RESEARCH ARTICLE

Potential of aedes breeding site with different types of container at plant nurseries in Klang

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Abstract:

Dengue fever which transmitted by the Aedes has been one of the most widespread vector-borne disease. The most important thing in reducing dengue infection cases is by controlling the populations of the *Aedes* mosquito by destructing *Aedes* breeding sites. Aedes breeding sites can be determined by conducting a larval survey. Most of the studies conducted were focusing on residential area. However, the mushrooming nursery within housing vicinity has become potential breeding places for aedes since it provides potential artificial and water source to the vector. There were nine plant nurseries have been observed, overall cleanliness and distance from nearest housing area were measured. During the observation, pots, plates, tires, and bucket mostly found in nurseries and may harboring Aedes, hence this study has focused on the container survey and larval collection. Noteworthily, a total of 68 containers were found positive for breeding sites, and most likely the artificial container with larger surface area harboring more than lesser surface area. Adding up to that situation, the relationship between the type of container and likelihood of breeding potential has scored a great observation too. To conclude, aedes breeding places in the nearby nursery might increased potential dengue fever transmission for nearby residences.

Keywords: Aedes breeding site, artificial container, container survey, plant nursery.

1. INTRODUCTION

Dengue fever is considered as one of the most widespread vector-borne diseases which has affected 128 countries in this world (Brady et al., 2012). According to the Ministry of Health, the total number of dengue cases in Malaysia for 2018 is 80,615 cases being reported with 147 deaths due to dengue. Most dengue cases occurred in Selangor state with 45,349 dengue cases followed by Federal Territory of Kuala Lumpur and Pulau Pinang with 7,591 cases and 6,071 cases, respectively. Due to the increase of the dengue cases in Malaysia, the most important thing in reducing the dengue infection cases is by controlling the populations of the Aedes mosquito since the transmission requires Aedes mosquito as the vectors. Hence, the control should be targeting the habitats that are more productive such as the container characteristic that led to the breeding of the Aedes. Aedes larvae usually can be found either in natural or artificial containers. The common container for the breeding site of the Aedes can be found discarded tires, flowerpots, flowerpot plates, rain barrels and other places that have stagnant water which is a suitable place for the mosquito to lay their eggs (Faiz Madzlan et al, 2016). Usually, the larval survey will be conducted at the housing area since it is near with the human population. Plant nursery can be an example of a potential Aedes breeding site as they have many artificial containers such as flower pot plates, rain barrels and canvas sheets. Besides that, Petaling Jaya City Council (MBPJ) Health and Environment Department director Dr Chitra Davi N. Vadivellu has said that Aedes larvae also can be found at the plant leaves that can hold water. Some studies have proved that Aedes sp. especially Ae. aegypti can breed in plant leaves such as ornamental bromeliads (André B. B. Wilke, 2018).

In response to this problem, this study aims to identify the potential Aedes breeding site at plant nursery. The purpose of this study also can help to identify the relationship between the type of container and larvae populations. Several plant nurseries in Klang will be selected for this study and the premise will be inspected to identify the potential breeding site and number of positive containers.

Reducing the Aedes breeding site is important in order to reduce the Dengue cases in an area. Identification of the Aedes breeding site is usually being conducted at the residential area since it is close to the community. Gardening

utensils were found as the common breeding site for the Aedes which include flower pot, flower pot plates and watering cans (Ramlee, 2013). However, these things also commonly can be found at the plant nursery. There is not much study being conducted at the plant nursery because most of the study is more focused at the community area such as residential area or institutional. Besides that, some of the ornamental plants such as Bromeliads has been proved can be a breeding site for Aedes (Wilke, 2018). This has increased the number of potential breeding sites at the plant nursery. So, this study is important to identify any natural or artificial breeding site that can be found in a plant nursery and if this potential breeding site can give impact to the Aedes population. Besides that, it also can help to educate the owner of the plant nurseries about the potential breeding sites of Aedes in the plant nurseries.

2. MATERIALS AND METHODS

The aim of this study is to identify the potential breeding sites of Aedes species at plant nursery. This study will be carried out using the quantitative research approach. Under the quantitative research approach, the observational research design that will be used for this study is the cross-sectional study. Setia (2016) has referred to the cross-sectional study as the measure of the outcome and the exposure in the study participant at the same time. The participant for this study will be selected using the convenience sampling where any plant nurseries in the study area that are able to take part for the study will be included. All the plant nurseries that are willing to take part in this study will be observed and inspected for the potential breeding sites.

The method of data collection that will be used for this study is survey and larval collection. The data will be collected via survey of all the containers either artificial or natural containers (Chen, 2009). The information such as type of container, number of potential containers, and container with positive breeding will be recorded. Then, the larvae will be collected using the dipping or pipette and will be placed in a universal bottle Each of the larvae samples taken will be labelled with the location being found and type of container. Next, the identification of larvae will be taken in the lab to identify the species of the larvae. The identification of the larvae has to be conducted to classify either it is Aedes aegypti, Aedes albopictus or others.

Based on data collection that has been made, descriptive analysis will be used to summarize the number of larvae collected at the different location and type of container found. Percentage of the potential breeding sites found and positive breeding sites will be determined to identify the highest type of container that could be the breeding site for the Aedes. Besides that, the index of available container (IAC) will be calculated for any potential breeding sites and Breeding Preference Ratio (BPR) will be calculated for any positive breeding site using above formula: -

$$\text{Breeding Preference Ratio (BPR)} = \frac{\text{Index of Contribution to Breeding Sites (ICBS)}}{\text{Index of Available Container (IAC)}}$$

$$\text{Index of Contribution to Breeding Sites (ICBS)} = \frac{\text{Amount of positive container for each type}}{\text{Total amount of positive container}}$$

$$\text{Index of available container (IAC)} = \frac{\text{Amount of potential container for each type}}{\text{Total amount of potential container}}$$

3. RESULTS AND DISCUSSION

3.1. Cleanliness and the distance of the plant nurseries from residential area

There are nine (9) plant nurseries have been observed in Kapar, Klang by using the cluster purposive sampling. Only nurseries with less than 100 meters distance were chosen with minimum size of 20-meter X 20-meter area. Nursery with lesser operating area were not included as they might be only a temporary plant storage area and does not have any permit of operation. Cleanliness was observed based on the plants and material arrangement and tagging area. The housekeeping and its roster were also observed and scored. Relative cleanliness rubric adapted from Rahman et al (2021) has suggested the score less than 2 is considered poor while score more that 4 is good.

In general, 33.30% of nurseries observed were in poor condition (Table 1). The indication has showed that lack of staff in maintaining the area as well as lack of awareness on the housekeeping benefit has potentially increased the possible Aedes breeding area due to less observation given to the water retention container. On top of that, cleanliness and proper housekeeping may reduce the possible aedes breeding places should the container were stacked properly and less expose to rain or plants watering activities. Poor housekeeping was also observed with the container were located with direct exposure to the rain and open, under the flowerpot rack. According to study by Rahman (2021), containers that located at outdoor in vegetation area has a higher chance for producing larvae. This is because this environment can enhance the mosquito breeding by enabling a resting site for the female mosquitoes to lay eggs and provide sugar feeding resource for larvae.

Table 1. Information regarding the plant nurseries

No.	Plant Nursery	Distance from nearest housing area (m)	Cleanliness
1	Plant Nursery 1	32	Good
2	Plant Nursery 2	22	Good
3	Plant Nursery 3	29	Good
4	Plant Nursery 4	45	Poor
5	Plant Nursery 5	98	Good
6	Plant Nursery 6	29	Good
7	Plant Nursery 7	44	Poor
8	Plant Nursery 8	20	Poor
9	Plant Nursery 9	69	Good

3.2. Determination of potential Aedes breeding sites at plant nurseries

Potential breeding places for Aedes can be varies between all possible artificial containers or possible natural, plant-based containers. In this study, the observation in the nurseries has leading to the tabulation data on possible containers for breeding places (Table 2). Throughout the observation, there were three type of container were mainly observed to be 'everywhere' in the nurseries, namely, flowerpot plates, bucket, and tires with the total number of 10, 13 and 13 respectively. It was noted that these items are essential for nurseries operation and thus, each nursery may or may not possibly turn these type of containers as potential aedes breeding places depending on the handling and upkeep of the surrounding area.

It was also observed that 14 bromeliads plants were found and observed to be highly potential as breeding places in 2 different nurseries. Bromeliads is a species of plants that have stackable – hard -leafy characteristic which may keep the water between their leaves. The water from plant watering, rain or from dews may collected at the heart of the plant thus provides good water source for breeding places. Maintaining bromeliads to be free as breeding places considered labour intensive thus need more workers to take care of this plants. Other container which was also found during the survey are plastic barrel, plastic bottle, watering can, canvas, and artificial ponds. These items also considered as highly potential breeding places due to their capacity that may have water retention capacity.

It was noteworthy that, most of the artificial container that has been inspected containing rainwater since it was a rainy season during the study period. Overall classification of natural and artificial container type found was depicted in Figure 1.

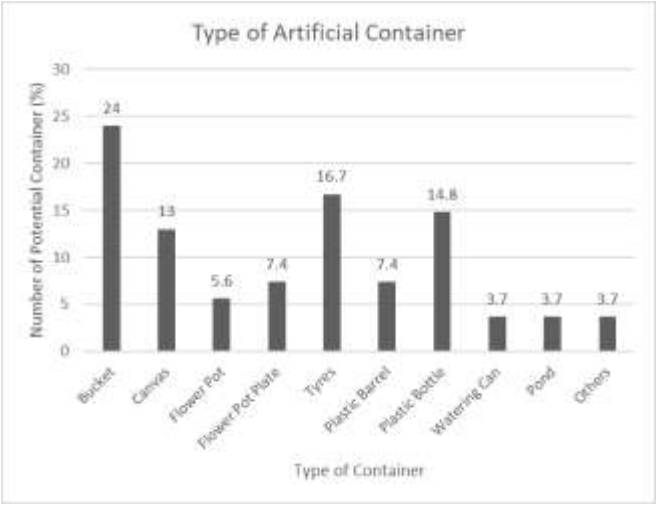

Figure 1. The percentage of potential artificial breeding places

From the result obtain for the container survey, bucket has the highest percentage for artificial container that can act as a potential breeding site with 24%. This is because most of the plant nurseries being observed will have bucket in their premise. This bucket usually will be used as a watering medium for the plant at the premise. Improper storage of the bucket could lead to higher chance to be a breeding site for the aedes. In this study, this type of container can was proved can be a breeding site for the aedes when one of them were positive with aedes albopictus. The stagnant water in the breeding container were due to the rainwater since the container were placed at an open area. Next, in this study tires were known as the second highest to act as the potential breeding sites for the aedes.

Table 2. Potential breeding sites at plant nurseries

Nurs	Type of Container												Potential Container	Container (+ve) Breeding
	Natural Container		Artificial Container											
	Bromeliads Plant	Others	Flowerpot	Flowerpot plate	Tyres	Canvas	Bucket	Plastic Barrel	Plastic bottle	Watering Can	Pond	Others		
1	10	-	-	1	1	5	-	3	-	-	2	1	22	2
2	-	-	-	1	-	1	-	-	-	-	-	-	2	1
3	4	-	2	-	5	-	-	-	-	-	-	-	11	2
4	-	-	-	3	-	-	3	-	-	-	-	-	6	-
5	-	-	-	-	4	-	3	-	-	-	-	1	8	3
6	-	-	-	-	-	-	2	1	-	-	-	-	3	1
7	-	-	-	2	-	-	-	-	-	2	-	-	4	-
8	-	-	1	-	2	1	3	-	8	-	-	-	15	-
9	-	-	-	3	1	-	2	-	-	-	-	-	6	2

Even though there were no positive breeding for this type of container in plant nursery, study by Lenhart (2006) has recommend being more aware on this type of container since it can produce up to 70% of aedes pupae especially during the rainy season. Adding up, a study by Gautam (2012) has proved that discarded tires can be a high-risk container since it also can be a breeding site for aedes during pre-monsoon and post monsoon season. Next, plastic bottle has the percentage of 14.8% to be accommodate in the plant nursery. The high percentage of plastic bottle in the plant nursery are due to the half-cut plastic bottles use at the legs of the shelves. This is a method used by the nursery owner to prevent the shelf leg from sinking into the muddy grown. Besides that, improper disposal of plastic bottle in the premise also contribute to the high percentage of plastic bottles

Next, a total of 13% of the potential place for breeding are from the canvas used to cover the soil at the plant nursery. The water accommodate on the canvas are due to the rainwater as well plant watering activities. However, this situation might be different during hot season since the water may be evaporated to the atmosphere, hence reducing the possible breeding places. Basically, improper placement of flowerpot plate can lead to water contained in it. The owner should place the unused flowerpot plates in the closed area or keep it in upside down manner to reduce the risk of being a harboring place for Aedes to lay their eggs with water in it. Other than that, plastic barrel used in the plant nursery to keep water for the plant can be a potential container since the plastic barrel does not been closed properly and the usage of abate is considered compulsory in preventing immature stage of aedes mosquito to develop. Lastly, container such as watering can, pond and other categories has the lowest percentage which are 3.7%. However, there is low possibility for the Aedes mosquitoes to breed if the container is regularly clean and practice a proper storage for the container to avoid any possibility for the Aedes mosquitoes to breed.

3.2. Calculation of Epidemiological Indexes by Container Classification

During the larval survey, there were 11 positive Aedes container were found in different artificial container which are flowerpot plates (3), tires (5), bucket (2) and barrel (1) respectively.

The larvae were identified as aedes genes since it has 2 lateral spines on each side of the thorax and a straight row of 7 to 12 comb scales at the 8th abdominal segment. The larvae were identified as *Aedes albopictus* since the comb scales exhibit a medical spine without subapical spines. Henceforth, in due to these expected findings, the calculation of the index of available container (IAC) was tabulated in Table 3. It was clearly evidence that ICBS of Aedes breeding in this study that focusing on the nine nurseries observed showed that the highest index contribution for breeding places is tires (0.4), followed by plastic barrel (0.25), and bucket (0.15) respectively.

Table 3. Index of available container by container classification

Characteristic	Type of container	Total number of potential containers	Total number of breeding sites	ICBS
Natural container	Bromeliad's plant	14	-	0
Artificial container	Bucket	13	2	0.15
	Canvas	7	-	0
	Flowerpot	3	-	0
	Flowerpot plate	10	3	0.

Tires	13	5	0.4
Plastic barrel	4	1	0.25
Plastic bottle	8	-	0
Watering can	2	-	0
Pond	2	-	0
Others	2	-	0

However, previous study related to container preference shows that bucket is known as the most frequent breeding sites. This is because the bucket has the capacity to contain considerable volume of water for the Aedes to breed (Nicolás Flaibani, 2020). Besides that, area of the opening surface for the container is large enough for the female mosquitoes to rest and breed on it while for bromeliads plant, even though it has the highest IAC, the probability for the mosquitoes to breed is low due to insufficient volume of water that contain in it. This can contribute to larval mortality due to lack of food, overcrowding and other sources of environmental stress (Shultis, 2009).

In this study tires have shown the highest ICBS with the reading of 0.4 in overall scoring. This number indicate that, among the highest potential in observed nurseries, it has 4-time higher chances in becoming the breeding places for Aedes. Similar finding was also observed by previous study by Dila (2020) and Rahman (2021) which were found that unused tires are among the highest for the contribution of Aedes breeding sites. This is because water that stored in used tyres are protected from the sunlight which make it more suitable places for the Aedes to breed since the female Aedes mosquitoes preferred to rest in less lighting area or shaded area (Dejene et al.,2015).

Next, container such as plastic barrel found in the plant nursery also has a substantial index of ICBS with score of 0.25, where it indicates that 1 out of 4 barrels found, there was a positive aedes breeding evidence in this study. This statement can be proved by previous study where the water barrel was also found to be the most frequent potential container even, so it was least found to have positive breeding as their opening surface of the barrel lead to less detectable containers (Nicolás Flaibani, 2020).

Overall observation and analysis from this study has indicated that containers found in the nurseries area could be potentially becoming Aedes breeding places. This substantial finding on the aedes breeding places as well as the cleanliness and distance from the nearest residential area has put an alarming situation if the nurseries was not taken into consideration for optimum prevention and control action by the authority. The fact that mosquito optimum flight range is between 200 meters and up to 2 kilometers with the help of transport may put community in alarming situation day by day. According to WHO, aedes mosquito could fly up to 400 meters to look for any water-filled containers to lay their eggs.

The good source of breeding container and water source in nurseries may lead to hidden breeding places should the authority only relying on the address basis of the dengue fever patient. It is important to keep an eye on possible capacity of 'less explored' area such as nurseries in their preventive and control action. Fogging and larvicidal activity should not only be focusing to the dengue prone residential areas, but to extend the capacity of possible breeding places within the vicinity of confirm dengue cases area.

4. CONCLUSION

This study helps to identify any natural and artificial container that has a potential to act as a breeding site for Aedes. Some of the container that can be found in the plant nursery include bucket, discarded tyres, plastic barrel, flower pot plate, flower pot, watering can, pond, plastic tray and water tank. Artifical container are found in the plant nursery has a potential to be a contributor for the increasing of Aedes population. Most of the potential container detected are due to the rain water or container that used to keep water for the used in plant nursery which is the best condition for Aedes to breed. Besides that, the relationship between the type of container and positive breeding site has been determine where they likely to breed in the container that has large surface area such as bucket and shaded area such as tyres and bucket. This study is conducted during rainy season since previous study shows that the population of Aedes larvae are the highest. Future consideration for determining regression of distance of nurseries to the number of confirm dengue cases might give a clearer view on possible hidden, less explored, less human density plant facilities onto which capabilities they may cause the dengue fever outbreak escalates time to time.

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