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# Ovitrap surveillance in Sarawak, Malaysia: A comprehensive study

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**Abstract.** This study reports the distribution and abundance of *Aedes* by using ovitrap surveillance and aims to provide the most recent information on dengue vector distribution in Sarawak State, Malaysia. The ovitrap index (OI) of *Aedes* larvae was found highest in urban residential area (mean OI = 90.97%), followed by suburban (69.70%), rural (65.45%) and remote (52.63%) residential areas. The mean number of *Aedes* larvae per ovitrap was also found to be significantly highest in urban residential area (26.47 ± 1.62) compared to other type of residential areas ($p<0.05$). Interestingly, no *Aedes aegypti* was observed in this study, but two species of *Armigeres* were found co-breeding with *Ae. albopictus*. This study reveals that *Ae. albopictus* is the dominant dengue vector in Sarawak State and all the surveyed residential areas are in risk of dengue transmission with OI > 10%.

## INTRODUCTION

Dengue and dengue haemorrhagic fever (severe dengue) remain to be an endemic infectious disease and a serious public health problem in Malaysia. Both *Aedes*–borne diseases are frequently reported in peninsular Malaysia and Borneo (Sabah and Sarawak) since the first nationwide outbreak in 1973 (Hii, 1977; Lee & Hishamudin, 1990; Chang *et al*., 1981). The dengue outbreak in Sarawak occurred for the first time in 1982 and since then, it has become public health concern (Chang & Jute, 1994). The recent report by Ministry of Health Malaysia showed that total accumulated reported cases of dengue until week 50 of year 2016 in Malaysia was 98,438, of which 2,688 cases were reported in Sarawak, representing an increase by 43.74% compared to 2015 (MOH, 2016).

The two major vectors involved in these infections are *Aedes aegypti* and *Aedes albopictus*. Both species were known to adapt well to urban and suburban areas where their larvae breed in artificial and natural containers near human dwellings (Chen *et al*., 2006). The presence of *Ae. aegypti* in Sarawak was first reported in Sibu by Macdonald *et al*. (1965) and subsequently in Kuching and Miri (Macdonald *et al*., 1967; Macdonald & Rajapaksa, 1972). A subsequent investigation was done by Chang and Jute in 1982 where 73 localities in seven divisions of the state were surveyed. Apart from these earlier investigations, no up-to-date information on the distribution pattern and population of both *Aedes* species in Sarawak is available from the state until the present day.

An ovitrap surveillance is the commonest sampling method to monitor *Aedes* mosquito populations (Service, 1992). According to Lee (1992), an ovitrap surveillance has been shown to be a more effective and sensitive technique especially when the *Aedes* infestation rates were low.

The aim of this study is to provide the latest information on the distribution of *Aedes* species in different types of residential areas in Sarawak, which could help in the local vector control programme, as well as supplementing earlier reports.

## MATERIALS AND METHODS

**Geographical description of study sites**
An ovitrap surveillance was conducted in 21 residential areas across 13 districts located in 8 divisions in Sarawak, Malaysia. The geographical and ecological description of the study sites are given in Table 1. The 21 residential areas were categorized according to their landscapes into urban, suburban, rural and remote areas.

**Ovitrap surveillance**
The ovitrap as described by Lee (1992) was used in this surveillance. Each ovitrap consisted of a 300 ml black plastic container with 6.5 cm in diameter, 9.0 cm in height and the opening measures 7.8cm in diameter. The outer wall of the container was coated with a layer of black oil paint. Fresh water was added to a level of 5.5 cm and an oviposition paddle made form hardboard (10 cm x 2.5 cm x 0.3cm) was placed diagonally with the rough surface upwards into each ovitrap.

Ovitraps were placed in not less than 10% of the houses in all residential areas. The ovitraps were placed outside the house but confined to the immediate vicinity of the house, i.e. car porch and corridor under the eave. The houses were chosen randomly.

**Identification of larvae**
The ovitraps were collected after 5 days and transported back to laboratory and the contents were poured individually into a labelled plastic container, together with the paddle. Overnight water (tap water exposed for 24–48 hr before using) was added into the container and a small piece (10mm) of fresh beef liver was added as larval food. The hatched larvae were subsequently counted and 3$^{rd}$ instar-larvae were identified to species level according to the key by Mahadevan & Cheong (1974). The larval numbers were recorded individually for each positive ovitrap.

**Data analysis**
All data obtained from this study was analysed as follow:

(1) Ovitrap Index (OI), the percentage of positive ovitrap against the total number of ovitraps recovered from each side.

(2) Mean number of larvae per recovered ovitrap.

All levels of statistical significance were determined at $P \leq 0.05$ by using statistical programme, student t-test and one-way ANOVA (SPSS® version 21.0; IBM, Armonk, NY).

## RESULTS

Table 2 shows the ovitrap index (OI) and the mean number of larvae per ovitrap of *Ae. albopictus* and *Armigeres* sp. obtained from 21 residential areas across 13 districts in Sarawak. All residential areas were categorized into urban, suburban, rural and remote according to their landscapes as shown in Table 1. *Aedes albopictus* was present in all localities with the OI ranging from 35.00% to 100% and mean number of larvae per ovitrap ranged from 2.74 ± 1.15 to 29.41 ± 6.64.

Comparisons between OI according to landscapes show that the OI of the urban residential area was significantly higher than rural, suburban and remote residential area ($p < 0.05$) with mean OI at 90.97 ± 1.59%, 69.76 ± 8.34%, 65.91 ± 3.88% and 52.63 ± 15.79%, respectively. In addition, significantly highest *Ae. albopictus* mean number of larvae per ovitrap was obtained from urban residential areas (26.47 ± 1.62) compared to rural areas (14.73 ± 2.95), suburban areas (13.55 ± 2.22)

Table 1. Geographical description of study sites in Sarawak, Malaysia

| Division | District | Study Sites | Coordinates | Elevation (meter a.s.l.) | Type of residential area |
|---|---|---|---|---|---|
| Kuching | Kuching | Lorong Siol Kandis | N 1º 34’ 31.8"; E 110º 21’ 50.6" | 5 | Urban |
| | | Petra Jaya | N 1º 34’ 43.0"; E 110º 20’ 25.8" | 7 | Urban |
| | Bau | Kampung Atas | N 1º 28’ 44.7"; E 110º 11’ 11.5" | 46 | Remote |
| | | Kampung Apar | N 1º 28’ 30.8"; E 110º 08’ 55.7" | 20 | Remote |
| Samarahan | Samarahan | Kampung Merdang Gayam | N 1º 27’ 34.4"; E 110º 24’ 59.5" | 14 | Rural |
| | | Kampung Merdang Lumut | N 1º 27’ 02.5"; E 110º 23’ 52.6" | 7 | Rural |
| | | Kampung Bukit Brangan | N 1º 26’ 40.4"; E 110º 23’ 16.7" | 17 | Suburban |
| | Serian | Kampung Melayu Tebakang | N 1º 12’ 27.8"; E 110º 33’ 57.2" | 19 | Suburban |
| Sibu | Sibu | Kiew Nang | N 2º 15’ 47.4"; E 111º 51’ 48.6" | 8 | Suburban |
| | Selangau | Pekan Selangau | N 2º 31’ 24.1"; E 112º 19’ 33.2" | 25 | Rural |
| Mukah | Mukah | Kampung Kuala Lama | N 2º 53’ 44.4"; E 112º 05’ 33.4" | 4 | Suburban |
| | | Bandar Baru Mukah | N 2º 53’ 34.0"; E 112º 05’ 33.9" | 3 | Suburban |
| | Dalat | Pekan Dalat | N 2º 44’ 20.7"; E 111º 56’ 14.9" | 6 | Rural |
| Miri | Miri | Kampung Siwa Jaya | N 4º 13’ 40.7"; E 113º 54’ 59.3" | 22 | Rural |
| | | Bandar Miri | N 4º 28’ 02.4"; E 114º 00’ 15.6" | 6 | Urban |
| | | Lutong, Kg. Tulang | N 4º 22’ 59.2"; E 113º 58’ 56.9" | 9 | Suburban |
| Bintulu | Bintulu | Kemena Jaya | N 3º 10’ 26.6"; E 113º 02’ 53.9" | 11 | Suburban |
| | | JKR Quarters | N 3º 10’ 42.4"; E 113º 02’ 55.3" | 19 | Suburban |
| | Tatau | Kampung Dagang Tatau | N 2º 52’ 33.7"; E 112º 51’ 12.6" | 7 | Rural |
| Sarikei | Sarikei | Pekan Sarikei | N 2º 07’ 40.9"; E 111º 31’ 7.85" | 3 | Suburban |
| Kapit | Kapit | Pekan Kapit | N 2º 01’ 1.11"; E 112º 56’ 14.9" | 18 | Rural |

Table 2. Ovitrap index and mean number of larvae per ovitrap (mean ± S.E.) of mosquito larvae collected from 21 study sites across Sarawak, Malaysia

| Study Sites | Landscape | OI (%) | Mean OI | Mean *Aedes albopictus* larvae per ovitrap | | Mean *Armigeres* larvae per ovitrap | |
|---|---|---|---|---|---|---|---|
| | | | | Each study site | Type of residential area | Each study site | Type of residential area |
| Bandar Miri, Miri | Urban | 90.91 | | 23.82 ± 3.31 | | | |
| Lorong Siol Kandis, Kuching | Urban | 88.24 | 90.97 ± 1.59 | 29.41 ± 6.64 | 26.47 ± 1.62 | | |
| Petra Jaya, Kuching | Urban | 93.75 | | 26.19 ± 6.11 | | | |
| Bandar Baru Mukah, Mukah | Suburban | 45.00 | | 4.25 ± 1.32 | | | |
| JKR Quarters, Bintulu | Suburban | 60.00 | | 15.47 ± 4.63 | | | |
| Kampung Bukit Brangan, Samarahan | Suburban | 73.68 | | 10.26 ± 2.22 | | | |
| Kampung Kuala Lama, Mukah | Suburban | 57.89 | | 13.05 ± 6.35 | | | |
| Kemena Jaya, Bintulu | Suburban | 72.73 | 69.70 ± 5.10 | 18.14 ± 4.81 | 13.55 ± 2.22 | | 0.11 ± 0.11 |
| Kiew Nang, Sibu | Suburban | 78.95 | | 12.84 ± 2.87 | | | |
| Lutong, Kg. Tulang, Miri | Suburban | 70.59 | | 7.71 ± 2.21 | | | |
| Pekan Sarikei, Sarikei | Suburban | 68.42 | | 12.63 ± 3.27 | | | |
| Kampung Melayu Tebakang, Serian | Suburban | 100.00 | | 27.64 ± 2.78 | | 0.97 ± 0.41 | |
| Kampung Dagang Tatau, Tatau | Rural | 47.37 | | 11.00 ± 4.04 | | | |
| Kampung Merdang Gayam, Samarahan | Rural | 87.50 | | 26.63 ± 7.24 | | | |
| Kampung Merdang Lumut, Samarahan | Rural | 80.00 | | 17.40 ± 9.46 | | 0.25 ± 0.16 | |
| Kampung Siwa Jaya, Miri | Rural | 63.64 | 65.46 ± 8.23 | 16.23 ± 4.29 | 14.73 ± 2.95 | | 0.06 ± 0.04 |
| Pekan Dalat, Dalat | Rural | 35.00 | | 4.20 ± 1.64 | | | |
| Pekan Kapit, Kapit | Rural | 92.31 | | 20.50 ± 4.10 | | | |
| Pekan Selangau, Selangau | Rural | 52.38 | | 7.14 ± 2.55 | | 0.18 ± 0.18 | |
| Kampung Atas, Bau | Remote | 68.42 | 52.63 ± 15.79 | 11.37 ± 4.49 | 7.06 ± 4.32 | 1.08 ± 0.60 | 0.90 ± 0.19 |
| Kampung Apar, Bau | Remote | 36.84 | | 2.74 ± 1.15 | | 0.71 ± 0.47 | |
| | | | p = 0.043 | | p = 0.026 | | p = 0.003 |

and remote areas (7.06 ± 4.32) ($p < 0.05$). There difference in larval numbers was significant among all residential areas ($p \leq 0.05$).

*Aedes aegypti* was not detected throughout the surveillance. On the other hand, *Armigeres* sp. was found co-breeding with *Ae. albopictus* from 5 residential areas, namely Kampung Melayu Tebakang (District: Serian, Division: Samarahan), Kampung Merdang Lumut (Samarahan, Samarahan), Pekan Selangau (Selangau, Sibu), Kampung Atas (Bau, Kuching) and Kampung Apar (Bau, Kuching) with mean larval number per ovitrap ranging from 0.18 ± 0.18 to 1.08 ± 0.60 (Table 2).

Further analyses of comparisons between OI and mean larval number per ovitrap according to landscapes are shown in Table 3 and Table 4, respectively. There was significant difference of OI between urban and other landscapes, but no significant difference of OI between suburban, rural and remote residential areas (Table 3). Table 4 reveals a significant difference between mean larval number per ovitrap obtained from urban and other landscapes; however, no significant difference between suburban, rural and remote residential areas was observed, indicating that density of the *Ae. albopictus* in urban residential areas were higher than other residential areas and distributed well with high OI observed in urban residential areas.

Table 5 shows mixed breeding of *Aedes albopictus* and *Armigeres* spp. larvae in residential areas in Sarawak. The percentage of mixed breeding ranged from 9.09 to 38.46%. The numbers of *Ae. albopictus* larvae were found 2.38 – 71.00 times higher than those of *Armigeres* sp. in mixed breeding ovitraps.

## DISCUSSION

*Aedes albopictus* is widespread, as detected in all localities in this study. Our results indicate that *Ae. albopictus* was more abundant in urban residential area and the density was significantly higher in urban area than those of other categories of residential areas, with mean ovitrap index 90.97 ± 1.59% and mean number of larvae per ovitrap 26.47 ± 1.62.

The differences in OI and mean number of larvae per ovitrap of *Ae. albopictus* between landscapes can possibly be the results of geo-physical and socio-environmental set up of the residential area with reference to location and basic amenities (Chang & Jute, 1982). According to Chang & Jute (1982), the density of *Ae. albocpictus* was higher in coastal and rural areas and comparatively low in urban and suburban areas due to the absence of basic amenities and the consequential water storage activities in coastal and rural area which in turn become breeding grounds for *Ae. albopictus*. The condition of recent rural areas is different from those reported by Chang & Jute (1982) 34 years ago. Road, communication, water supply and garbage disposal system have been since improved, and an effective vector control programme is now actively implemented by local authorities, thus reducing water storage activities and the number of breeding grounds for *Ae. albopictus*. However, still in several rural and remote areas, the lack of basic amenities has led to indiscriminate disposal of garbage and many water holding containers were still used widely, similarly as reported by Chang & Jute (1982).

Table 3. Comparison of mean ovitrap index (OI) between landscapes

| p value | Urban | Suburban | Rural | Remote |
|---|---|---|---|---|
| Urban | – | 0.043 | 0.021 | 0.049 |
| Suburban | – | – | 0.654 | 0.212 |
| Rural | – | – | – | 0.488 |

Table 4. Comparison of mean number of larvae per ovitrap between landscapes

| p value | Urban | Suburban | Rural | Remote |
|---|---|---|---|---|
| Urban | – | 0.010 | 0.039 | 0.015 |
| Suburban | – | – | 0.751 | 0.240 |
| Rural | – | – | – | 0.248 |
| Remote | – | – | – | – |

Table 5. Mixed breeding of *Aedes albopictus* and *Armigeres* spp. larvae in residential areas in Sarawak

| Study sites | Collected Ovitrap | Positive ovitrap | | Mixed breeding ovitrap | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | n | % | n | % | Ovitrap No. | Number of Larvae: *Ae. albopictus* | Number of Larvae: *Armigeres* spp. | Ratio of *Ae. albopictus* : *Armigeres* spp. |
| Kampung Atas, Bau | 19 | 13 | 68.42 | 5 | 38.46 | 1 | 3 | 1 | 3.00 : 1.00 |
| | | | | | | 2 | 71 | 1 | 71.00 : 1.00 |
| | | | | | | 3 | 53 | 2 | 26.50 : 1.00 |
| | | | | | | 4 | 13 | 1 | 13.00 : 1.00 |
| | | | | | | 5 | 21 | 8 | 2.63 : 1.00 |
| Kampung Melayu Tebakang, Serian | 36 | 36 | 100.00 | 4 | 11.11 | 1 | 45 | 7 | 6.43 : 1.00 |
| | | | | | | 2 | 18 | 6 | 3.00 : 1.00 |
| | | | | | | 3 | 49 | 8 | 6.13 : 1.00 |
| | | | | | | 4 | 19 | 8 | 2.38 : 1.00 |
| Kampung Merdang Lumut, Samarahan | 10 | 8 | 80.00 | 2 | 25.00 | 1 | 8 | 1 | 8.00 : 1.00 |
| | | | | | | 2 | 41 | 1 | 41.00 : 1.00 |
| Pekan Selangau, Selangau | 21 | 11 | 52.38 | 1 | 9.09 | 1 | 5 | 2 | 2.50 : 1.00 |
| Kampung Apar, Bau | 19 | 7 | 36.84 | 1 | 14.29 | 1 | 15 | 3 | 5.00 : 1.00 |

*Aedes albopictus* is well known as a semidomestic breeder in urban areas where it feeds on humans and domestic animals and oviposits in natural and artificial water containers near human dwellings (Hawley, 1988). Heavy vegetation was observed around the urban areas and a variety of man-made breeding grounds for *Ae. albopictus*, such as plastic rubbish and water ditches yielded by urban activities was also observed.

*Aedes aegypti* was previously reported in Sibu (Macdonald *et al.*, 1965), Kuching (Macdonald *et al.*, 1964; Surtees, 1970) and Miri (Macdonald & Rajapaksa, 1972). A survey done by Chang and Jute (1982) in 1980 found that *Ae. aegypti* was present in 37 localities out of 73. Interestingly, no *Ae. aegypti* was recovered from this study. Chang & Jute (1982) also reported that *Ae. aegypti* had been eliminated in 5 urban localities after 3 years of vector control programme since 1978. Chan *et al.* (1971) reported that *Ae. aegypti* breeds predominately inside houses while *Ae. albopictus* breeds mainly outside houses. Most of the control programme targets indoor areas due to intensive malaria vector control in the past 3 decades (Tee, 2000), whereas the outdoor breeding behavior of *Ae. albopcitus* might have increased their survival when they were hidden in the inner deep of heavy vegetation where control application hardly reached. In the long run, the population of *Ae. aegypti* was lowered and thus *Ae. albopictus* become dominant in urban area due to the absence of interspecies competition in outdoor breeding sites. The lack of proper means of transportation from urban to other areas could also affect the migration of *Ae. aegypti* in the past 3 decades (Chang & Jute, 1982) and this might be the reason why the populations of *Ae. aegypti* were unable to spread while suppressed by the control programme. Barrera (1992) reported that *Ae. albopictus* could withstand starvation longer than *Ae. aegypti* when reared on oak leaves, in other word, the heavy coverage of vegetation around the residential area favors the *Ae. albopictus* populations. With all the factors may explain why *Ae. albopictus* become a dominant species in urban residential areas. In suburban and rural areas, the distribution is somewhat similar although ovitrap index in urban area was higher. Both residential areas share the similarity of geo-physical and socio-environment factors such as water supply, shop lots and residential. The human population and activities which provide more food source and favorable habitats for *Ae. albopictus* contribute to higher OI in suburban residential area than rural and remote residential area.

Larvae of *Armigeres* spp. were also found co-breeding with *Ae. albopcitus* in 5 residential areas. *Armigeres kesseli* and *Armigeres subalbatus* are commonly found close to human dwellings and may adapt to breeding habitats similar to *Aedes* mosquitoes such as artificial containers, coconut shells, hollow bamboos and mostly polluted water (Pandian & Chandrashekaran, 1980; Rajavel, 1992, Nurin-Zulkifli *et al.*, 2015). The larvae of *Armigeres* spp. are voracious biter that had been reported to be predacious (Buddle, 1928) as well as cannibalistic (Rajavel, 1992). The presence of *Armigeres* sp. increases the interspecies competition as well as the predation on the *Ae. albopictus* larvae.

The factors contributing to the failure of establishment of *Ae. aegypti* in all residential areas when compared to data reported by Chang & Jute (1982) are not fully understood. The absence of *Ae. aegypti* was also previously reported in an university campus and an island of peninsular Malaysia (Norafikah *et al.*, 2009; Chen *et al.*, 2009; Lau *et al.*, 2013; Noor-Afizah *et al.*, 2015). This phenomenon was probably due to lack of favorable breeding foci of *Ae. aegypti* (Norafikah *et al.*, 2009; Chen *et al.*, 2009; Lau *et al.*, 2013). Noor-Afizah *et al.* (2015) suggested that other *Aedes* species was prevented from establishing themselves because the population of *Ae. albopictus* was so dominant, as the establishment of *Ae. albopictus* was associated with reduction in the abundance and range of *Ae. aegypti*. In our study, *Ae. albopictus* has been able to establish itself well better than *Ae. aegypti* in all residential areas. In other words, *Ae. albopictus* is the dominant vector and incriminated for the transmission of dengue fever. Moreover, all the surveyed residential areas are in high risk of dengue transmission where OI was more than 10% (Tham, 2000). Ovitrap surveillance is a key component of any local integrated vector management to quantify human risk to dengue fever by determining the presence of local vector and abundance. Thus, local authorities should implement ovitrap surveillance frequently and carry out more effective integrated vector management (IVM) to prevent dengue fever outbreak. Public awareness and usage of personal protection measures against mosquitoes should be promoted in order to reduce the exposure to mosquito bites.

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