SPATIAL DISTRIBUTION OF MOSQUITO VECTOR IN DENGUE OUTBREAK AREAS IN KUALA LUMPUR AND SELANGOR, MALAYSIA

Wan Najdah Wan Mohamad Ali^{1,2}, Rohani Ahmad¹, Zurainee Mohamed Nor^{2*}, Thuraiya Abdul Rahman¹ & Yvonne Ai-Lian Lim²

> ¹Medical Entomology Unit,
> Institute for Medical Research, National Institute of Health, Ministry of Health Malaysia,
> 50588 Kuala Lumpur, Malaysia.
> ²Department of Parasitology, Faculty of Medicine, University of Malaya,
> 50603 Kuala Lumpur, Malaysia.
> *Corresponding author: *zuraineemn@um.edu.my*

ABSTRACT

In Malaysia, the control of dengue is mainly through the identification and reduction of mosquito vector breeding sites. In this study, a larval survey was conducted from June 2017 until December 2018 to determine the spatial distribution of dengue vectors in the 132 dengue hotspots outbreak areas in Kuala Lumpur and Selangor. Molecular methods were performed in order to detect the presence of transovarial dengue virus in larvae collected, while the density of the breeding habitat and *Aedes* larval population were determine using spatial analysis. Map of Dengue virus (DENV) distribution were generated to illustrate the trend of dengue outbreak. This study showed that larval survey was an effective method to detect the presence of dengue virus transmission in immature Aedes aegypti and Aedes albopictus. This study also demonstrated that plastic container was the highest source of breeding habitat for Aedes mosquito, whereas blocked drain and tyre were the most favourable breeding habitats for Ae. aegypti and Ae. albopictus, respectively. Pearson's correlation coefficient shows that mosquito density was not correlated with the DENV infection. In conclusion, current study shows that dengue transmission risk in Kuala Lumpur and Selangor remain high despite the outbreak response conducted by the health authority due to high density of Aedes population and the presence of DENV infection within the larvae population in the area. Therefore, new outbreak response methods such as public mandatory involvement in community-based control program to ensure success in management of resource reduction are necessary to ensure that the risk of dengue infection can be eliminated.

Keywords: Aedes distribution, dengue vector, dengue hotspots, spatial analysis

ABSTRAK

Di Malaysia, kaedah kawalan denggi yang utama adalah pengenalpastian dan penghapusan tempat pembiakan nyamuk vektor. Dalam kajian ini, tinjauan larva dilakukan dari bulan Jun 2017 hingga Disember 2018 untuk menentukan taburan spasial vektor denggi di 132 kawasan

wabak hotspot denggi di Kuala Lumpur dan Selangor. Kaedah molekular digunakan untuk mengesan kehadiran virus denggi (DENV) transovarial pada larva yang dikumpul. Sementara itu, kepadatan habitat pembiakan dan populasi larva Aedes ditentukan menggunakan kaedah analisis spasial. Peta taburan DENV dihasilkan untuk menunjukkan trend wabak denggi. Kajian ini menunjukkan bahawa tinjauan larva adalah kaedah yang berkesan untuk mengesan kehadiran penularan DENV di dalam larva Aedes aegypti dan Aedes albopictus. Kajian ini juga menunjukkan bahawa bekas plastik merupakan sumber habitat pembiakan nyamuk Aedes yang paling tinggi. Habitat pembiakan yang paling digemari oleh *Ae. aegypti* adalah longkang yang tersumbat manakala tayar merupakan habitat pembiakan yang paling digemari oleh Ae. albopictus. Ujian Korelasi Pearson menunjukkan bahawa kepadatan nyamuk tidak berkorelasi dengan jangkitan DENV. Kesimpulannya, kajian ini menunjukkan bahawa risiko penularan denggi di Kuala Lumpur dan Selangor tetap tinggi walaupun tindak balas wabak telah dilakukan oleh pihak kesihatan kerana kepadatan populasi Aedes yang tinggi dan adanya jangkitan DENV dalam populasi larva di kawasan tersebut. Oleh yang demikian, kaedah penularan wabak yang baru seperti penglibatan wajib masyarakat dalam program kawalan berasaskan komuniti untuk memastikan kejayaan dalam pengurusan pengurangan sumber diperlukan untuk memastikan risiko jangkitan denggi dapat dihapuskan.

Kata kunci: Taburan Aedes, vektor denggi, hotspot denggi, analisis spasial

INTRODUCTION

Dengue has become one of the fastest growing mosquito-borne diseases in the world (WHO 2010). Globally, dengue infections constitute a significant public health burden (Jahan et al. 2016). The four serotypes of dengue virus (DENV) (Flaviviridae: genus *Flavivirus*) are responsible for up to 50 million cases of dengue fever (DF) annually and approximately 500, 000 cases progress to the more severe type, dengue haemorrhagic fever (DHF). Infection by one serotype unfortunately does not confer immunity to the other three, and sequential infections may predispose one to developing DHF (Bhoomiboonchoo et al. 2015; Edelman 2011).

In recent decades, Malaysia has become a dengue hyper-endemic country with the cocirculation of the four dengue serotypes (Jahan et al. 2016). The dengue situation in Malaysia has worsened with an increasing number of reported cases and death during the last decade (Mia et al. 2013). In 2019, there were 130, 101 dengue cases with 182 death (MOHM 2020). To date the control of the disease have been limited to mosquito abatement, as no licensed vaccines or antivirals are available (Anderson et al. 2006).

The virus is transmitted to humans through the bite of infected female *Ae. aegypti* and *Ae. albopictus* (Chen et al. 2004; Rohani et al. 1997; Rohani et al. 2007). Female mosquitoes remain infectious for their entire lives and have the potential to transmit virus during each human feeding. In Malaysia *Ae. aegypti* and *Ae. albopictus* are generally found in two different geographical categories, suburban and urban. They are found mostly near the vicinity of human habitation, as the adult mosquitoes need humans' blood for their meals (Lee 1992). *Ae. aegypti* can easily found at developing area with less vegetation (Chen et al. 2006) and tends to breed in variety of assorted water-holding containers found in and around homes (Lenhart et al. 2005) while *Ae. albopictus* known as an outdoor species, lives in vegetated area (Chen et al. 2006) and breeds in artificial and natural containers near human dwellings (Saleeza et al. 2011).

Dengue outbreaks were partly attributed to localized increases of *Aedes* mosquitoes in the dengue outbreak areas (Messina et al. 2019; Rohani et al. 2011; Rohani et al. 2018). The increasing trend of dengue highlights the need for a more systematic surveillance and reporting of the disease (Mia et al. 2013). Vector surveillance provides estimates of population densities and viral infection rates which are necessary to predict epidemics of dengue and implementing remedial measures (Rohani et al. 1997).

A major problem pose by dengue is the high number of non-detectable breeding sites. This problem however is about to be resolved. With advances in satellite technology the resolution of spatial imagery is likely to increase, where it allows the estimation of additional predictors to appraise where humans and vectors interact (Louis et al. 2014). Thus, dengue risk map can be powerful tools to facilitate decision making in public health, ranging from surveillance to prediction maps. The objective of this study was to determine the spatial distribution of both *Ae. aegypti* and *Ae. albopictus* using larval survey method and to generate map of dengue transmission risk in dengue hotspot areas in Kuala Lumpur and Selangor, Malaysia.

MATERIALS AND METHODS

Study Area and Collection Sites

Entomological surveys were conducted from June 2017 until December 2018 in 132 dengue hotspots localities across Kuala Lumpur and Selangor, Malaysia (Figure 1). Kuala Lumpur is one of the territories under the Federal Territories of Malaysia and the capital city of Malaysia. Selangor meanwhile is one of the states in Malaysia that comprises of nine districts as follows: Sabak Bernam, Hulu Selangor, Kuala Selangor, Gombak, Petaling, Hulu Langat, Kuala Langat, Klang, and Sepang. All localities included in the study were coded according to locality number (Loc. No.) and categories into three geographical categories: urban, suburban or rural (Mohd Syariefudin et al. 2016; Samruhaizad et 2014: (Appendix A) al. https://www.iqiglobal.com/blog/3-common-myths-of-urban-suburban-areas/). Geographical category classification based on Department of Statistics Malaysia. The selection of the localities was based on constant occurrence of dengue cases obtained from idengue web (http://idengue.arsm.gov.my/) published by Malaysian Space Agency and Vector Borne Disease Control Programme, Ministry of Health Malaysia (iDengue 2018).

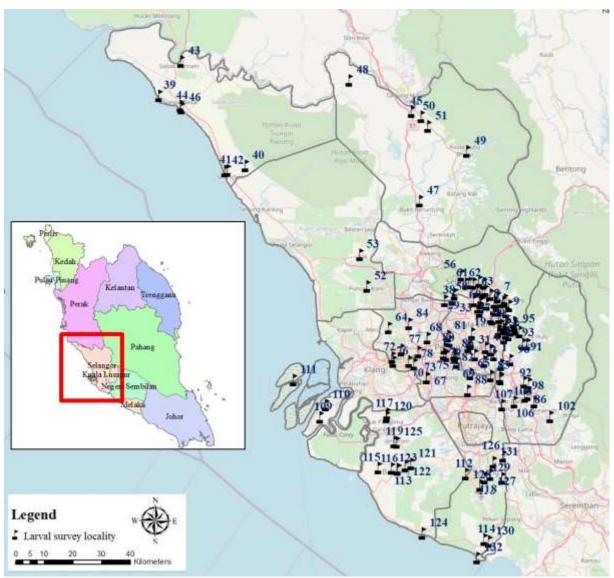


Figure 1. Map of the 132 study localities in Kuala Lumpur and Selangor

Larval survey

A house to house and peridomestic area survey was carried out in order to detect mosquito larval breeding sites. Two groups of surveyors which consist of four persons in each group were deployed to conduct larval survey in each locality starting from 5.00 pm to 7.30 pm. All potential breeding sites found within the premises compound in the sampling areas were visually inspected for the presence of mosquito larvae and pupae using torchlight. Among potential breeding sites anticipated include tin cans, tires, earthen jars or any other containers that can trap water. Water from containers was poured into white plastic trays. When *Aedes* mosquito larvae are present, larvae were collected by using standard dipping method World Health Organization (WHO 1975). Then, larvae were collected by means of a pipette and transferred to a bottle and properly labelled. Care is taken to ensure that the larvae and pupae collected remain alive and undamaged until arrival at the insectarium.

Larvae Identification

The larvae and pupae collected were transported to the insectarium. The larvae and pupae were colonized in the plastic trays at 28°C and 80% relative humidity. Partially cook liver was served

to the larvae. The pupated larvae were collected from the plastic tray using a pipette and transferred into a small plastic container before placed in the cage for emerging. Ten percent glucose supplemented with 1% vitamin B complex was supplied to the newly emerged adult mosquitoes. The adults were identified using standard taxonomic keys (Jefry et al. 2012). The adult mosquitoes were then pooled according to habitat type and locality with maximum 20 adults per pool in 1.5 ml micro centrifuge tube and then stored at -80°C fridge until used.

Detection of Dengue Virus (DENV)

The previously pooled mosquitoes were then assayed for DENV detection. Briefly 210μ l of nuclease-free, double-distilled water was added to the tube and the mosquito were homogenized on ice using a homogenizer attached to a Pellet Pestle Motor (Kontes, Japan). The homogenized samples were then centrifuged at 5000 x g for 10 minutes at 4°C. QIAmp Viral RNA Mini Kit (Qiagen) was used to extract the viral RNA from the mosquito homogenates according to the manufacturer's guidelines. Extracted RNA was kept at -80°C until further used.

For the detection of DENV, reverse transcriptase-polymerase chain reaction (RT-PCR) universal (5'employed using the dengue primers of D1 was (5'-TCAATATGCTGAAACGCGCGAGAAACCG-3') forward and D2 for TTGCACCAACAGTCAATGTCTTCAGCTTC-3') for reverse. The protocols and primers described by Lanciotti et. al (1992) were used with slight modification. The PCR product size with 511bp will appear on the in 1.5% agarose gel.

Data Analysis

All data obtained from this study was analysed for the distribution of *Aedes* mosquito collected by species, types of breeding containers and the *Aedes* spp. occurrence, mean larvae population: Total larvae population divide to total breeding habitat, Minimum infection rate (MIR): Number of positive pools divide to total number of adult female tested times 1000.

Findings of the survey were then statistically analysed using Statistical Package for Social Science (SPSS) version 23. The normality of the distribution was determined using a Kolmogorov-Smirnov goodness-of-fit test for variables. The test indicated that all the scores did fit a normal distribution. Thus, parametric test was performed for statistical analyses. Pearson correlation analysis was conducted in order to determine the significant correlation within the variables. T-test and Two-way ANOVA analysis test were conducted in order to determine the significant different within the variables.

Spatial Analysis

The coordinates of habitat that contain mosquito larvae were marked using a hand-held Geographic Positioning System (GPS), [Garmin GPSMAP[®] 60CSx] and processed and built in ArcMap 10.1 GIS (ESRI). Basic digital administrative boundaries map of state of Kuala Lumpur and Selangor was free downloaded from <u>https://gadm.org/download_country_v3.html</u> and used as a base map. Coordinate system used in digital maps was WGS1984. Coordinates of habitat containing mosquito larvae were later integrated to a GIS to quantify spatial heterogeneity in the associated area.

Spatial density map of *Aedes*-positive breeding sites, *Ae. aegypti* and *Ae. albopictus* and total *Aedes* larvae population in dengue hotspots areas in Kuala Lumpur and Selangor were created using ArcMap 10.1 program and Arc toolbox by spatial analyst tools of point density. Area with highest density is indicated by red colour, medium high density by orange colour,

medium density by yellow colour, medium low density by light green colour and the low density by green colour.

Geodatabase Dengue Transmission Risk

Based on the density of *Aedes*-positive breeding site, *Ae. aegypti* and *Ae. albopictus* larval population, the level of dengue transmission risk in the study areas were determined by spatial analyst tools of point density method.

RESULTS

Distribution of *Aedes* **Populations**

Throughout this study, 1,040 potential breeding sites were identified and a total of 585 were confirmed as *Aedes*-positive larval habitats. The *Aedes*-positive habitats were then classified into two classes: (a) artificial breeding site and (b) natural breeding site. The artificial breeding sites consist of six types: 1) aluminium containers (cans, metal drum, and cooking pots), 2) concrete tanks (clay jars, vase, discard sink and toilet bowl), 3) glass containers (broken glasses and discard aquarium), 4) plastic containers (bottle, basins, buckets or pails and canvas), 5) polystyrene containers and 6) tyres. The natural breeding site consist of three types: 1) non-manmade (plant part, discarded coconut shells, ground pool and tree hole), 2) blocked drain and 3) blocked gutter. Because drains and gutters are building structures, mosquito breeding that occurs is natural and not due to human action, thus blocked drain and blocked gutter were categorized as a natural breeding site. Out of nine types of breeding site, plastic containers were the highest number of breeding sites recorded (33.85%), followed by concrete tanks (15.90%), polystyrene containers (11.62%), non-manmade (10.94%), tyres (9.40%), aluminium containers (7.69%), glass containers (5.13%), blocked drain (3.59%) and blocked gutter (1.88%).

Only two *Aedes* species, *Ae. albopictus* and *Ae. aegypti* were captured during this study. The distribution of the two *Aedes* species in relation to habitat type were compared (Figure 2). It was demonstrated that not only *Ae. albopictus* shown to be present in all habitat types but it was also found occupying larger number of individual breeding site than *Ae. aegypti* except for blocked drain. *Ae. aegypti* were also found present in all habitat types but the number of each habitat harbouring them were less than *Ae. albopictus*. Larvae of *Ae. albopictus* and *Ae. aegypti* were found coexisted in 134 out of 585 habitats which involve almost all habitat types except blocked gutter habitat. It is interesting to note that in tyre and plastic container habitat types, the number of each breeding habitat that have both species co-existed were more that those that harbour *Ae. aegypti* alone.

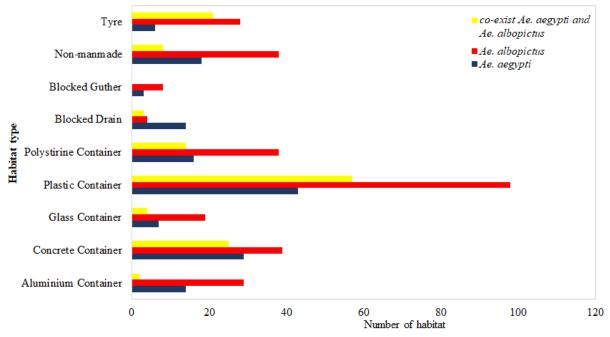


Figure 2. Distribution of *Ae. aegypti* and *Ae. albopictus* larvae based on number of each habitat types

Table 1 shows the number of *Ae. aegypti* and *Ae. albopictus* larval population in relation to the habitat types. The total number of *Ae. albopictus* larvae collected during the survey was almost double (4,438) to that of *Ae. aegypti* (2,454) as the number of *Ae. albopictus* larval population was found higher in almost all habitat types than *Ae. aegypti*, except in the blocked drain and their number was almost equal in natural habitat type.

Table 1.	Mean of larvae collected from each habitat type for Ae. aegypti and Ae.
	albopictus

Habitat Type		Ae. aegypti	Ae. albopictus		
Habitat Type	n	Mean±SE	F	Mean±SE	F
Aluminium container	45	1.93±0.49 ^a	9.22**	7.38 ± 2.42^{ab}	3.88**
Concrete container	93	5.15±0.76 ^a		6.55 ± 0.90^{a}	
Glass container	30	3.67 ± 0.92^{a}		$9.80{\pm}2.19^{ab}$	
Plastic container	198	3.22 ± 0.34^{a}		7.83 ± 0.72^{a}	
Polystyrene container	68	2.34±0.41 ^a		5.57 ± 0.67^{a}	
Blocked drain	21	13.62±2.81 ^b		2.71 ± 1.13^{a}	
Blocked gutter	11	3.27 ± 2.06^{a}		10.55 ± 4.61^{ab}	
Non-manmade	64	3.38 ± 0.68^{a}		5.67 ± 0.73^{a}	
Tyre	55	$4.60{\pm}1.06^{a}$		13.40 ± 1.86^{b}	

Values followed by different letters within a column for each species are significantly different (Two-way ANOVA followed by Tukey test, P < 0.05)

The difference in the number of larval populations between *Ae. aegypti* and *Ae. albopictus* in relation to each habitat types were then statistically analysed, and they were found to be significantly different for all habitat types [aluminium container (t_{88} = -2.24, P<0.05), glass container (t_{58} = -2.59, P<0.05), plastic container (t_{394} = -5.829, P<0.05), polystyrene

container t_{134} = -4.09, P<0.05), blocked drain (t_{40} = 3.61, P<0.05), blocked gutter (t_{20} = -1.440, P<0.05), non-manmade (t_{124} = -2.30, P<0.05) and tyre (t_{108} = -4.11, P<0.05) habitat type] except concrete container habitat (t_{184} = -1.19, P>0.05).

The mean of larvae collected from each habitat types for *Ae. aegypti* and *Ae. albopictus* were shown in Table 1. For *Ae. aegypti*, the mean of larvae collected was found highest in blocked drain (13.62±2.80) and lowest in aluminium container (1.93±0.49). The difference in mean of larvae collected between habitat types for *Ae. aegypti* was found to be significantly different ($F_{8, 576}$ = 9.22, P<0.01) between blocked drain and eight other habitat types. As for *Ae. albopictus*, tyre habitat type shows the highest mean of larvae collected (13.40±1.86) while blocked drain shows the lowest (2.71±1.13). For *Ae. albopictus*, the difference in mean of larvae collected between habitat types indicated that tyre habitat type means of larvae collected was significantly different with concrete container, plastic container, polystyrene container, blocked drain and natural breeding habitat (F_{8, 576}= 3.88, P<0.01).

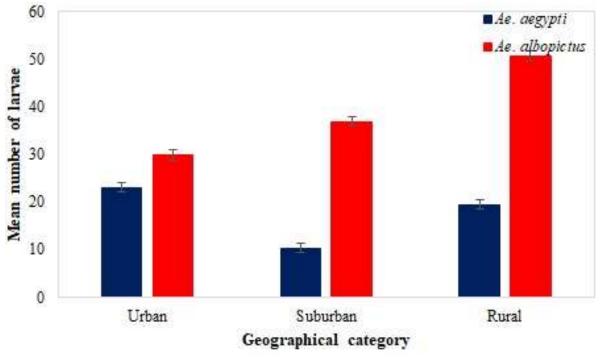


Figure 3. Summary of mean of *Ae. aegypti* and *Ae. albopictus* collected from 3 geographical categories

Figure 3 shows the mean number of larvae for *Ae. aegypti* and *Ae. albopictus* collected from three geographical categories (urban, suburban and rural). *Ae. aegypti* mean of larvae collected was the highest for urban category (23.10 ± 3.70) followed by rural category (19.44 ± 10.96) and then suburban category (10.32 ± 4.96) while for *Ae. albopictus*, rural category (50.67 ± 10.96) shows the highest mean of larvae collected followed by suburban category (36.89 ± 4.96) and urban (29.87 ± 3.70) category. Two-way ANOVA test performed demonstrated significant difference within interactions of geographical categories and *Aedes* species towards number of larvae collected ($F_{5, 258}=4.37$, P<0.05).

Spatial Density of *Aedes*-positive Breeding Sites, *Aedes* Larval Population and Dengue Transmission Risk

Figure 4 is the map developed to illustrate spatial density of the *Aedes*-positive breeding sites in Kuala Lumpur and Selangor. The map clearly shows that the localities with densest *Aedes*positive breeding sites were located mainly in Kuala Lumpur. All together there were 31 localities out of 38 hotspot localities identified in Kuala Lumpur that have densest *Aedes*positive breeding habitats. In Gombak district there were nine localities out of 10 hotspot localities listed while in Hulu Langat district there were 10 localities out of 22 hotspot localities. Other districts were Petaling with five localities out of 22 hotspot localities, Klang with one locality out of three hotspot localities, Kuala Langat with six localities out of 14 hotspot localities and Sepang with three localities out of seven hotspot localities (Appendix A).

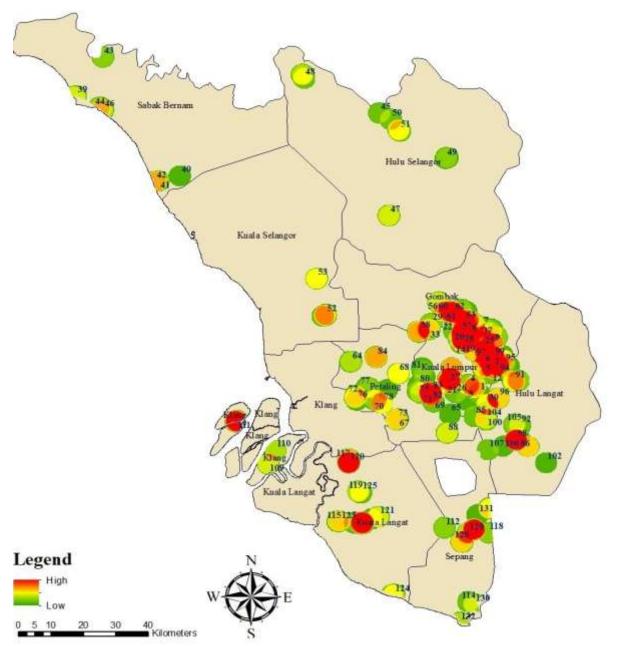


Figure 4. Map showing spatial density of *Aedes* breeding habitat in Kuala Lumpur and Selangor

The spatial density map for *Aedes* larval population were developed and presented as Figure 5a (for *Ae. aegypti*) and Figure 5b (for *Ae. albopictus*) and detailed in Appendix A. Both maps clearly show that the densest larvae population for both species were located in Kuala Lumpur. Out of 31 localities with densest *Aedes*-positive breeding sites, 25 localities had breeding habitats highly populated with *Aedes* larvae. Ten localities were having breeding sites highly populated with *Ae. aegypti* larvae and three localities having breeding sites populated by both species. With nine localities in Gombak district identified as densest *Aedes*-positive breeding sites, two localities were found having breeding sites which were heavily populated by *Ae. aegypti* larvae and one locality had their breeding sites populated by *Ae. albopictus* larvae of both species. Another district in Selangor that demonstrated highest density of the *Aedes*-positive breeding sites is Petaling district with five localities. Two of the localities were found to have their breeding habitats highly populated by *Ae. albopictus* larvae of both species. Two of the localities were found to have their breeding sites is Petaling district with five localities. Two of the localities were found to have their breeding habitats highly populated by *Ae. albopictus* larvae and while breeding habitats highly populated by *Ae. albopictus* larvae of both species.

As for Klang district which had only one locality with densest *Aedes*-positive breeding site, the species inhabiting the breeding sites at this location was mainly *Ae. aegypti*. With 10 densest *Aedes*-positive breeding sites detected in Hulu Langat district, breeding sites at Loc. No: 98 was found highly populated by *Ae. aegypti* larvae, two localities were found highly populated with *Ae. albopictus* larvae, while in seven other localities their breeding habitats were populated by both species.

With six densest *Aedes*-positive breeding sites, Kuala Langat district had five localities had breeding habitats highly populated with *Aedes* larva. Four of those localities where their breeding sites were highly populated with *Ae. aegypti* and *Ae. albopictus* larva while one locality was found to have breeding habitats with high population of *Ae. albopictus* larvae. The final district that demonstrated localities with densest *Aedes*-positive breeding sites was Sepang district. There were three localities involved. Only two localities had breeding habitats highly populated with *Aedes* larva and both were found having breeding sites that consist high population of *Ae. albopictus* larvae.

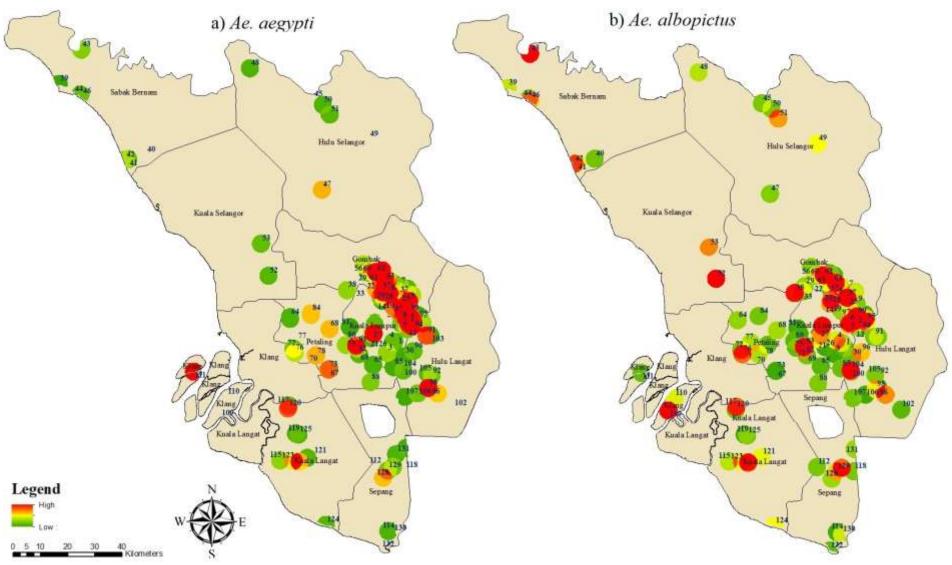


Figure 5. Map showing spatial density of (a) *Ae. aegypti* and (b) *Ae. albopictus* larvae population in Kuala Lumpur and Selangor

Based on the density of *Ae. aegypti and Ae. albopictus* larval population, the dengue transmission risk for all study localities were determined and presented in Appendix A. Figure 5 is the map that depicts the spatial density of dengue transmission risk in Kuala Lumpur and Selangor. Based on Figure 6, 61 localities with highest dengue transmission risk were mainly located in Kuala Lumpur involving 27 localities, nine localities in Gombak district, seven localities in Petaling district, eight localities in Hulu Langat district, five localities in Kuala Langat district, two localities in Sepang district, and one locality each in Klang, Sabak Bernam and Kuala Selangor district.

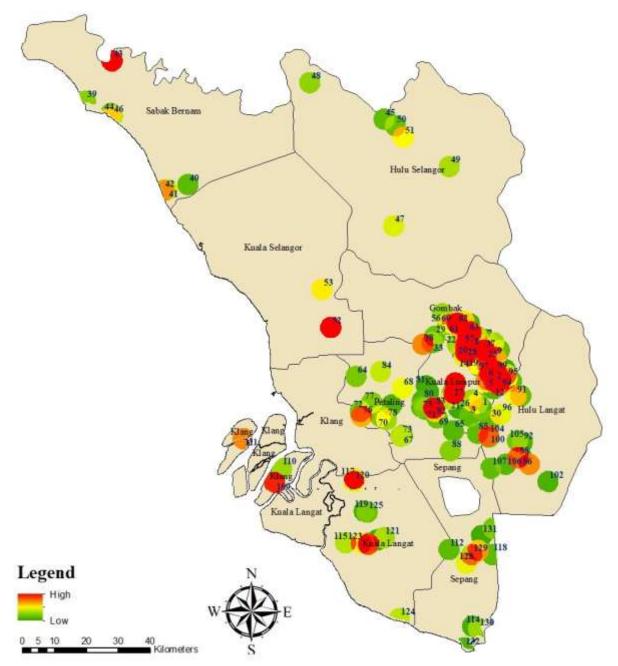


Figure 6. Map of spatial distribution of dengue transmission risk in Kuala Lumpur and Selangor

Based on the above findings, this study observes situation where not all localities that have breeding habitat with densest *Aedes* larval population, demonstrated high dengue transmission risk. For examples, in Hulu Langat district there were 10 localities that had breeding habitats highly populated with *Aedes* larvae, but only eight localities were demonstrated as having high dengue transmission risk. Another situation observed was, localities found not having breeding habitat densely populated with *Aedes* larvae, were showing high dengue transmission risk. Klang district for example, the one locality (Loc. No: 111) with densest *Aedes* larval population, was not found having highest dengue transmission risk but demonstrated by another locality (Loc. No: 109) instead. Similarly, in Petaling district, previously identified as having five localities having breeding habitats highly populated with *Aedes* larvae, was found to have seven localities (including the five listed earlier) demonstrated highest dengue transmission risk.

Distribution of DENV Positive Pools

A total of 303 pools of adult mosquitoes were successfully collected from the study areas. All pools were tested and only 51 pools (16.83%) were confirmed positive for DENV. These 51 DENV-positive pools were those collected from 26 study localities. From the 26 localities, *Ae. aegypti*-positive pools were collected from six localities while *Ae. albopictus* pools were collected from 12 localities There were eight localities where both *Ae. aegypti* and *Ae. albopictus* pools were collected (Table 2).

Locality	Looolity	Number of Pos	Number of Positive DENV Pool				
Number	Locality	Ae. aegypti	Ae. albopictus				
1	Angkasa Condominium		1				
6	Blok M Pandan Jaya		1				
13	Flat Sect. 1 Wangsa Maju	1					
24	PPR Sri Pantai		1				
26	Tmn. Bahagia Kuchai		1				
37	Tmn. Sri Rampai		1				
44	Tmn. Berkat Sg. Besar	1					
53	Tmn. Cempaka Sari Ijok	1	2				
57	Kg. Changkat Greenwood		2				
58	Kg. Laksamana Gombak	2	1				
60	Selayang Baru		1				
68	Apt. Sri Meranti D'mansara	1					
70	Dataran Otomobil		1				
72	Flat Nilam Sari S7 S.Alam	1					
74	Flat Tmn. Dato Harun	1	2				
81	SS22 Damansara		1				
94	Flat Seri Nilam Ampang	1					
95	Flat Tmn. Dagang Permai		1				
98	Tmn. Bkt Mewah Kajang	1					
104	Tmn. Taming Jaya	1	2				
109	Apt. Samudera Pulau Indah		6				
113	Tmn. Aman Banting	2	1				
115	Tmn. Banting Baru	1	3				
117	Tmn. Perwira TPG	2	2				
120	Tmn. Seri Medan Jaya	3	1				
124	Tmn. Tanjong Sepat		1				
	Total	19	32				

Table 2Distribution of 51 DENV-positive pools collected 26 localities

Figure 7 is the map that illustrates the distribution of *Ae. aegypti* and *Ae. albopictus*-DENV positive pools in Kuala Lumpur and Selangor. Similar to maps developed earlier (*Aedes* breeding habitat, *Aedes* larva population and DENV transmission risk), this map also demonstrated that *Ae. aegypti* and *Ae. albopictus*-DENV positive pools were located mainly in Kuala Lumpur and its surrounding districts. Not many *Ae. aegypti* and *Ae. albopictus*-DENV positive pools were collected from localities farther from Kuala Lumpur. Based on Pearson correlation test, this study indicates that the density of *Ae. aegypti* and *Ae. albopictus* larva population was not significantly correlated with virus infectivity (r₉₉= -0.108, P= 0.282) (r₁₁₆= -0.149, P= 0.054) respectively.

Table 3 shows the Minimum Infection Rate (MIR) value for *Aedes* spp. collected in Kuala Lumpur and Selangor. Six DENV-positive pools that consist of one *Ae. aegypti* pool and five *Ae. albopictus* pools were collected from Kuala Lumpur while the other 45 DENV-positive pools consist of 18 *Ae. aegypti* pools and 27 *Ae. albopictus* pools were collected from Selangor. Based on these numbers, the MIR value of *Ae. albopictus* and *Ae. aegypti* mosquito for Kuala Lumpur and Selangor were determined. For Kuala Lumpur, *Ae. albopictus* mosquito was found to have MIR value which was five times higher (5.05) than *Ae. aegypti* (1.17). In contrast, *Ae. aegypti* was the one that showed higher MIR (11.24) which was almost two times higher than *Ae. albopictus* (7.83) for Selangor.

Aedes species	No. of mosquitoes	No. of pools	No. of positive pools (%)	MIR
Kuala Lumpur				
Ae. aegypti	853	71	1(1.41%)	1.17
Ae. albopictus	991	72	5(6.94%)	5.05
Selangor				
Ae. aegypti	1601	46	18(39.13%)	11.24
Ae. albopictus	3447	114	27(25.44%)	7.83

 Table 3.
 The number of DENV-positive mosquito pool and the MIR value for Kuala

 Lumpur and Selangor

Wan Najdah et al.

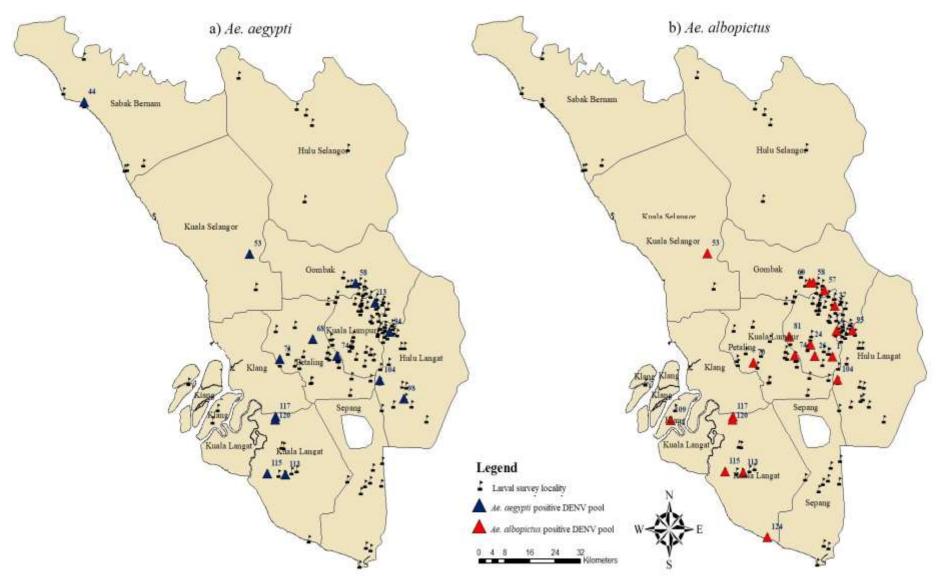


Figure 7. Distribution of a) *Ae. aegypti*-DENV positive pools and b) *Ae. Albopictus* DENV positive pools in Kuala Lumpur and Selangor

ISSN 1394-5130

DISCUSSION

There were 132 localities in Kuala Lumpur and Selangor that have been identified as hotspot areas for dengue by idengue web (*http://idengue.arsm.gov.my/*). This study examined the mosquito breeding habitat, larva population, dengue virus infectivity in larva population and the MIR value of two *Aedes* spp caught in these hotspot areas in order to establish localities with high dengue transmission risk and to determine the competent species in term of transmitting the virus. Several maps were developed to illustrate areas with densest breeding habitat, larva population and DENV-positive larva population to provide a more accurate picture of the actual dengue situation in the identified hotspot areas. Such information is useful in relation to implementation of effective dengue control program.

Out of 1,040 breeding habitats examined during the study, only half of them were detected positive for Aedes spp. and only two Aedes species were identified during the study, namely Ae. aegypti and Ae. albopictus. Both species are the main vectors and responsible for the transmission of DENV in Malaysia (Nazri et al. 2013a). The Aedes-positive breeding habitat determined in this study consist of nine types namely aluminium containers, concrete tanks, glass containers, plastic containers, polystyrene containers, tyres, non-manmade, blocked drain and blocked gutter. This study revealed that majority (83.59%) of Aedes-positive breeding sites were those that are artificial in nature (plastic containers, polystyrene container, concrete containers, tyre, aluminium container and glass container) and they are clearly the main contributors of mosquito breeding sites in these hotspot areas. This finding is in accordance with study by Rohani et. al. (2014) who reported that artificial breeding habitat contributes 93% of the Aedes larval habitat detected at dengue outbreak areas in Malaysia. Saleeza et. al. (2013) meanwhile reported that artificial breeding habitat contributes 88% of the Aedes larval habitat collected from Putrajaya, Malaysia. Although Putrajaya is a well-planned city, the presence of artificial breeding habitats in the area has led to the existence of Aedes population. Nonetheless, it has been proven that by regularly draining or eliminating the artificial containers have shown effectively decreasing the mosquito breeding grounds (MOHM 2015). What is needed is for the community not to simply throw garbage everywhere as they please and possibly organise regular clean-up campaign to maintain cleanliness in the area.

Out of nine habitat types listed in this study, plastic container was the highest distributor of larval habitat. This finding is supported by several studies performed at several states in Malaysia (Chen et al. 2009; Mohd Amierul et al. 2018; Rahim et al. 2018; Rohani et al. 2014; Rozilawati et al. 2015) and at other countries like India (Vijayakumar et al. 2014) and in Bangladesh (Islama et al. 2019). Plastic characteristics such as durability, easy recycle, light weight and low cost, have inspired manufacturers to produce products made from plastic for a wide variety of uses (Rahim et al. 2018). Thus, make plastic container among the most commonly used and discarded items in any community. Sadly, such item has been proven over and over by many studies as the major contributor of *Aedes* mosquito habitat and this study is no exception.

According to Paul et. al. (2018) different types of containers may serve differently as habitat for the production of immature mosquitoes. Several studies have reported that both *Aedes* spp. tend to display specific preference towards certain type of container for oviposition (Chatterjee et al. 2015; Faiz et al. 2017; Rohani et al. 2014). In this current study the highest mean of *Ae. aegypti* larvae was obtained from blocked drain which could indicate that this type of breeding habitat as preferred habitat for this species. Chen et al. (2005), has stated that

stagnant clear water in drains serves as good artificial larval containers for *Ae. aegypti*. This study discovered that stagnant clear water caused by blocked drains were available in most of high-rise premises (flat and apartment) in the study localities.

As for Ae. albopictus, this study demonstrated that tyres may be its favourite habitat type. According to Reiter (1998), global trade in tyres plays a major role in Ae. albopictus expansion. History has proven that tyres being such a favourable breeding grounds despite of climate preference. After World War II a considerable amount of waste tyres from military vehicle and aircraft from US were left scattered in abroad countries. When the tires were returned to the US, a large number of Ae. albopictus were unintentionally shipped together from South Pacific and other countries. Despite of different climates, it managed to thrive in the US because of the tyres. 20 years later, Ae. albopictus was again transported from the Vietnam following Vietnam war and successfully established in the US (Pratt et al. 1946). A review studied by Yee (2008) on the tyres as habitats for mosquito summarized that 32 mosquitos' species have been documented as tyre-inhabited species and Ae. albopictus was the most abundant in the south of Eastern United States. Study by Rohani et al. (2001), in urban and rural areas in 12 states of Malaysia described that waste tyres are the well-known favourite breeding habitat for Ae. albopictus. Similarly, discarded tyre was reported as the most preferred breeding site for Ae. albopictus in Udaipur, India (Meena & Choudhary 2019) and in Dares Salaam (Philbert & Ijumba 2013).

Examining the population of the two species based on three geographical categories (urban, semi-urban and rural) demonstrated that *Ae. albopictus* was the dominant mosquito species compared to *Ae. aegypti* for all geographical categories. This finding was in accordance with several studies (Faiz et al. 2017; Nazri et al. 2013b; Rohani et al. 2014; Rozilawati et al. 2015; Saleeza et al. 2011). Individually, *Ae. aegypti* collected from the three geographical categories showed those collected from urban category was significantly higher compared to the one collected at rural and suburban categories. The fact that *Ae. aegypti* is an anthropohilic mosquito and highly adapted to the domestic environment, there is always a positive correlation between abundance and increasing urbanization (Higa 2011). In contrary, *Ae. albopictus* larvae collected from suburban and urban categories. Being an exophagic and exophilic mosquito, *Ae. albopictus* as pointed out by many researchers prefers to breed in containers surrounded by vegetation in rural (Kamgang et al. 2010; Rohani et al. 2014) and suburban areas (Ho et al. 2014; Rozilawati et al. 2014; Rozilawati et al. 2015).

For a control program that aims to eliminate *Aedes* population in an area to be successful, it is important to locate the area and this area has to be the one with high dengue transmission risk. By developing a map (Figure 6) the distribution of such area could be seen clearly where from the total of 132 hotspot areas only 61 were actually identified of having high dengue transmission risk and they were clearly located in Kuala Lumpur and its nearby districts.

This study also examined the occurrence of transovarial transmission of DENV at all study areas since transovarial transmission of *Aedes* larvae caught in wild has been reported before in Malaysia (Nor Aliza et al. 2019; Rohani et al. 2014). Based on 26 localities where transovarial transmission has occurred, attempt to see correlation between transovarial transmission and the density of larval population at study areas showed that they were not significantly correlated. This finding is in accordance with a study conducted in Yogyakarta City (Rahayu et al. 2019). Study by Pena-Garcia et al. (2016), in Colombian cities however

showed otherwise where low correlation between the mosquito density and the infected mosquito was observed. Nonetheless, the researcher did explain in his report that DENV transmission dynamics cannot be explained by mosquito density alone, since the mechanism includes a complex network of variables such as vector capacity, human immunity heterogeneity, and abiotic variables such as temperature.

It was revealed that transovarial DENV in wild larvae was associated with dengue outbreak (Lee & Rohani 2005) which means early detection of DENV through transovarial transmission therefore could actually be used as an indicator for dengue transmission in an area and urgent remedial surveillance steps should be conducted before imminent outbreak occurs. It is therefore very crucial for areas with high dengue transmission risk and with transovarial transmission DENV given priority for control program to be conducted compared to low risk areas to ensure successful preventive measures and the risk of the upcoming threat, rather than a reaction to an outbreak event (Hassan et al. 2012).

As mentioned earlier, map of dengue transmission risk for all hotspot areas (Figure 6) demonstrated the localities with high dengue transmission risk were mainly located in Kuala Lumpur and Gombak, followed by few localities in Kuala Langat, Klang and Kuala Selangor, Sepang and Sabak Bernam. Almost similar finding was presented by Hassan et al. (2012), who reported that dengue transmission risk in Kuala Lumpur and the neighbouring districts like Hulu Langat, Petaling and Gombak, was high. This situation is most likely contributed by the rapid development and urbanization that took place in Klang valley (Kuala Lumpur and the surrounding areas) that leads to formation of temporary breeding sites for *Aedes* mosquito.

By examining the transovarial transmission of DENV at study areas, it also allows the Minimum Infection Rate (MIR) value for both species to be measured. MIR is an indicator of arbovirus prevalence in a mosquito population where it is believed that the risk of arbovirus transmission to humans and animals is increases as the infection rate increases (Bustamante & Lord 2010). This study shows an opposite MIR values between *Aedes* spp collected from Kuala Lumpur and Selangor. For Kuala Lumpur, MIR value for *Ae. albopictus* pools was higher than *Ae. aegypti* while in Selangor MIR value for *Ae. albopictus* was lower than *Ae. aegypti* suggesting that both species can be a competent vector where *Ae. albopictus* is more competent compared to *Ae. aegypti* in Kuala Lumpur but *Ae. aegypti* in the competent one in Selangor. There were several studies however that reported *Ae. albopictus* is the less competent species in transmitting DENV as compare to *Ae. aegypti* (Alto et al. 2008; Kamgang et al. 2019; Rezza 2012).

CONCLUSION

In conclusion, this study has successfully established that not all hotspot areas presented high density of breeding sites and larval population, and not all hotspot areas are area with high dengue transmission risk. Furthermore, localities with high dengue transmission risk were located mainly in Kuala Lumpur and its neighbouring districts. Transovarial transmission were not significantly correlated with density of larval population, and finally both species shown to be a competent vector but at different location.

ACKNOWLEDGEMENT

The authors are grateful to the Director-General of Health, Malaysia for permission to publish this paper. We especially thanked the staff of Medical Entomology Unit of IMR without whose diligence and hard work under difficult field conditions this research would not have been accomplished. This study was conducted under grant NIH-866334-8009(b).

REFERENCES

- Alto, B.W., Reiskind, M.H. & Lounibos, L.P. 2008. Size alters susceptibility of vectors to dengue virus infection and dissemination. *American Journal of Tropical Medicine and Hygiene* 79(5): 688–695.
- Anderson, J.R. & Rico-Hesse., R. 2006. *Aedes aegypti* vectorial capacity is determined by the infecting genotype of dengue virus. *American Journal of Tropical Medicine and Hygiene* 75(5): 886-892.
- Bhoomiboonchoo, P., Nisalak, A., Chansatiporn, N., In-Kyu, Y., Kalayanarooj, S., Thipayamongkolgu, M., Endy, T., Rothman, A.L., Green, S., Srikiatkhachorn, A., Buddhari, D., Mammen, P.M. & Gibbons, R.V., 2015. Sequential dengue virus infections detected in active and passive surveillance programs in Thailand, 1994– 2010. BMC Public Health 15: 250.
- Bustamante, D.M. & Lord, C.C. 2010. Sources of error in the estimation of mosquito infection rates used to assess risk of arbovirus transmission. *American Journal of Tropical Medicine and Hygiene* 82(6): 1172–1184.
- Chatterjee, S., Chakraborty, A. & Sinha, S.K., 2015. Spatial distribution & physicochemical characterization of the breeding habitats of *Aedes aegypti* in & around Kolkata, West Bengal, India. *The Indian Journal of Medical Research* 142 (1): S79–S86.
- Chen, L.H. & Wilson, M.E., 2004. Transmission of dengue virus without mosquito vector: nosocomial mucocutaneous transmission and other routes of transmission. *Clinical Infectious Diseases* 39 (6): e56-e60.
- Chen, C.D., Seleena, B., Mohd Masri, S., Chiang, Y.F., Lee, H.L., Nazni, W.A. & Sofian-Azirun, M., 2005. Dengue vector surveillance in urban residential and settlement areas in Selangor, Malaysia. *Tropical Biomedicine* 22(1): 39–43.
- Chen, C.D., Seleena, B., Nazni, W.A., Lee, H.L., Masri, S.M., Chiang, Y.F. & Sofian-Azirun, M., 2006. Dengue vector surveillance in endemic area in Kuala Lumpur city centre and Selangor state, Malaysia. *Dengue Bulletin* 30: 197-203.
- Chen, C., Lee, H.L., Stella-Wong, S., Lau, K. & Sofian-Azirun, M., 2009. Container survey of mosquito breeding sites in a university campus in Kuala Lumpur, Malaysia. *Dengue Bulletin* 33: 187-193.
- Edelman, R. 2011. Unique challenges faced by the clinical evaluation of dengue vaccines. *Expert Review of Vaccines* 10(2): 133-136.
- Faiz, M., Nazri, C.D., Nurmahirah, Z., Siti Nur Anis, H., Chua, S.T. & Siti Nazrina, C. 2017. Profiling of dengue vectors breeding habitat at urban resisdental areas in Shah Alam, Malaysia. *Serangga* 22(2): 185-216.
- Hassan, H., Shamarina, S. & Nor, R.H. 2012. Risk mapping of dengue in Selangor and Kuala Lumpur, Malaysia. *Geospatial Health* 7(1): 21-25.

- Higa, Y. 2011. Dengue vectors and their spatial distribution. *Tropical Medicine and Health* 39(4): 17-27.
- Ho, L.Y., Loh, T.S. & Yam, L.A. 2014. Surveillance and resistance status of *Aedes* population in two suburban residential areas in Kampar town, Perak, Malaysia. *Tropical Biomedicine* 31(3): 441–448.
- Islam, S., Haqueb, C.E., Hossainc, S. & Rochond, K. 2019. Role of container type, behavioural, and ecological factors in *Aedes* pupal production in Dhaka, Bangladesh: An application of zero-inflated negative binomial model. *Acta Tropica* 193: 50–59.
- iDengue, 2018. Malaysian Space Agency (MYSA) and Disease Control Division (BKP), Ministry of Health Malaysia (MOHM), 2018. https://idengue.mysa.gov.my/ide_v3/index.php. [8 April 2021]
- IQI, 2017. 3 common myths of urban & suburban areas. https://www.iqiglobal.com/blog/3-common-myths-of-urban-suburban-areas/ [April 2021]
- Jahan, N.K., Ahmad, M.P., Dhanoa, A., Meng, C.Y., Ming, L.W., Reidpath, D.D., Allotey, P., Zaini, A., Phipps, M.E., Fatt, Q.K., Rabu, A.B., Sirajueen, R., Ahmad Fatan A.A.B., Ghafar, F.a., Ahmad H.B., Othman, I. & SyedHassan, S. 2016. A community-based prospective cohort study of dengue viral infection in Malaysia. *Infectious Diseases of Poverty* 5: 76.
- Jeffery, J., Rohela, M., Muslimin, M., Abdul Aziz, S.M.N., Jamaiah, I., Kumar, S., Tan, T.C., Lim, Y.A.L., Nissapatorn, V. & Abdul-Aziz, N.M., 2012. *Illustrated Keys: Some Mosquitoes of Peninsular Malaysia*. Malaysia: University of Malaya Press.
- Kamgang, B., Happi, J.Y., Boisier, P., Njiokou, F., Herve, J.P., Simard, F. & Paupy, C. 2010. Geographic and ecological distribution of the dengue and chikungunya virus vectors *Aedes aegypti* and *Aedes albopictus* in three major Cameroonian towns. *Medical and Veterinary Entomology* 24(2): 132-41.
- Kamgang, B., Vazeille, M., Tedjou, A.N., Wilson-Bahun, T.A., Yougang, A.P., Mousson, L., Wondji, C.S. & Anna-Bella, F., 2019. Risk of dengue in Central Africa: Vector competence studies with *Aedes aegypti* and *Aedes albopictus* (Diptera: Culicidae) populations and dengue 2 virus. *PLoS Neglected Tropical Diseases* 13(12): e0007985.
- Lanciotti, R.S., Calisher, C.H., Gubler, D.J., Chang, G.J. & Vordam, A.V., 1992. Rapid detection and typing of dengue viruses from clinical samples by using reverse transcriptase-polymerase chain reaction. *Journal of Clinical Microbiology* 30: 545-551.
- Lee, H.L. 1992. *Aedes* ovitrap and larval survey in several suburban community in Selangor, Malaysia. *Mosquito Borne Diseases Bulletin* 9(1): 9-15.
- Lee, H.L. & Rohani, A., 2005. Transovarian transmission of dengue in *Aedes* vectors in relation to dengue outbreak in an urban area in Malaysia. *Dengue Bull*. 29: 106–11.
- Lenhart, A.E., Mario, W., Hector, C. & Axel, K., 2005. Building a better ovitrap for detecting *Aedes aegypti* oviposition. *Acta Tropica* 96: 56-59.

ISSN 1394-5130

- Louis, V.R., Revati, P., Olaf, H., Pitcha, R., Annelies, W.S., Yesim, T. & Dambach, P, 2014. Modelling tools for dengue risk mapping – a systematic review. *International Journal* of Health Geographics 13: 50
- Meena, A.R. & Choudhary, N.L., 2019. Container breeding preference of *Aedes albopictus* in urban environment. *International Journal of Mosquito Research* 6(5): 44-47.
- Messina, J.P., Oliver, J.B., Nick, G., Moritz, U.G.K., Wint, G.R.W., Ray, S.E., Pigott, D.M., Shearer, F.M., Johnson, K., Earl, L., Marczak, L.B., Shirude, S., Weaver, N.D., Gilbert, M., Velayudhan, R., Jones, P., Jaenish, T., Scott, T.W., Reiner Jr, R.C. & Hay, S.I. 2019. The current and future global distribution and population at risk of dengue. *Nature Microbiology* 4: 1508-1515.
- Mia, M.S., Rawshan, A.B., Er, A.C., Raja Datuk Zaharaton, R.Z.A. & Joy, J.P. 2013. Trends of dengue infections in Malaysia, 2000-2010. *Asian Pacific Journal of Tropical Medicine* 462-466.
- Ministry of Health Malaysia (MOHM), 2015. Garis Panduan Pencegahan dan Pengawalan Nyamuk Aedes di Tapak-tapak Pembinaan. Vector Borne Disease Sector, Disease Control Division, Ministry of Health Malaysia: Putrajaya, Malaysia. https://idengue.mysa.gov.my/pdf/SOP_constructionsite.pdf [23 March 2018].
- Ministry of Health Malaysia (MOHM), 2020. Kenyataan akhbar khas Ketua Pengarah Kesihatan Malaysia berkenaan situasi semasa demam denggi, zika dan chikungunya di Malaysia tahun 2019 dan inisiatif KKM untuk memerangi denggi tahun 2020. Putrajaya: Minstry of Health. https://www.moh.gov.my/index.php/database____stores/store_view_page/21/1290 [2 February 2021].
- Mohd Amierul, F.M., Mohd Hatta, M., Noor Aliza, L. & Hanipah, S. 2018. Study on key *Aedes* spp breeding containers in dengue outbreak localities in Cheras district, Kuala Lumpur. *International Journal of Mosquito Research* 5(2): 23-30.
- Mohd Syariefudin, A., Mohd Mahadee, I. & Mansor, M.N. 2016. Kesepaduan sosial dan kejiranan di kawasan rukun tetangga. *Jurnal Kinabalu* 19: 53-75.
- Nazri, C.D, Abu Hassan, A., Ishak, A.R. & Rodziah, I. 2013a. Assessing the risk of dengue fever based on the epidemiological, environmental and entomological variables. *Procedia-Social and Behavioral Sciences* 105: 183-194.
- Nazri, C.D., Abu Hassan, A. & Rodziah, I., 2013b. Habitat Characterization of *Aedes sp.* breeding in urban hotspot area. *Procedia Social and Behavioral Sciences* 85: 100–109.
- Nor Aliza, A.R., Harvie, S., Nur Ain Marduah, R., Lela, S., Siti Fairouz, I., Razitasham. S. & Marlini, O., 2019. Detection of transovarial dengue virus in *Aedes albopictus* from selected localities in Kuching and Samarahan divisions, Sarawak, Malaysia by reverse transcriptase polymerase chain reaction (RT-PCR). *Serangga* 24(2): 145-158.

- Paul, K.K., Dhar-Chowdhury, P., Haque, C.E., Al-Amin, H.M., Goswami, D.R., Kafi, M.A.H., Drebot, M.A., Lindsay, L.R., Ahsan, G.U. & Brooks, W.A. 2018. Risk factors for the presence of dengue vector mosquitoes, and determinants of their prevalence and larval site selection in Dhaka, Bangladesh. *PLoS ONE* 13(6): e0199457.
- Philbert, A. & Ijumba, J.N. 2013. Preferred breeding habitats of *Aedes aegypti* (Diptera: Culicidae) mosquito and its public health implications in Dares Salaam, Tanzania. *Journal of Environmental Research and Management* 4(10): 344-351.
- Peña-García, V.H., Triana-Chávez, O., Mejía-Jaramillo, A.M., Díaz, F.J., Gómez-Palacio, A. & Arboleda-Sánchez, S., 2016. Infection rates by dengue virus in mosquitoes and the influence of temperature may be related to different endemicity patterns in three Colombian cities. . *International Journal of Environmental Research and Public Health* 13(7): 734.
- Pratt, J.J., Heterick, R.H., Harrison, J.B. & Haber, L. 1946. Tires as a factor in the transportation of mosquitoes by ships. *The Military Surgeon* 99(6): 785–788.
- Rahayu, A., Saraswati, U., Supriyati, E., Kumalawati, D.A., Hermantara, R., Rovik, A., Daniwijaya, E.W., Fitriana, I., Setyawan, S., Ahmad, R.A., Wardana, D.S., Indriani, C., Utarini, A., Tantowijoyo, W. & Arguni, E. 2019. Prevalence and distribution of dengue virus in *Aedes aegypti* in Yogyakarta city before deployment of *Wolbachia* infected *Aedes aegypti*. *International Journal of Environmental Research and Public Health* 16:1742.
- Rahim, J., Ahmad, A.H., Maimusa, A.H. & Irfan, S. 2018. Updated abundance and distribution of *Aedes albopictus* (Skuse) (Diptera: Culicidae) in Penang Island, Malaysia. *Tropical Biomedicine* 35(2): 308–320.
- Reiter, P. 1998. *Aedes albopictus* and the world trade in used tires, 1988-1995: The shape of things to come? *Journal of the American Mosquito Control Association* 14(1): 83-94.
- Rezza, G. 2012. Aedes albopictus and the re-emergence of dengue. Biomed Central Public Health 12: 1-3.
- Rohani, A., Asmaliza, I., Zainah, S. & Lee, H.L., 1997. Detection of dengue virus from field *Aedes aegypti* and *Aedes albopictus* adults and larvae. *Southeast Asian Journal of Tropical Medicine and Public Health* 28(1): 138-142.
- Rohani, A., Abdullah, A.G., Ong, Y.F., Saadiyah, I., Zamree, I. & Lee, H.L. 2001. Survey of mosquito larvae distribution in container habitats collected from urban and rural areas in major town of Malaysia. *Tropical Biomedicine* 18(1): 41-49.
- Rohani, A., Suzilah, I., Malinda, M., Anuar, I., Mohd Mazlan, I., Salmah Maszaitun, M., Topek, O., Tanrang, Y., Ooi, S.C., Rozilawati, H. & Lee, H.L. 2011. Aedes larval population dynamics and risk for dengue epidemics in Malaysia. Tropical Biomedicine 28(2): 237-248.
- Rohani, A., Zamree, I., Lee, H.L., Mustafakamal, I., Norjaiza, M.J. & Kamilan, D. 2007. Detection of transovarial dengue virus from field –caught *Aedes aegypti* and *Ae*.

ISSN 1394-5130

albopictus larvae using c6/36 cell culture and reverse transcriptase-polymerase chain reaction (RT-PCR) techniques. *Dengue Bulletin* 31:47-57.

- Rohani, A., Aidil Azahary, A.R., Malinda, M., Zurainee, M.N., Rozilawati, H., Wan Najdah, W.M.A. & Lee, H.L. 2014. Eco-virological survey of *Aedes* mosquito in selected outbreak areas in Malaysia. *Journal of Vector & Borne Diseases* 327-332.
- Rohani, A., Suzilah, I., Wan Najdah, W.M.A., Topek, O., Ibrahim, M. & Lee, H.L. 2018. Factor determining dengue outbreak in Malaysia. *Plos One* 13(2): e0193326.
- Rozilawati, H., Tanaselvi, K., Nazni, W.A., Mohd Masri, S., Zairi, J., Adanan, C.R. & Lee, H.L. 2015. Surveillance of *Aedes albopictus* Skuse breeding preference in selected dengue outbreak localities, peninsular Malaysia. *Tropical Biomedicine* 32(1): 49–64.
- Saleeza, S.N.R., Norma-Rashid, Y. & Sofian-Azirun, M. 2011. Mosquitoes larval breeding habitat in urban and suburban areas, peninsular Malaysia. *World Academic of Science, Engineering and Technology* 58: 569-573.
- Saleeza, S.N.R., Norma-Rashid, Y. & Sofian-Azirun, M. 2013. Mosquito species and outdoor breeding places in residential areas in Malaysia. Southeast Asian Journal of Tropical Medicine and Public Health 44(6): 963-969.
- Samruhaizad, S., Jamaluddin, M.J. & Azahan, A. 2014. Isu perbandaran dan kualiti hidup penduduk pinggir bandar. *International Journal of the Malay World and Civilization* 2 (1): 63-75.
- Yee, D.A. 2008. Tires as habitats for mosquitoes: A review of studies within the Eastern United States. *Journal of Medical Entomology* 45(4): 581-593.
- Vijayakumar, K., Sudheesh Kumar, T.K., Zinia, T.N., Farook, U. & Anu, K., 2014. A study on container breeding mosquitoes with special reference to *Aedes* (Stegomyia) *aegypti* and *Aedes albopictus* in Thiruvananthapuram district, India. *Journal of Vector Borne Diseases* 51: 27–32.
- WHO (World Health Organization). 1975. Manual on Practical Entomology in Malaria. Part II. Methods and Techniques. Geneva: World Health Organization. https://apps.who.int/iris/bitstream/handle/10665/42481/WHO_OFFSET_13_(part2).p df;jsessionid=2EE8931F204F7A8C3C4F5EBB898BF70E?sequence=2 [18 July 2019].

WHO (World Health Organization). 2010. Dengue: the fastest growing mosquito-borne disease in the world. http://www.who.int/neglected_diseases/integrated_media /integrated_media _2010_Dengue_vs_malaria/en/ [18 July 2019].

							Density level				
Location number	Name of location	State	District	Latitude	Longitude	Category	Breeding sites	<i>Ae. aegypti</i> population	Ae. albopictus population	Dengue transmission risk	
1	Angkasa Condominium	KL	KL	3.0792547	101.72989	Ur	4	NA	3	2	
2	Apt. Bunga Raya Pandan Indah	KL	KL	3.130967	101.75746	Ur	5	5	5	5	
3	Apt. Permai	KL	KL	3.064228	101.70627	Ur	1	2	1	2	
4	Apt. Sri Penara	KL	KL	3.096531	101.71009	Ur	4	NA	4	3	
5	Blok L Pandan Jaya	KL	KL	3.133395	101.73973	Ur	5	5	5	5	
6	Blok M Pandan Jaya	KL	KL	3.1379276	101.74038	Ur	5	5	5	5	
7	Desa Melawati	KL	KL	3.21706	101.73761	Ur	5	3	NA	5	
8	Flat Danau Kota	KL	KL	3.2001023	101.71229	Ur	5	5	4	5	
9	Flat Keramat AU3/1	KL	KL	3.181583	101.75862	Ur	5	5	3	5	
10	Flat KTMB	KL	KL	3.1307708	101.67752	Ur	5	5	5	5	
11	Flat Pandan Mewah	KL	KL	3.1270924	101.76492	Ur	5	5	5	5	
12	Flat Tmn. Muda	KL	KL	3.119335	101.75388	Ur	5	5	NA	5	
13	Flat Sect. 1 Wangsa Maju	KL	KL	3.204492	101.73084	Ur	5	5	5	5	
14	Flat Seri Negeri Sembilan	KL	KL	3.182955	101.69559	Ur	5	5	5	5	
15	Flat Sri Kelantan	KL	KL	3.1920277	101.69123	Ur	5	5	NA	5	
16	Flat Sri Labuan Bndr Tun Razak	KL	KL	3.090105	101.72072	Ur	5	2	4	4	
17	Flat Sri Perlis 2	KL	KL	3.167957	101.72218	Ur	5	5	3	4	
18	Kg. Chubadak	KL	KL	3.202868	101.69696	Ur	5	5	NA	5	
19	Kg. Dato' Keramat	KL	KL	3.16442	101.72549	Ur	5	NA	3	4	

ISSN 1394-5130

20	Kg. Padang Balang	KL	KL	3.209852	101.69898	Ur	5	5	4	5
21	Kg. Pantai Dalam	KL	KL	3.098856	101.6685	Ur	5	5	4	5
22	PPR Bt Muda	KL	KL	3.208516	101.6817	Ur	5	5	NA	5
23	PPR Kg. Limau	KL	KL	3.1096496	101.67529	Ur	5	5	5	5
24	PPR Sri Pantai	KL	KL	3.10661	101.67414	Ur	5	NA	4	5
25	Sect. 10 Wangsa Maju	KL	KL	3.1856725	101.73913	Ur	5	5	5	5
26	Tmn. Bahagia Kuchai	KL	KL	3.0802513	101.68438	Ur	1	NA	1	1
27	Tmn. Bukit Angkasa	KL	KL	3.10968	101.6709	Ur	5	5	5	5
28	Tmn. Dato' Senu	KL	KL	3.1918894	101.69834	Ur	5	5	5	5
29	Tmn. Daya	KL	KL	3.223722	101.63631	Ur	4	NA	3	2
30	Tmn. Delima Cheras	KL	KL	3.056337	101.74706	Ur	5	1	2	2
31	Tmn. Jaya	KL	KL	3.090602	101.7235	Ur	5	2	4	4
32	Tmn. Keramat	KL	KL	3.16822	101.74181	Ur	5	5	4	5
33	Tmn. Kepong	KL	KL	3.211735	101.63147	Ur	3	NA	5	5
34	Tmn. Melati	KL	KL	3.2230721	101.72126	Ur	5	5	5	5
35	Tmn. Melawati	KL	KL	3.21126	101.75444	Ur	4	3	3	3
36	Tmn. Permata G	KL	KL	3.205594	101.75137	Ur	5	NA	5	5
37	Tmn. Sri Rampai	KL	KL	3.1967026	101.73571	Ur	5	3	5	5
38	Tmn. Wangsa Permai	KL	KL	3.207762	101.61254	Ur	5	2	5	5
39	Kg. Bagan Sg. Burong	Sel	SB	3.693138	100.9374	Ru	2	1	2	2
40	Kg. Site A Sekinchan	Sel	SB	3.526706	101.14226	Ru	1	NA	2	1
41	Tmn. Aman Jaya	Sel	SB	3.516439	101.1004	Ru	4	2	4	4
42	Tmn. Ria Sekinchan	Sel	SB	3.514923	101.0938	Ru	4	2	4	4
43	Tmn. Aman SB	Sel	SB	3.773819	100.99018	Ru	2	2	5	5
44	Tmn. Berkat Sg. Besar	Sel	SB	3.6686009	100.98805	Ru	4	2	4	4
45	Tmn. Bernam Jaya	Sel	SB	3.657203	101.5345	Ru	2	NA	2	1
46	Tmn. Padu Permai	Sel	SB	3.663586	100.99127	Ru	4	NA	4	4
47	Apt. Teratai Bkt. Beruntung	Sel	HS	3.4440838	101.55378	Ru	2	4	2	2
48	Felda Gedangsa	Sel	HS	3.730298	101.38596	Ru	3	1	2	2

49	Kg. Asam Kubang	Sel	HS	3.5627287	101.66524	Ru	2	NA	3	2
50	Tmn. Rajawali	Sel	HS	3.6443086	101.55716	Ru	1	1	2	1
51	Tmn. Tempua Bistari	Sel	HS	3.6214216	101.57323	Ru	3	1	4	3
52	Jln Cakera Purnama Puncak Alam	Sel	KS	3.2416006	101.42836	S/Ur	4	1	5	5
53	Tmn. Cempaka Sari Ijok	Sel	KS	3.317228	101.41069	Ru	3	1	4	3
54	Apt. Ukay Bistari	Sel	G	3.196008	101.76565	Ur	5	5	NA	5
55	Flat Tmn. Sri Batu Bt Caves	Sel	G	3.232261	101.69087	S/Ur	5	5	5	5
56	Kepong Baru	Sel	G	3.269022	101.6516	Ur	3	2	2	2
57	Kg. Changkat Greenwood	Sel	G	3.232268	101.70937	S/Ur	5	5	5	5
58	Kg. Laksamana G	Sel	G	3.2500488	101.68132	S/Ur	5	5	5	5
59	Pinggiran Bt. Caves	Sel	G	3.254441	101.69484	S/Ur	5	5	5	5
60	Selayang Baru	Sel	G	3.250246	101.6716	S/Ur	5	5	5	5
61	Selayang Indah	Sel	G	3.248906	101.66236	S/Ur	5	5	5	5
62	Tmn. Sri G F9	Sel	G	3.248931	101.70578	Ur	5	NA	5	5
63	Tmn. G	Sel	G	3.228704	101.70228	Ur	5	5	NA	5
64	Alam Budiman Setia Alam	Sel	Р	3.143955	101.47884	S/Ur	2	1	2	2
65	Apt. Enggang Kinrara	Sel	Р	3.0353618	101.6736	S/Ur	1	1	1	1
66	Apt. Perdana S. Alam	Sel	Р	3.086411	101.54976	Ur	1	NA	1	1
67	Apt. Sri Ixora	Sel	Р	3.02554	101.57097	Ur	3	4	NA	2
68	Apt. Sri Meranti D'mansara	Sel	Р	3.1201594	101.57142	Ur	3	4	2	3
69	Apt. Vista Lavender	Sel	Р	3.0609346	101.64109	Ur	5	5	5	5
70	Dataran Otomobil	Sel	Р	3.064471	101.52743	Ur	4	NA	2	3
71	Desa Mentari	Sel	Р	3.079073	101.6162	Ur	5	NA	5	5
72	Flat Nilam Sari S7 S.Alam	Sel	Р	3.0740792	101.48721	Ur	3	3	5	5
73	Flat Sect. 27 S. Alam	Sel	Р	3.024581	101.56868	Ur	3	4	1	2

ISSN 1394-5130

74	Flat Tmn. Dato Harun	Sel	Р	3.080868	101.63432	Ur	5	5	5	5
75	Mentari Court	Sel	Р	3.082515	101.60999	Ur	3	2	NA	2
76	Sect. 7 S. Alam	Sel	Р	3.0640912	101.48975	Ur	3	3	5	5
77	Sect. 8 S. Alam	Sel	Р	3.09149	101.50997	Ur	2	NA	2	2
78	Sect. 20 S. Alam	Sel	Р	3.058137	101.53972	Ur	4	4	NA	3
79	SS2 PJ	Sel	Р	3.1126565	101.62375	Ur	2	NA	1	1
80	SS3 PJ	Sel	Р	3.0955793	101.61196	Ur	3	NA	2	2
81	SS22 Damansara	Sel	Р	3.12463	101.61962	Ur	2	1	2	1
82	Tmn. Medan Baru	Sel	Р	3.074182	101.63692	Ur	5	5	5	5
83	Tmn. Medan Cahaya	Sel	Р	3.0821003	101.63597	Ur	5	NA	5	5
84	Tmn. Subang baru	Sel	Р	3.153433	101.52794	Ur	4	1	2	2
85	Tmn. Sg. Besi Indah	Sel	Р	3.0303115	101.72186	Ur	2	1	2	2
86	Apt. Damai Mewah Kajang	Sel	HT	2.984724	101.80909	S/Ur	5	NA	3	4
87	Apt. PKNS Tmn. Dagang	Sel	HT	3.1467369	101.75896	Ur	5	5	5	5
88	Apt. Putra Permai	Sel	HT	2.9952795	101.66781	S/Ur	2	2	2	2
89	Apt. Saujana Ampang	Sel	HT	3.1341009	101.78981	S/Ur	4	2	NA	4
90	Apt. Seri Baiduri Ampang	Sel	HT	3.157395	101.77695	S/Ur	5	NA	5	5
91	Apt. Tmn. Perkasa	Sel	HT	3.1046657	101.79899	S/Ur	4	4	2	4
92	Condo. Ivory	Sel	HT	3.011841	101.81156	S/Ur	3	2	NA	2
93	Flat Kemboja Tmn. Ampang Indah	Sel	HT	3.148234	101.77921	Ur	5	5	5	5
94	Flat Seri Nilam Ampang	Sel	HT	3.137372	101.76811	Ur	5	5	5	5
95	Flat Tmn. Dagang Permai	Sel	HT	3.13975	101.78088	Ur	5	5	5	5
96	Kg. Sg. Raya Bt 9	Sel	HT	3.068074	101.76848	S/Ur	3	2	4	3
97	PPR Hiliran Ampang	Sel	HT	3.150067	101.74623	Ur	5	5	5	5
98	Tmn. Bkt Mewah Kajang	Sel	HT	2.98165	101.80322	Ur	5	5	3	4

99	Tmn. Dato' Ahmad Razali	Sel	HT	3.153209	101.75902	Ur	5	5	5	5
100	Tmn. Maju Jaya	Sel	HT	3.02268	101.74505	Ur	3	2	5	4
101	Tmn. Nirwana Ampang	Sel	HT	3.1414363	101.75229	Ur	5	5	5	5
102	Tmn. Pelangi Semenyih	Sel	HT	2.9334997	101.86274	S/Ur	1	NA	1	1
103	Tmn Sri Nanding	Sel	HT	3.105123	101.80845	S/Ur	4	4	2	4
104	Tmn. Taming Jaya	Sel	HT	3.0257636	101.74369	S/Ur	3	2	5	4
105	Tmn. Taming Impian	Sel	HT	3.0153012	101.80175	S/Ur	3	2	2	2
106	Sect. 7 Bdr Baru Bangi	Sel	HT	2.968499	101.77849	S/Ur	1	2	1	1
107	Sect. 9 Bdr. Baru Bangi	Sel	HT	2.960517	101.74856	S/Ur	2	1	2	1
108	Sect. 1 Bdr. Teknologi Kajang	Sel	HT	2.9684449	101.82569	S/Ur	4	4	4	4
109	Apt. Samudera Pulau Indah	Sel	K	2.931429	101.31605	Ru	3	NA	5	5
110	Kg. Pinang	Sel	Κ	2.9603505	101.33014	Ru	2	NA	2	2
111	Pulau Ketam	Sel	Κ	3.019856	101.25315	Ru	5	5	1	4
112	Apt. Langat Utama	Sel	KT	2.7976084	101.66266	S/Ur	2	NA	2	2
113	Tmn. Aman Banting	Sel	KT	2.8078908	101.50137	S/Ur	5	5	5	5
114	Tmn. Aman Sg. Pelek	Sel	KT	2.644232	101.7096	S/Ur	2	1	2	2
115	Tmn. Banting Baru	Sel	KT	2.811065	101.45549	S/Ur	4	2	2	2
116	Tmn. Gembira Banting	Sel	KT	2.8095877	101.50024	S/Ur	5	NA	5	5
117	Tmn. Perwira TPG	Sel	KT	2.940149	101.47577	S/Ur	5	5	5	5
118	Tmn. Salak Indah	Sel	KT	2.787509	101.7489	Ru	2	NA	2	2
119	Tmn. Seri Jarom	Sel	KT	2.874016	101.4929	Ru	3	2	2	2
120	Tmn. Seri Medan Jaya	Sel	KT	2.9333627	101.47417	Ru	5	5	5	5
121	Tmn. Sg. Emas Banting	Sel	KT	2.821003	101.5328	Ru	3	NA	3	2
122	Tmn. Sri Telok Datok	Sel	KT	2.8168332	101.51943	Ru	5	1	NA	2
123	Tmn. Sri Putra	Sel	KT	2.81011	101.48685	Ru	5	5	5	5
124	Tmn. Tanjong Sepat	Sel	KT	2.6591975	101.56308	Ru	3	1	3	2
125	Tmn. Yayasan	Sel	KT	2.871896	101.49884	Ru	3	2	2	2

126	Desa Vista	Sel	S	2.840105	101.75108	S/Ur	3	1	2	2
127	Kg. Baru Lanjut S	Sel	S	2.7956047	101.72062	Ru	5	2	5	5
128	KLIA DownTown	Sel	S	2.769819	101.69718	Ru	5	4	4	4
129	Tmn. Dataran Abadi	Sel	S	2.7871758	101.70795	Ru	5	2	5	5
130	Tmn. Ria 2 Bagan Lalang	Sel	S	2.638971	101.72144	Ru	2	NA	2	2
131	Tmn. Seroja BBST	Sel	S	2.825295	101.7277	Ru	1	1	NA	1
132	Tmn. Sri Bayu Indah Bagan Lalang	Sel	S	2.600953	101.69272	Ru	2	NA	2	1

State: Kuala Lumpur (KL); Selangor (Sel)

District: Kuala Lumpur (KL); Sabak Bernam (SB); Hulu Selangor (HS); Kuala Selangor (KS); Gombak (G); Petaling (P); Hulu Langat (HT); Kuala Langat (KT); Klang (K) Sepang (S)

Category: Urban (Ur); Rural (Ru); SubUrban (S/Ur)

Density level: High (5); Medium high (4); Medium (3); Medium low (2); Low (1); Not available (NA)