eggs will hatch. Besides, mosquito populations are monitored in vector control programme to prevent any dengue occurrence. Therefore, greater concern should be focused on mosquito stages surviving into adults which have a potential to transmit dengue viruses. WHO (Focks, 2003) has suggested the use of pupal index instead of larval index, as the pupal stage is relatively closer to adult and has higher chances of emerging to adults. However, in real situation, it is difficult to use pupal index compared to larval index, as pupae are difficult to be found in the field compared to larvae (Lee HL, unpublished document).

Meteorological data

Rainfall, temperature and relative humidity data were obtained from the Malaysian Meteorological Department.

Data analysis

Data were analysed as follows:

(a) Ovitrap Index (OI), the percentage of positive ovitrap against the total number of ovitraps recovered for each ovitrap surveillance from each study site, and

(b) Mean number of *Ae. aegypti* and *Ae. albopictus* larvae per recovered ovitrap.

All level of significance was determined at $p = 0.05$ by using a statistical programme with student t-test and one way ANOVA (SPSS v10). The correlation analyses were Spearman rank-order correlations. The significant correlation was determined at $r \geq 0.5$.

RESULTS AND DISCUSSION

Table 2 describes the ovitrap index (OI) and the mean number larvae per ovitrap of *Ae. aegypti* and *Ae. albopictus* obtained from eight weeks outdoor ovitrap surveillance in Varsity Lake. The results showed the abundance of *Aedes* population in Varsity Lake with OI range of 60.00% – 90.00%. The mean number of larvae per ovitrap of *Ae. albopictus* ranged from 11.23 ± 2.42 – 43.80 ± 6.22; while *Ae. aegypti* was not available in this study area.

Table 2. Ovitrap index and mean number of larvae per ovitrap of *Ae. aegypti* and *Ae. albopictus* obtained from 8 weeks outdoor ovitrap surveillance in Varsity Lake

Ovitrap Surveillance	Ovitrap Index, %	Mean number ± SE larvae per ovitrap			
		Ae. aegypti	ANOVA	*Ae. albopictus*	ANOVA
Week 1	60.00	0.00 ± 0.00		11.23 ± 2.42	
Week 2	86.67	0.00 ± 0.00		43.80 ± 6.22	
Week 3	76.67	0.00 ± 0.00		22.53 ± 5.44	
Week 4	70.00	0.00 ± 0.00		28.57 ± 5.97	
Week 5	83.33	0.00 ± 0.00	F = 999.99	21.23 ± 3.93	F = 4.31
Week 6	86.67	0.00 ± 0.00	P < 0.05	32.53 ± 5.02	P < 0.05
Week 7	73.08	0.00 ± 0.00		22.04 ± 4.94	
Week 8	90.00	0.00 ± 0.00		17.63 ± 3.54	
Mean	78.30 ± 3.63	0.00 ± 0.00	–	24.94 ± 3.53	–

$p > 0.05$ = not significantly different
$p \leq 0.05$ = significantly different
SE = standard error

The OI and the mean number of larvae per ovitrap of *Ae. albopictus* obtained from eight weeks outdoor and indoor ovitrap surveillance in Seventh Residential College are described by Table 3. The OI which indicates the abundance of *Aedes* mosquitoes population in outdoor and indoor ranged from 73.33% – 93.33% and 0.00% – 30.00%, respectively. The results indicate that *Ae. aegypti* populations were not found either outdoors or indoors in Seventh Residential College. However, the mean number of larvae per ovitrap of *Ae. albopictus* obtained outdoors and indoors in Seventh Residential College ranged from 19.33 ± 4.55 – 35.27 ± 5.46 and 0 – 5.90 ± 3.55, respectively.

In comparison with *Ae. albopictus* populations obtained from Varsity Lake and Seventh Residential College within the study period, there was no significant difference between the populations in both study area (Table 4). The study showed a significant correlation between OI and mean number of larvae per ovitrap of outdoor *Ae. albopictus* populations in both study sites ($r = 0.794$, $p < 0.05$) (Figure 1).

The *Ae. albopictus* population survey throughout all eight weeks ovitrap surveillance indicated a weekly variation of *Ae. albopictus* population ($p < 0.05$). This is in contrast to a study by Chen (2006) in that there was no weekly variation of *Ae. aegypti* and *Ae. albopictus* populations obtained from Taman Samudera and Kg. Banjar in Selangor, Malaysia.

Ae. aegypti was found neither indoors nor outdoors in both study areas. This finding is in contrast with the studies conducted by De Lima-Camara *et al.* (2006) who suggested that *Ae. aegypti* adults captured in urban areas preferred to rest inside houses and in areas with high human density; a behavior that favoured vector-human contact. However, the existence of indoor *Ae. albopictus* population in Seventh Residential College in this study supported studies by Ali *et al.* (2003) where they proved that *Ae. albopictus* was also present indoors. Moreover, the indoor condition of Seventh Residential College was generally clean, with minimal natural containers. Piped water supply is also available and thus, there is no need for the residents to store water. Therefore, it may not be the preferred breeding condition for *Ae. aegypti* mosquitoes.

The use of ovitraps is practical for monitoring populations of *Aedes* sp. (Masuh *et al.*, 2008). As previously described, more

Table 3. Ovitrap index and mean number of larvae per ovitrap of *Ae. albopictus* obtained from 8 weeks outdoor and indoor ovitrap surveillance in Seventh Residential College

Ovitrap Surveillance	Ovitrap Index, %		Mean number ± SE larvae per ovitrap			
	Outdoor	Indoor	Outdoor	ANOVA	Indoor	ANOVA
Week 1	73.33	30.00	19.33 ± 4.55		5.90 ± 3.55	
Week 2	76.67	10.00	29.83 ± 4.38		0.10 ± 0.10	
Week 3	88.46	0.00	24.11 ± 4.11		0.00 ± 0.00	
Week 4	80.00	0.00	26.73 ± 4.52		0.00 ± 0.00	
Week 5	53.33	0.00	19.93 ± 5.97	F = 1.00	0.00 ± 0.00	F = 2.75
Week 6	84.62	0.00	26.45 ± 6.05	P > 0.05	0.00 ± 0.00	P < 0.05
Week 7	80.00	0.00	26.87 ± 5.95		0.00 ± 0.00	
Week 8	93.33	0.00	35.27 ± 5.46		0.00 ± 0.00	
Mean	78.72 ± 4.28	5.00 ± 3.78	26.07 ± 1.83	–	0.75 ± 0.74	–

$p > 0.05$ = not significantly different
$p \leq 0.05$ = significantly different
SE = standard error

Table 4. Comparison of mean number larvae per ovitrap of *Ae. albopictus* obtained from 8 weeks outdoor ovitrap surveillance in Varsity Lake and Seventh Residential College

Ovitrap Surveillance	Mean number ± SE larvae per ovitrap		Level of significant
	Varsity Lake	Seventh Residential College	
Week 1	11.23 ± 2.42	19.33 ± 4.55	$p > 0.05$
Week 2	43.80 ± 6.22	29.83 ± 4.38	$p > 0.05$
Week 3	22.53 ± 5.44	24.11 ± 4.11	$p > 0.05$
Week 4	28.57 ± 5.97	26.73 ± 4.52	$p > 0.05$
Week 5	21.23 ± 3.93	19.93 ± 5.97	$p > 0.05$
Week 6	32.53 ± 5.02	26.45 ± 6.05	$p > 0.05$
Week 7	22.04 ± 4.94	26.87 ± 5.95	$p > 0.05$
Week 8	17.63 ± 3.54	35.27 ± 5.46	$p < 0.05$
Mean	24.94 ± 3.53	26.07 ± 1.83	$p > 0.05$

$p > 0.05$ = not significantly different
$p \leq 0.05$ = significantly different
SE = standard error

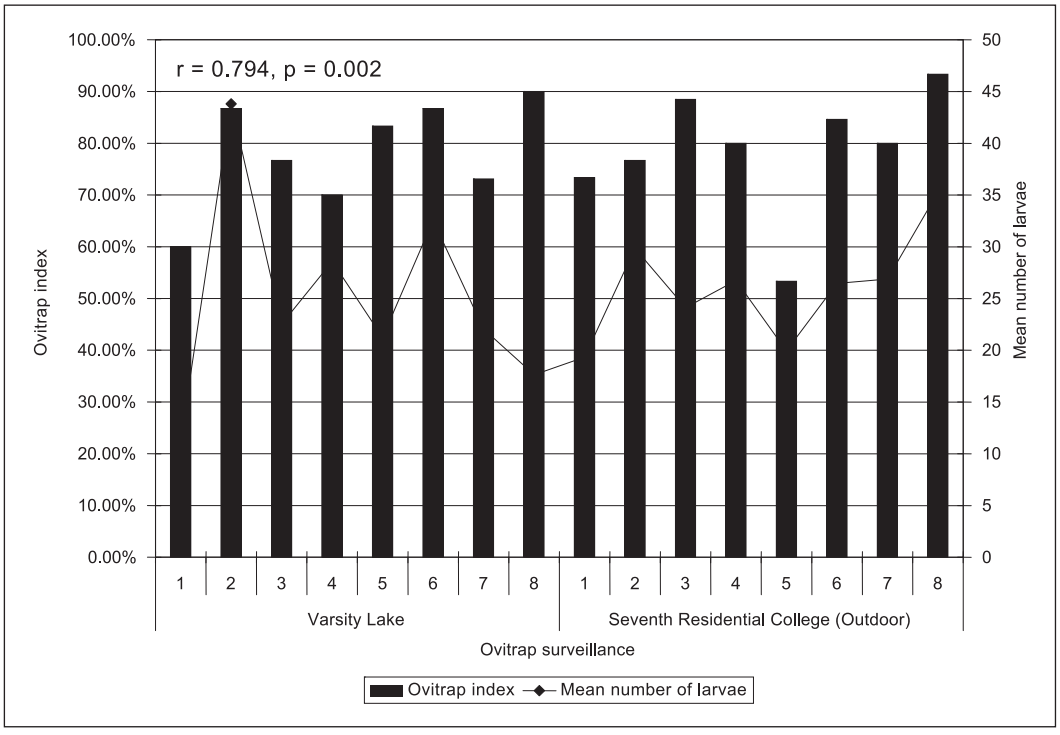

Figure 1. Correlation between ovitrap index and mean number of *Ae. albopictus* larvae collected from Varsity Lake and Seventh Residential College.

than 90.00% of outdoor OI was recorded using 30 ovitraps in every study area chosen in this study. Moreover, up to 30.00% of indoor OI was presented with the use of only 10 ovitraps in Seventh Residential College. These findings supported previous studies

worldwide which also showed that ovitrap is a sensitive tool in detecting the vector populations in nature. As such, local studies by Rozilawati *et al.* (2005) indicated an OI of 40.00% – 100.00% and 66.00% to 100.00% in untreated and deltamethrin-treated areas, respectively. In addition, Romero-Vivas & Falconar (2005) also reported that the mean ovitrap premise index (OPI) was 98.2% for an urban area in Colombia compared to the mean larval premise index (LPI) of the same area with only 59.2%. These findings were in parallel with previous studies by Marques *et al.* (1993) who showed that in Brazil, the ovitrap was more efficient than larval-traps and were positive even in the presence of natural breeding grounds. Not only that, Cardoso Junior *et al.* (1996) also reported that the ovitraps placed in Catanduva showed positiveness for *Ae. aegypti* two months after the control research, while Breteau Index became positive only at the fourth month after the end of the referred research. In fact, a strong correlation between OI and mean number of *Ae. albopictus* larvae collected from both areas presented in this study indicated that ovitrap surveillance conducted has provided a clear evidence on the presence and distribution of potential dengue vectors in the study areas.

Aedes aegypti, in particular, is highly adapted to human settlements (Gubler, 1988; Rodhain & Rosen, 1997); while *Ae. albopictus* is commonly found outdoors and breeds in all types of natural containers (Foo *et al.*, 1985; Sucharit *et al.*, 1978). However, studies by Chiaravalloti-Neto *et al.* (2002) showed that *Ae. albopictus* was found in greater proportions close to dwellings and presented greater degrees of association in natural and discarded containers, compared to *Ae. aegypti*. Their studies support our results where the mean number of larvae per ovitrap of outdoor and indoor *Ae. albopictus* populations obtained were higher compared to *Ae. aegypti* in both study areas.

Furthermore, the availability of natural potential breeding sites such as bamboo tree, banana tree, tree holes and pandan leaves (*Pandanus* sp.) in both study areas contributed to the high density of *Ae. albopictus*. The unmanaged rubbish and dry leaves available in Varsity Lake also encouraged the breeding of *Aedes* mosquitoes as the sites which contained sufficient nutrition were likely to support larval development (Strickman & Kittayapong, 2003).

Chakravarti & Kumaria (2005) suggested that analysis of three climatic factors such as rainfall, temperature and relative humidity were very important as these factors could affect the mosquito breeding activities. Our studies also showed that the mean number of *Ae. albopictus* larvae was significantly correlated with rainfall ($r = 0.584$, $p < 0.05$) (Figure 2). However, there was no significant correlation between the mean number larvae per ovitrap of *Ae. albopictus* obtained from eight weeks indoor and outdoor ovitrap surveillance in both study areas with the temperature ($r = 0.153$, $p > 0.05$) (Figure 3) and relative humidity ($r = -0.162$, $p > 0.05$) (Figure 4). Okogun *et al.* (2003) reported that rainfall is an important factor which regulates the abundance of outdoor breeding mosquito populations. Beside this, the wet seasons are associated with higher prevalence levels of mosquito-borne diseases (Okogun *et al.*, 2003). Rozilawati *et al.* (2007) also found that there was a strong correlation between rainfall and egg population of *Ae. albopictus* in Malaysia. Gubler *et al.* (2001) suggested that most of vector-borne diseases exhibit a distinctive seasonal pattern and climatic factors such as rainfall, temperature, and other weather variables are claimed to affect both the vector and the pathogen they transmit in many ways. Therefore, four climatic variables were tested as well in this study in both study areas. According to Tong *et al.* (2002), relative humidity influences the longevity, mating, dispersal, feeding behavior and oviposition of mosquitoes. They also claimed that mosquitoes generally survive longer and disperse further at high humidity.

In conclusion, since no *Ae. aegypti* mosquitoes were captured in both study areas, thus *Ae. albopictus* was the most likely principal dengue vector in the respective areas. Besides that, this study also proved that ovitrapping technique is still a reliable

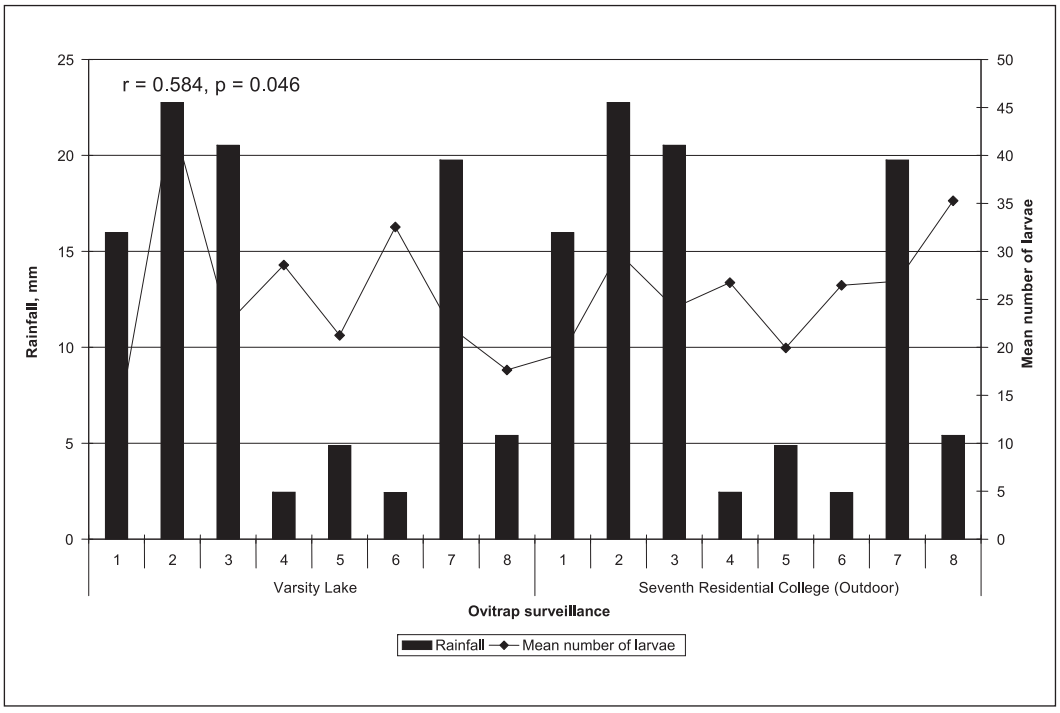

Figure 2. Correlation between rainfall and mean number of *Ae. albopictus* larvae collected from Varsity Lake and Seventh Residential College.

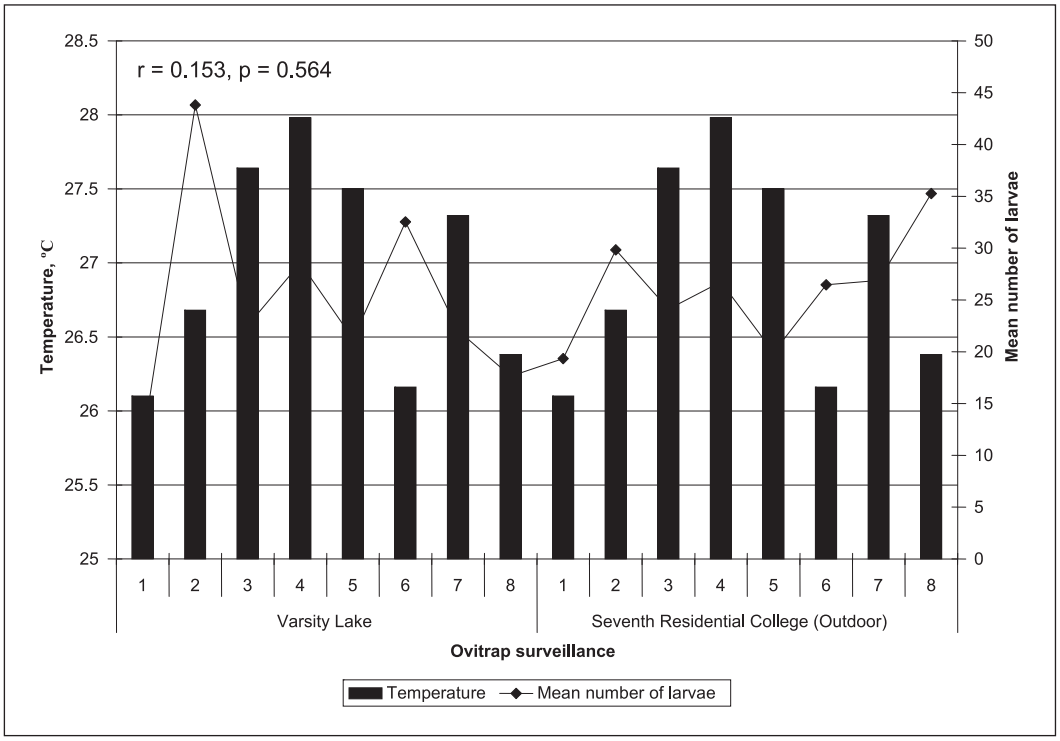

Figure 3. Correlation between temperature and mean number of *Ae. albopictus* larvae collected from Varsity Lake and Seventh Residential College.

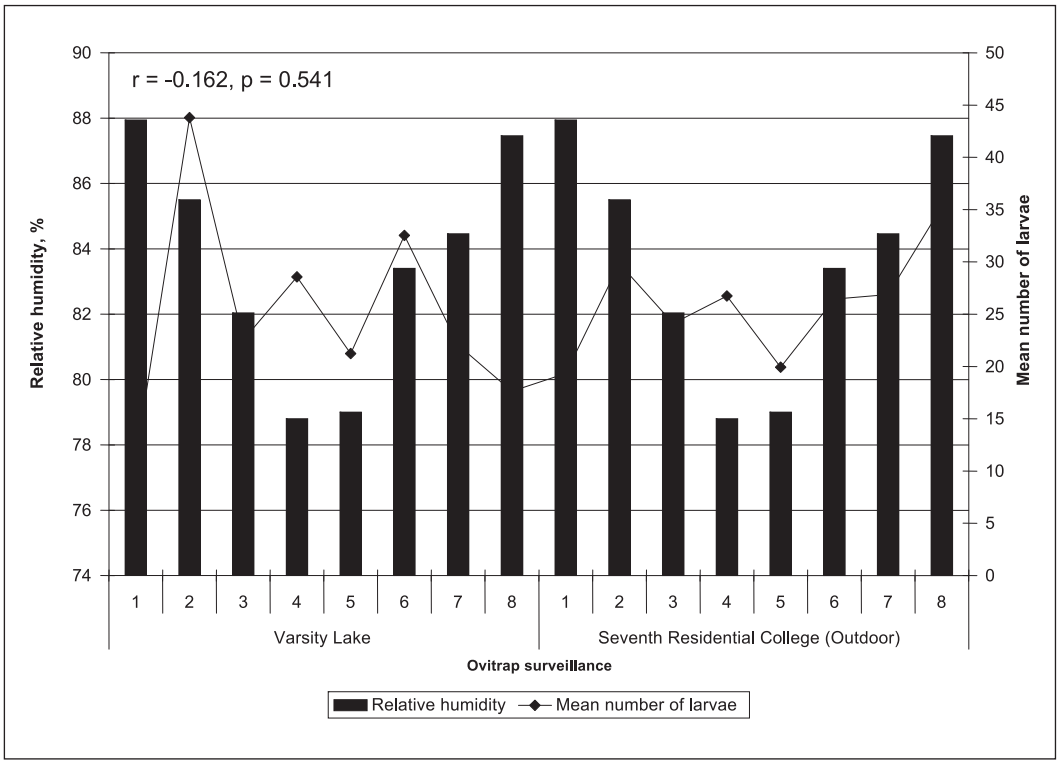

Figure 4. Correlation between relative humidity and mean number of *Ae. albopictus* larvae collected from Varsity Lake and Seventh Residential College.

and sensitive tool for early detection of dengue vectors in natural environment in comparison with larval survey.

Integrated vector management (IVM) such as source reduction, surveillance studies, insecticide application, biological control, education and public awareness as well as personal protection should be implemented in the campus in order to monitor and control the populations of both dengue vectors within the campus. Routine adulticiding should also be carried out in the university campus to suppress the *Aedes* populations, especially when the ovitrap index is 10% or more (Lee, 1992b).

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