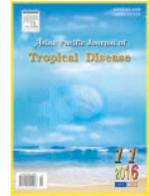




Contents lists available at ScienceDirect

## Asian Pacific Journal of Tropical Disease

journal homepage: www.elsevier.com/locate/apjtd



Dengue research

doi: 10.1016/S2222-1808(16)61143-X

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## Current observation on *Aedes* mosquitoes: A survey on implication of dengue infection, human lifestyle and preventive measure among Malaysia resident in urban and sub-urban areas

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## ARTICLE INFO

## Article history:

Received 25 Aug 2016

Received in revised form 7 Sep, 2nd

revised form 13 Sep 2016

Accepted 24 Sep 2016

Available online 8 Oct 2016

## Keywords:

*Aedes*

Dengue

Lifestyle

Mosquito

Survey

## ABSTRACT

**Objective:** To evaluate the current knowledge level and information on *Aedes* mosquitoes and dengue among Penang resident in urban (Sungai Dua) and sub-urban areas (Batu Maung).

**Methods:** The questionnaire comprises of twenty-three questions, surveyed on 202 respondents regarding socio-demographic, the observation of general *Aedes* mosquito behaviour, dengue infection threat, preventive measures against mosquitoes and lifestyle (on usage of artificial light) in changing the behaviour of *Aedes* mosquitoes.

**Results:** The respondents in sub-urban Batu Maung showed less knowledge level regarding *Aedes* mosquitoes as compared to respondents in urban Sungai Dua. There was a positive association between biting frequency of mosquitoes with the lifestyle of respondents (in relation to the light use), the use of personal protection and fogging operations conducted by the Ministry of Health. There is a positive relationship between the knowledge of preferred mosquitoes resting place and the respondent aged between 17 to 24 (78.2%) in sub-urban Batu Maung. Urban Sungai Dua respondents showed a significant relation between the use of lights and the resting places for *Aedes* mosquitoes. The preventive measures chosen by both sub-distinct areas against mosquitobites were significantly affected by the biting time and the mosquitoes biting frequency ( $P < 0.05$ ).

**Conclusions:** In this study, we concluded that there is a direct link between the knowledge of the *Aedes* biting frequency with dengue infection, human lifestyle on the light use and preventive measures against mosquitoes. However, the level of knowledge is poor on the subject of *Aedes* mosquitoes and dengue for both urban and sub-urban respondents.

## 1. Introduction

Malaysia, a tropical with warm climate provide a conducive condition for breeding site of dengue vector mosquitoes, *Aedes albopictus* (*Ae. albopictus*) and *Aedes aegypti* (*Ae. aegypti*). Malaysia was ranked the third among countries in the Western Pacific Region on the number of reported dengue cases from 1991 until 2007[1]. Dengue was first reported in Malaysia during the early 1900s and became a serious public health problem since the 1970s[2]. Starting from the year of 1982, Malaysia faced significant outbreaks of dengue fever with a gradually increasing number of cases and fatalities each

year. The disease had become endemic since the early 1990s, with yearly and frequent outbreaks there after[1] till 2008.

In year 2013, reported dengue cases in Malaysia rose up to 43348 cases, which was closer to the highest peak reported in 2008 with 49335 cases[3]. The number of dengue cases continued to rise, where on the 12th September 2015, Malaysia experienced a staggering 85488 of dengue cases with 234 deaths[4]. Over the past decades, major dengue disease outbreaks occurred in a cyclical pattern involving dengue virus type 1 (DENV-1), DENV-2, DENV-3 and DENV-4 serotypes[5]. Recent data showed that during the dengue outbreak in 2013 (Malaysia), DENV-2 serotypes overtook a pre-existing DENV-3 and DENV-4 as the dominance virus pattern since 2012[3].

To control the dengue problem, various vector control efforts have been used throughout the decades. The control efforts mainly target environmental management and purging larval breeding sites. However, early control methods relied heavily on chemical insecticide

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Foundation Project: Supported by the Fundamental Research Grant Scheme by the Ministry of Education and Universiti Sains Malaysia (203/PBIOLOGI/6711359).

The journal implements double-blind peer review practiced by specially invited international editorial board members.

compared to other safer methods[6]. Even though chemical control is successful in reducing the vector diseases to occur, but this method facing challenges on the emergence and development of insecticide resistance in dengue vector especially *Aedes* species[7].

*Aedes*, a vector of dengue was found to be widespread in human habitation which is mostly influenced by poor sanitation. Educational programs such as campaigns, talks and surveys need to disseminate to the community as a preventive measure to reduce the risks of outbreaks[8]. The knowledge on dengue vector and mosquitoes was found to be adequate, but important relationship concerning mosquitoes, human behavior and dengue transmission is still lacking[9]. In Thailand, survey participants thought they had no ability to prevent the dengue fever and the responsibility to prevent the disease is delegated to public health staff. They were ignorant on the importance of eliminating *Aedes* breeding grounds and only 15.27% attempted to destroy the *Aedes* breeding places[10].

Changes in the modern human lifestyle and urbanization might influence the behavior of *Aedes* mosquitoes, further affecting the transmission and infection of dengue viruses. In this paper, we aim to find out: the current level of knowledge among people in the urban and sub-urban areas toward *Aedes* mosquitoes and dengue, and how changes in human lifestyles might influence *Aedes* mosquitoes'. This study was conducted to understand the level of knowledge, preventive measure and effects of lifestyle among residence in two sub-distinct areas of Penang urban (Sungai Dua) and sub-urban areas (Batu Maung).

## 2. Material and methods

### 2.1. Study setting

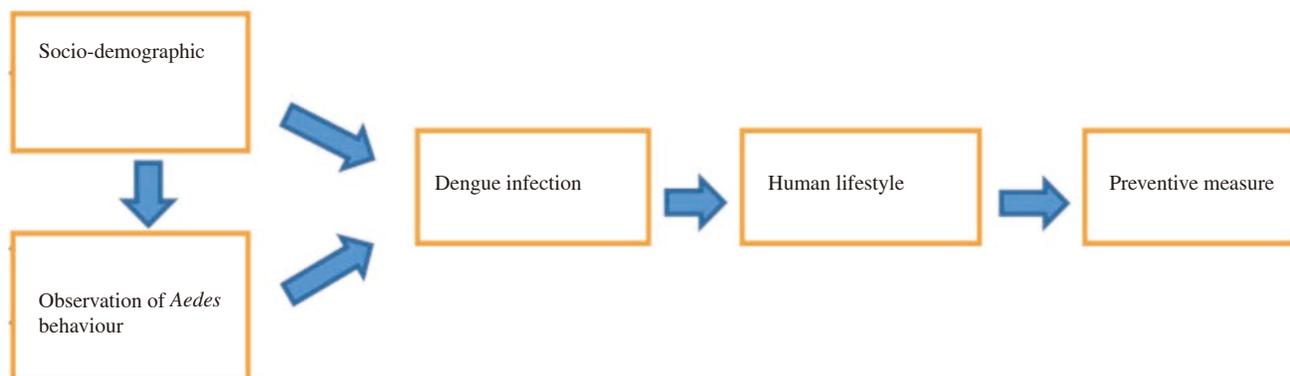
The survey was carried out in Penang Island, Malaysia, between November 2014 until Jun 2015. A total of 202 respondents with a minimum age of 17 years and above was approached personally to fill in the survey form. Two locations were chosen for this study: (1) Batu Maung area (sub-urban) which is near the sea and approximately 1 km from Penang international airport (5°17.058' N, 100°16.898' E), and (2) Sungai Dua, Penang area (urban) (5°20.920' N, 100°17.923' E) with 101 respondents from each site. Urban areas is defined as

a main city which includes the inner city with a bigger population density, whereas the sub-urban is defined as an area that is adjacent to the city. In addition, urban areas are more crowded in terms of people with establishments of transportation, and public facilities compared to the suburban territories[11]. Both areas were selected due to Batu Maung sub-urban was listed as a dengue hotspot since 2005, whereas Sungai Dua is known as a common recurring dengue hotspot in Pulau Pinang[12].

We assumed the conservative response distribution was most statistically possible at 50%. To capture a representative sample of the resident population of 6344 at Sungai Dua, and 7236 at Batu Maung with 80% confidence and a 7% margin of error, we estimated that a total of at least 83 samples was required for both Sungai Dua and Batu Maung areas, respectively (Raosoft Inc sample size calculator).

### 2.2. Study design and instruments

The survey was semi-structured. The second and third sections in the questionnaire focused on the attitude and knowledge on dengue fever sections from previous studies[8,13,14]. The questionnaire was pre-tested, and translated into Bahasa Malaysia (National language of Malaysia) with the original meaning retained. The survey was performed either in English or Malay language depending on the necessity. This paper-based questionnaire comprises of 23 questions and was divided into 5 main sections which relates to: (1) the demographic characteristics of respondent (gender, age, education and occupation) consisted of 4 questions; (2) respondent's observation on general *Aedes*' mosquitoes behavior and dengue, with 5 questions; (3) dengue infection faced by the respondent with 3 questions; (4) human lifestyle on usage of light changing behavior of *Aedes* mosquitoes with 5 question; (5) preventive measure against mosquitoes including personal protection and fogging activities in respondent's area with 6 questions. The relationship between socio-demographic, observation of *Aedes* mosquitoes by respondent, dengue infection, respondent lifestyle and preventive measure is displayed in Figure 1. Participation in this study was voluntary and no incentive was provided. Respondents names and address were not recorded to abide the confidentiality and consent of the respondents.



**Figure 1.** Diagram of the relationship between socio-demographic and observation of *Aedes* behaviour of dengue infection faced by respondent, respondent lifestyle and preventive measure.

### 2.3. Statistical analysis

The survey data were analyzed using SPSS version 20. Data were analyzed using descriptive statistics based on frequency, and percentage. Questionnaire data reliability was analyzed using reliability test of Cronbach's alpha. Coefficient associations were assessed using the *Chi-square* test as an appropriate comparison response between location areas of sub-urban (Batu Maung) and urban (Sungai Dua). A *P*-value with  $< 0.05$  was considered as significant value.

For analysis between relationship of socio-demographic (gender, age and education level) on dengue infection, lifestyle and prevention, we used multinomial logistic regression in JMP 11 software. We applied the same method on the observation of *Aedes* behaviour (frequency of biting, biting time and resting place for *Aedes*) to relate the effects on dengue infection, lifestyle and preventive measures. The best fit model using a stepwise procedure was calculated for covariates stated above and ran separately between urban and sub-urban areas. Least significant covariates were excluded until the model provides the best fit reliable equation.

The knowledge level of respondent was calculated based on Bloom's cut off points. A score within 80%–100% of correct answer defines as good knowledge, a score of 60%–79% as fair knowledge and 50% or less constitutes as poor knowledge.

## 3. Results

### 3.1. Reliability test of questionnaire

A total of 19 questions with the exception of four socio-demographic questions were analyzed for reliability test. Cronbach-alpha values of 0.312 for the questionnaire indicated low reliability. However, Kaiser-Meyer-Olkin measure of sampling adequacy indicated the questionnaire samples were mediocre (Kaiser-Meyer-Olkin = 0.501). The Bartlett's test of sphericity showed a significance value ( $\chi^2 = 583.91$ ,  $df = 171$ ,  $P = 0.00$ ), which authenticated that validity and suitability of the respondent collected for the study.

### 3.2. Socio-demographic characteristics of respondents

Table 1 shows the demographic of the 202 respondents of urban and developed sub-urban areas. Among them, 56.4% of respondents were female and 48.5% were male ( $P > 0.05$ ). The age of respondents ranged between 17 and 65 years above. A majority of 45.5% from the respondents were between 17 to 24 years of age. The level of education among respondents was considered as high where the majority of the respondents had received the secondary education (72.8%), and 21.3% had received tertiary education. Most respondents were students (44.1%) followed by private company workers (25.2%) and government staff (12.9%). The results showed that there were significant differences between two areas of urban and sub-urban ( $P < 0.05$ ) for age group, level education and occupation of respondents.

There was no significant differences between gender of respondents ( $P = 0.156$ ) which indicated an equal number between gender was selected for both sub-distinct areas in this study.

**Table 1**

Socio-demographic characteristics of respondents. *n* (%).

Factor		Sub-urban	Urban	Total	<i>P</i> -value
Gender	Male	39 (38.6)	49 (48.5)	88 (43.6)	0.156
	Female	62 (61.4)	52 (51.5)	114 (56.4)	
Age group	17–24	79 (78.2)	13 (12.9)	92 (45.5)	0.000
	25–34	10 (9.9)	30 (29.7)	40 (19.8)	
	35–44	6 (5.9)	13 (12.9)	19 (9.4)	
	45–54	3 (3.0)	15 (14.9)	18 (8.9)	
	55–64	3 (3.0)	20 (19.8)	23 (11.4)	
	> 65	0 (0.0)	10 (9.9)	10 (5.0)	
Level of education	Primary	1 (1.0)	11 (10.9)	12 (5.9)	0.000
	Secondary	88 (87.1)	59 (58.4)	147 (72.8)	
	Tertiary	12 (11.9)	31 (30.7)	43 (21.3)	
Occupation	Housewife	0 (0.0)	21 (20.8)	21 (10.4)	0.000
	Student	79 (78.2)	10 (9.9)	89 (44.1)	
	Government worker	10 (9.9)	16 (15.8)	26 (12.9)	
	Private company worker	12 (11.9)	39 (38.6)	51 (25.2)	
	Unemployed	0 (0.0)	4 (4.0)	4 (2.0)	
	Retired	0 (0.0)	11 (10.9)	11 (5.4)	

### 3.3. Observation and knowledge related to *Aedes* mosquitoes' behavior and dengue

During a single night, the majority of respondents had experienced being bitten by mosquitoes for about 1 to 2 times with 35.1% for both urban and sub-urban areas, while 30.7% did not suffer mosquito bites. Respondents from the urban Sungai Dua were significantly less bitten (zero bitten at 45.5%) compared to the respondents in sub-urban Batu Maung at 15.8% ( $P < 0.05$ ; Table 2).

**Table 2**

Observation and knowledge related to *Aedes* mosquitoes' behavior and dengue. *n* (%).

Question	Sub-urban	Urban	Total	<i>P</i> -value
K1: Frequency of biting by mosquitoes during a single night				
> 5	12 (11.9)	17 (16.8)	29 (14.4)	0.000
3–4	27 (26.7)	13 (12.9)	40 (19.8)	
1–2	46 (45.5)	25 (24.8)	71 (35.1)	
0 (None)	16 (15.8)	46 (45.5)	62 (30.7)	
K2: Mosquito biting time				
12 am–4 am	37 (36.6)	14 (13.9)	51 (25.2)	0.002
4 am–8 am	2 (2.0)	7 (6.9)	9 (4.5)	
8 am–12 pm	5 (5.0)	5 (5.0)	10 (5.0)	
12 pm–4 pm	7 (6.9)	3 (3.0)	10 (5.0)	
4 pm–8 pm	18 (17.8)	30 (29.7)	48 (23.7)	
8 pm–12 am	32 (31.7)	42 (41.6)	74 (36.6)	
K3: Mosquito resting place in the house				
In the room	36 (35.6)	32 (31.7)	68 (33.7)	0.288
Toilet	15 (14.9)	16 (15.8)	31 (15.3)	
Kitchen	6 (5.9)	14 (13.9)	20 (9.9)	
Outside the house	44 (43.6)	39 (38.6)	83 (41.1)	
K4: <i>Ae. albopictus</i> transmit dengue viruses				
Yes	56 (55.4)	60 (59.4)	116 (57.4)	0.005
No	20 (19.8)	32 (31.7)	52 (25.7)	
Maybe	25 (24.8)	9 (8.9)	34 (16.8)	
K5: Increasing number of mosquitoes, increases the dengue cases				
Yes	82 (81.2)	74 (73.3)	156 (77.2)	0.095
No	5 (5.0)	14 (13.9)	19 (9.4)	
Maybe	14 (13.9)	13 (12.9)	27 (13.4)	

K: Observation and knowledge of *Aedes*'s behavior.

A total of 68.3% of respondent living in sub-urban experienced prolonged mosquito biting hours between 8.00 pm to 4.00 am, while respondents in the urban areas were mostly bitten between 8.00 pm to 12.00 am (41.6%). Based on the result in Table 2, most of the respondents in both areas found mosquitoes resting outside of their house area. This is followed by the room, toilet and kitchen with no significant differences between both urban and sub-urban ( $P = 0.288$ ).

More than 50% from the total of respondents in sub-urban were aware that dengue virus is transmitted by *Ae. albopictus*. Unfortunately, 31.7% of respondents in the urban Sungai Dua were not as well informed by disagreeing that *Ae. albopictus* did not transmit dengue virus. However, majority of respondent from both areas agreed with the statement that the rise in mosquito population will increase the number of cases of dengue cases (urban 73.3%; sub-urban 81.2%) and no significant differences were found for this question between both urban and sub-urban ( $P = 0.095$ ).

### 3.4. Dengue infection on respondents

Table 3 shows that a high number of respondents in sub-urban Batu Maung were infected with dengue virus (16.8%) compared to urban Sungai Dua areas (13.9%). However, 4.0% of respondents in the urban Sungai Dua experienced recurring dengue infection (more than two times). No significant differences were found ( $P > 0.05$ ) for the dengue infection occurrence and re-occurrence for both areas.

**Table 3**

Dengue infection on respondents. *n* (%).

Question	Sub-urban	Urban	Total	<i>P</i> -value
DI1: Infected with dengue virus				
Yes	20 (19.8)	14 (13.9)	34 (16.8)	0.259
No	81 (80.2)	87 (86.1)	168 (83.2)	
DI2: Frequency of dengue infection				
> 2	0 (0.0)	2 (2.0)	2 (1.0)	0.212
2	2 (2.0)	2 (2.0)	4 (2.0)	
1	18 (17.8)	10 (9.9)	28 (13.9)	
0 (None)	81 (80.2)	87 (86.1)	168 (83.2)	
DI3: Mosquitoes live longer when it is infected with DENV				
Yes	26 (25.7)	13 (12.9)	39 (19.3)	0.001
No	14 (13.9)	35 (34.7)	49 (24.3)	
Maybe	61 (60.4)	53 (52.5)	114 (56.4)	

DI: Dengue infection.

More than half of the total respondents in both areas lacked on the information and knowledge regarding the effect of dengue virus on the mosquito survivability, with 56.4% respondents chose unsure for this question. However, the result showed significant differences between these two areas ( $P = 0.001$ ) on dengue infection question with more respondent in sub-urban Batu Maung by agreeing with the statement above by 25.7% compared to urban Sungai Dua with only 12.9%.

### 3.5. Human lifestyles on the usage of light and *Aedes* behaviour

Table 4 shows the effects of human lifestyle on light usage towards

mosquitoes behavior. More respondents in urban areas believed that 24 h light-on in the food stall increased the chances of getting bitten by mosquitoes (42.6%), whereas the majority of respondent in sub-urban answered "maybe" (54.5%). There was a significant difference between both areas ( $P < 0.05$ ) regarding the question above. More than half of the total respondents from both areas agreed that light played a vital role in mosquito behavior, resulting in less mosquitoes bites if the light was switched off ( $P = 0.216$ ).

A majority of respondents (90.1%) in both areas usually switched off their bedroom light before going to bed. However, the respondents in both areas left the lights on in various other places in the house during night time with the kitchen was the main choice followed by toilet and living room. Respondents in urban areas are most likely to save energy by switching off all of the light in their house (46.5%) and significant differences were found between respondents' choice in these two areas ( $P = 0.000$ ).

**Table 4**

Human lifestyle on the usage of light and *Aedes* behaviour. *n* (%).

Question	Sub-urban	Urban	Total	<i>P</i> -value
HL1: 24 h light-on stall increases the chances of getting bitten by mosquitoes				
Yes	26 (25.7)	43 (42.6)	69 (34.2)	0.001
No	20 (19.8)	29 (28.7)	49 (24.3)	
Maybe	55 (54.5)	29 (28.7)	84 (41.6)	
HL2: Light alters mosquito behavior and make human less being bitten by mosquitoes				
Yes	46 (45.5)	58 (57.4)	104 (51.5)	0.216
No	32 (31.7)	27 (26.7)	59 (29.2)	
Maybe	23 (22.8)	16 (15.8)	39 (19.3)	
HL3: Switch off the light in the bedroom before going to bed				
Yes	91 (90.1)	91 (90.1)	182 (90.1)	1.000
No	10 (9.9)	10 (9.9)	20 (9.9)	
HL4: Other places leave with a light on during night time				
Toilet	22 (21.8)	23 (22.8)	45 (22.3)	0.000
Living room	23 (22.8)	12 (11.9)	35 (17.3)	
Kitchen	34 (33.7)	17 (16.8)	51 (25.2)	
Other place	3 (3.0)	2 (2.0)	5 (2.5)	
None	19 (18.8)	47 (46.5)	66 (32.7)	
HL5: Other factors attract mosquitoes to bite humans				
Carbon dioxide	4 (4.0)	10 (9.9)	14 (6.9)	0.002
Sweat	18 (17.8)	14 (13.9)	32 (15.8)	
Dark cloth	76 (75.2)	60 (59.4)	136 (67.3)	
Other factor	3 (3.0)	17 (16.8)	20 (9.9)	

HL: Human lifestyle.

Dark colored clothing (72.5% for urban and 59.4% for sub-urban) is considered as the most important factor attracting mosquitoes to bite human followed by sweat, carbon dioxide and other factors such as a dark place, exposure of skin by human, locations environment (humid and dark), bushes and type of blood chosen by respondents in both areas. There was a significant difference between these two areas for their choices on factors attracting mosquitoes to bite humans ( $P = 0.002$ ).

### 3.6. Preventive measures of *Aedes* mosquitoes

The majority of respondents with 60.4% sub-urban and 53.4% urban reported that fogging were rarely conducted in their residential area (Table 5). The majority of respondents in sub-urban and urban

areas agreed that more fogging operation will be administered in conjunction with the number of dengue cases. More than 46% of respondents in both areas believed that insecticide fogging operation is effective in killing mosquitoes.

**Table 5**

Preventive measure of *Aedes* mosquitoes. n (%).

Question	Sub-urban	Urban	Total	P-value
PV1: Frequent of fogging operations conducted				
Once a week	7 (6.9)	9 (8.9)	16 (7.9)	0.000
Once a month	16 (15.8)	23 (22.8)	39 (19.3)	
Twice a month	17 (16.8)	15 (14.9)	32 (15.8)	
Rarely	61 (60.4)	54 (53.5)	115 (56.9)	
PV2: More fogging activities during high reported dengue				
Yes	80 (79.2)	84 (83.2)	164 (81.2)	0.142
No	5 (5.0)	9 (8.9)	14 (6.9)	
Maybe	16 (15.8)	8 (7.9)	24 (11.9)	
PV4: Fogging operation effective in killing the mosquitoes				
Very effective	11 (10.9)	7 (6.9)	18 (8.9)	0.268
Effective	48 (47.5)	47 (46.5)	95 (47.0)	
Less effective	35 (34.7)	32 (31.7)	67 (33.2)	
Not effective	7 (6.9)	15 (14.9)	22 (10.9)	
PV5: For personal protection method against mosquitoes				
Aerosol	58 (57.4)	45 (44.6)	103 (51.0)	0.187
Mosquito electric mat	12 (11.9)	16 (15.8)	28 (13.9)	
Mosquito coil	31 (30.7)	40 (39.6)	71 (35.1)	
PV6: Excessive- use of mosquito insecticides will cause less death of mosquitoes				
Yes	18 (17.6)	42 (41.6)	60 (29.7)	0.000
No	25 (24.8)	31 (30.7)	56 (27.7)	
Maybe	58 (57.4)	28 (27.7)	86 (42.6)	
PV7: Insecticide will affect the development and survivability of mosquitoes				
Yes	29 (28.7)	48 (47.5)	77 (38.1)	0.000
No	11 (10.9)	24 (23.8)	35 (17.3)	
Maybe	61 (60.4)	29 (28.7)	90 (44.6)	

PV: Preventive measure.

As a personal protection against mosquitoes, 51% of total respondents from both areas chose mosquito aerosol spray as the main preventive method, and mosquito coil as the second option. A total of 57.4% of the respondents in sub-urban shared an opinion that neither agreed nor disagreed with the statement of excessive use of mosquito insecticide will cause less mosquitoes death, whereas 41.6% of urban are as respondents agreed with this statement. Similar answer was received from both urban and sub-urban areas for the statement of that insecticide could give an effect to mosquitoes fitness cost in terms of development and survivability.

Knowledgeable respondents in Sungai Dua urban areas on dengue and *Aedes* mosquitoes' behaviour were significantly related to age

and education, whereas the selection of preventive measure against *Aedes* mosquitoes significantly related to gender and age as showed in multivariate analysis ( $P < 0.05$ ; Table 6). Based on the survey distributed to the 101 respondents living in Sungai Dua urban area, multivariate model on frequency of biting by *Aedes* mosquitoes was influenced by education, dengue infection, human lifestyle using 24 h constant light which alter mosquito behaviour and increase the biting frequency. Nevertheless, 91% of the respondents in the urban areas preferred to switch off their bedroom light before sleeping to reduce mosquito bite. The frequency biting model is also influenced by fogging and personal protection used by respondents (all factors significant at  $P < 0.05$ ,  $y = 2.561$ ,  $-0.285$  dengue infection,  $-0.493$  education,  $-0.224$  human lifestyle 1,  $-0.670$  human lifestyle 3,  $-0.215$  preventive measure 4; Table 6). Biting time by *Aedes* mosquitoes was significantly related to age and preventive measure used by respondent (Table 6). Whereas, resting place for *Aedes* mosquitoes in the room (31.7%) and outside the house (38.6%) was significantly related to the use of lights ( $P < 0.05$ ; Table 6).

The situation in the sub-urban area was quite different with gender influencing the opinion on the fogging frequency. Meanwhile, the frequency of mosquito bite was related to the age of the respondent. People with higher education significantly believed that more fogging activities will be conducted during a high number of dengue cases ( $P = 0.044$ ). Based on the multivariate model summarize in Table 7, the frequency mosquito bites during a single night was influenced by the observations on the capability of *Ae. albopictus* to transmit dengue viruses and the frequent of fogging operations in the residential sub-urban area of Batu Maung ( $y = 1.747 + 0.289$  observation and knowledge of *Aedes*'s behavior  $K4 + 0.216$  preventive measure  $P1$ ;  $P = 0.01$ ). The mosquitoes biting time were significantly influenced by the statement that insecticide will affect the development and survivability of mosquitoes (estimated = 0.553;  $P < 0.05$ ; Table 7). Resting place of mosquitoes in sub-urban area indicated preference outside of the house (43.6%) and inside the room (35.6%) was significantly related to the number of respondents infected with dengue and the frequency of being infected with dengue virus during their lifetime. Also, significantly related to the age of the respondent with the majority of age is between 17 and 24 (78.2%). ( $P < 0.05$ ; 0.204 age, 0.85 dengue infection 1, 1.189 dengue infection 2; Table 7).

**Table 6**

Best fit multinomial regression model for respondents in the urban Sungai Dua area for demographic characteristic (gender, age, education) and observation and knowledge on *Aedes*'s behaviour on affecting dengue infection, human lifestyle, and preventive measure.

Factor	Model	Adjusted R <sup>2</sup>	F	P-value
Gender	$y = 1.320 \text{ int}^* + 0.846 \text{ DI1} - 0.593 \text{ DI2} + 0.040 \text{ K5} - 0.182 \text{ K5} + 0.198 \text{ PV3} - 0.102 \text{ PV4}$	0.158	4.119	0.001
Age	$y = 0.596 \text{ int}^* + -0.0842 \text{ K2} - 0.164 \text{ K4} + -0.348 \text{ PV3}$	0.175	3.117	0.002
Education	$y = 1.405 \text{ int}^* + -0.0807 \text{ K1}$	0.073	2.965	0.023
The frequency of being bitten by mosquitoes	$y = 2.561 \text{ int}^* + -0.285 \text{ DI3} + -0.493 \text{ Education} + -0.224 \text{ HL1} - 0.670 \text{ HL3} + -0.215 \text{ PV4}$	0.204	4.651	< 0.001
Mosquito biting time	$y = 1.749 \text{ int}^* + 0.861 \text{ Age} + -0.616 \text{ PV3} + 0.716 \text{ PV4} + -0.476 \text{ K4}$	0.114	2.836	0.010
Mosquito resting place	$y = 2.255 \text{ int}^* + -0.320 \text{ HL1}$	0.052	2.815	0.043

\*Int: Intercept; all possible two-way interaction; \*K: Observation and knowledge of *Aedes*'s behavior; DI: Dengue infection; HL: Human lifestyle; PV: Preventive measure; \*P-value is based on 2 log likelihood regression using backward stepwise method.

**Table 7**

Best fit multinomial regression model for respondents in the sub-urban Batu Maung area for demographic characteristic (gender, age, education) and observation and knowledge on *Aedes*'s behaviour on affecting dengue infection, human lifestyle, and preventive measure.

Factor	Model	Adjusted R <sup>2</sup>	F	P-value
Gender	$y = 0.891 \text{ int}^* + -0.167 \text{ Occupation} + 0.101 \text{ PV1}$	0.320	8.855	< 0.001
Age	$y = 1.424 \text{ int}^* + -0.843 \text{ Education} + 0.321 \text{ Occupation} + 0.261 \text{ K1}$	0.251	4.716	< 0.001
Education	$y = 1.482 \text{ int}^* + -0.117 \text{ Age} + 0.090 \text{ Occupation} + 0.210 \text{ PV2}$	0.162	3.409	0.001
The frequency of being bitten by mosquitoes	$y = 1.747 \text{ int}^* + 0.289 \text{ K4} + 0.216 \text{ PV1}$	0.080	3.901	0.010
Mosquito biting time	$y = 3.191 \text{ int}^* + 0.554 \text{ PV6}$	0.118	3.685	0.004
Mosquito resting place	$y = -1.588 \text{ int}^* + 0.204 \text{ Age} + 0.857 \text{ DI1} + 1.189 \text{ DI2}$	0.164	4.275	< 0.001

\*Int: Intercept; all possible two-way interaction; \*K: Observation and knowledge of *Aedes*'s behavior; DI: Dengue infection; HL: Human lifestyle; PV: Preventive measure; \*P-value is based on 2 log likelihood regression using backward stepwise method.

Level of knowledge was significantly different between the sub-urban and urban area ( $\chi^2 = 7.833$ ;  $P = 0.020$ ; Table 8). Respondent in the urban area showed slightly better in knowledge about *Aedes* and dengue as compared to the sub-urban respondent. However, more than half of the respondents showed poor knowledge for both urban and sub-urban areas (urban 50.5%; sub-urban 69.3%).

**Table 8**

Knowledge level of respondents in the urban and sub-urban area about *Aedes* and Dengue based on Bloom's cut off point analysis. n (%).

Knowledge level	Sub-urban	Urban	Total
Good	5 (5.00)	11 (10.90)	16 (7.90)
Fair	26 (25.70)	39 (38.96)	65 (32.20)
Poor	70 (69.30)	51 (50.50)	121 (59.90)
Total	101 (100.00)	101 (100.00)	202 (100.00)

#### 4. Discussion

There were limitations to this study that need to be considered. Most of the respondents in sub-urban Batu Maung were secondary students from Batu Maung high school due to the lack of cooperation from the residents in the Batu Maung area. While in urban Sungai Dua, the surveys were done in residential areas which consist of more various ages of respondents. Input from respondents of various ages among residents in Batu Maung would give a clear indication on their knowledge, lifestyle and preventive measure on dengue and *Aedes*. Younger respondents might have lacked of experience towards *Aedes* knowledge compared to the adult respondents who have had more exposure about mosquitoes.

The Cronbach's alpha value on the knowledge questionnaire indicates a low reliability due to the questionnaire was based upon variety cognitive information and it was affected by the age difference from two sub-distinct areas and unlike the rest of the question; the questions were made up of unconnected knowledge points. The greater variety of knowledge responses could affect the value of Cronbach's alpha. However, the low reliability value were not common, but it does not indicate the validity of the questionnaire[15].

Respondents in sub-urban area was bitten by *Aedes* mosquitoes more frequently than the respondents in the urban areas. The risk of dengue fever outbreak is higher for the residence in the sub-urban area. A mathematical model by Kyle and Harris (2008) showed the basic reproductive rate of dengue virus (Ro) measures the

transmission capacity of viruses which includes the human-biting rate of the mosquitoes. The human-biting rate of mosquitoes is a key variable in dengue virus transmission and often defined as the average number of secondary transmission caused by the infectious individual[16]. Our study found that the three main factors linked to mosquitoes biting frequency are the changes in human lifestyle, in which by using 24 h light-on, preventive measure selected by the respondent and the frequency of fogging in the urban areas. Whereas for sub-urban areas, only preventive measure were linked to the mosquitoes biting frequency. A study by Oki *et al.*[17] indicated that the fogging operation will reduce the dengue cases, if conducted immediately after the reports of outbreak, thus reducing the natural epidemic of dengue. There was an abrupt immediate reduction in the indoor resting density of *Ae. aegypti* when peridomestic fogging was conducted, which significantly reduced about 50% of the parity rate of the indoor landing mosquito populations and the indoor human landing collection on the day after fogging treatment[18].

People in the sub-urban Batu Maung experienced a longer mosquito biting hour starting from 8.00 pm to 4.00 am. There are two possible reasons: (1) *Ae. albopictus* showed a peak of diurnal biting activity after sunrise and before sunset[19] and, (2) peak biting activity of *Aedes* during early morning in the secondary forest area[20]. In which, the sub-urban Batu Maung is surrounded with high coverage of vegetation areas, unlike the urban Sungai Dua area. There is also a possibility of only a small shift in the biting time cycle of *Aedes* mosquitoes because mosquitoes were already inhibited inside the house. As reported by Chen[21], the peak diel biting of *Aedes* occurred from 600–900 h in the morning. Other than that, the respondents in the sub-urban area might have mistook the *Aedes* species of mosquitoes with another dominant species, *Culex quinquefasciatus*. The peak biting activity of this nocturnal *Culex* species started at 1800 h, reaching the peak at 2400 h and gradually decreased till 600 h[22]. The selection of the preventive measure used by respondents, time of applying the insecticide and the method of applying the insecticide may influenced the biting time of mosquitoes.

The majority of respondents in both areas stated that mosquitoes are mostly resting outside their house area. *Ae. albopictus* was observed resting in canopies, open clearings or water containers[23]. High density of mosquitoes found outdoor were influenced by the presence of vegetation which clearly provided a suitable resting

place for *Aedes* mosquitoes[24]. Our study suggested that the usage of 24 h lighting inside the house may influenced the *Aedes* mosquitoes. The mosquitoes were outside the house as they preferred to rest in the dark places. Sarwar *et al.*[25] found mosquitoes response towards light is negatively phototactic and the majority of mosquito species is fully attracted by dark and shaded areas[26].

A total of 72.2% respondents in both sub-urban and urban areas agreed that an increase in mosquito population will increase the number of dengue cases. Higher number of mosquitoes will further increase human-mosquito contact[27]. Several studies showed in the regions where dengue is present, vector abundance and diseases incidence rise measurably accompanied by an increase in the temperature and humidity[28]. Other factors such a temporal change in the population immunity or the introduction of new serotypes can significantly affect dengue cases along with the increase in mosquito population[28,29]. Although more than 50% respondents in both areas were aware that dengue virus is transmitted by *Ae. albopictus*, they lacked on the knowledge regarding the behavior of *Ae. albopictus* species such as biting time, and the effect of insecticide on mosquitoes. Both of *Ae. albopictus* and *Ae. aegypti* are the main epidemic vector of DENV and Chikungunya virus worldwide[30,31].

A total of 19.8% respondents in Batu Maung and 13.9% in Sungai Dua have been infected with dengue virus. Higher number of female respondent (aged 19 to 24) was infected with dengue. Age is found to be a significant risk factor associated with dengue seropositivity. Dengue seroprevalence in human will increase for every 10 years of age. Older humans faces a higher chance of being infected with the virus[31]. Gender is one of the contributing factors towards the risk of severe dengue and death, with female and children are mainly victims of several series of dengue shock syndrome cases[32]. The reasons for the majority of females amongst severe and fatal cases is suggested due to differences in health-care seeking behavior. Women were reluctant to seek early medical care, leading severe condition[33]. The physiological and immunological difference exists between gender may explain the high infection among women[32]. The respondents live in the urban Sungai Dua have experienced recurrence of more than 2 times of dengue virus infection. Sungai Dua area has been known as dengue hotspot area in Penang as reported by Mohiddin *et al.*[34]. Dengue is caused by 4 distinct dengue virus serotypes (DENV 1, 2, 3 and 4) which are transmitted to human by *Aedes* mosquitoes[35] and the infection with one serotype is capable of producing life-long immunity to to specific serotype, but only a temporary immunity to other serotypes[36]. Regional data for 2002 to 2012 from the Sentinel Surveillance System in Malaysia reported a heterogenous distribution of DENV serotypes in Malaysia, with DENV 4 dominating in Penang, followed by DENV 3 and DENV 1[37]. It is possible that the 4% respondent might be infected by different types of dengue viruses in their lifetime.

A total of 42.6% of the urban respondents in Sungai Dua believed that 24 h lighting from food stalls might increase the chances of

getting bitten by mosquitoes. While 54.5% respondents in the sub-urban Batu Maung stated unsure on this hypothesis. The different response to this statement may be due to difference in community structure. The urban Sungai Dua is surrounded with 24 h food stall but not in the sub-urban Batu Maung. Beck[38] believed that flight activity in *Ae. aegypti* appears to be controlled by an endogenous rhythm that may be phase-set by both light-on and light-off. It is possible that 24 h light-on might increases the chances of getting bitten by mosquitoes because a wider chance of the exposure to mosquitoes due to respond to light intensity level influencing the mosquitoes flight activity[39]. If the insects tends to avoid the light, whether a diffuse or a beam of artificial light, it is considered as negatively phototactic. However, regardless of their negative phototactic responses, some of mosquitoes response to artificial light[40] leads to an exploited design of an extremely efficient traps for mosquito surveillance.

The respondents have a good knowledge regarding the factors that attracts mosquitoes to bite human in both areas. *Ae. aegypti* appears to be attracted to dark objects such as dark clothing or clothes hanging on a wall or dark surfaces[41], human breath (carbon dioxide), sweat and urine[42]. Sweat from armpit, hand or forearm encourages a higher rate of probing by *Ae. aegypti*, suggesting secretions of both apocrine and erine sweat glands was stimulated[43]. As a preventive measure for personal protection, most of the respondents in both areas used mosquito aerosol spray as it is easy to apply followed by mosquito coil and mosquito electric mat. In India, the majority of people preferred a mosquito electric mat[44], and mosquito coil[45,46]. The dengue guidelines by World Health Organization[47] suggested when indoors mosquito biting occurs, the control such as household insecticides aerosol product, mosquito coil and other insecticide vaporizer can be used to reduce biting activity of mosquitoes. The main tools combating mosquitoes borne-disease also includes the use of adulticides of space spray (*i.e.* fogging) and applications of larvicides to aquatic habitat such in pool, and water reservoirs[48].

The respondents in the urban Sungai Dua might have experienced and witnessed that the excessive use of mosquito insecticide will lead to less death of mosquitoes and make them better observers as compared to the respondents in Batu Maung. There is a possibility that the respondents have never been exposed about this information by the government or experts. Chemical insecticide is capable of developing resistance to the mosquitoes after few generations. In the long term, mosquito adaptation causes them become stronger than before[49]. Insecticide resistance happens when a population of insect is subjected to insecticide exposure over a long time or at a very frequent rate[50].

The respondent for both urban and sub-urban areas reported fogging rarely conducted in their residential area. However, more fogging has been done in the Sungai Dua area as this area is a well known as the dengue hotspot area. The normal fogging activities were conducted every three days during the dengue outbreak until

the outbreak is considered over by the Ministry of Health Penang. The effectiveness of fogging is only for a limited amount of time and must be continuously sprayed[49]. It is possible that the respondents were not aware the fogging activity in their residential area because they received no notification or they had missed the notice. Fogging is usually conducted during working day before the respondent reached home from work. Fogging operation will be administered as soon as dengue cases reported in the area based on level two of dengue emergency response[47].

It can be concluded that the overall knowledge about mosquitoes and dengue is at a poor level, especially those living in the suburban areas. The knowledge among students need to be improved by increasing awareness about dengue either at school level or in the social media. More exposure to the information regarding the effect of excessive-use of insecticide, and the effect of DENV viruses to mosquitoes should be handed out to the public either by the government or private institution to ensure they have further understanding on the dengue transmission and resistance problems. Closing the gap between knowledge, current lifestyle and preventive measure can further reduce the *Aedes* mosquito populations and to curb the dengue infections among people.

### Conflict of interest statement

We declare that we have no conflict of interest.

### Acknowledgments

The authors would like to thank for all support and technical assistance during this project. We are grateful to the volunteers and residence from all two localities for their active participation in this survey. This project was partially funded by the Fundamental Research Grant Scheme by the Ministry of Education and Universiti Sains Malaysia (203/PBIOLOGI/6711359).

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