#### A comparison of different types of ovitraps for outdoor monitoring of *Aedes* mosquitoes in Kuala Lumpur

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Abstract. Dengue is a significant public health problem in Malaysia and vector surveillance is one of the important components in a vector control program. Routinely vector surveillance in Malaysia is performed through larval surveys. However, larval surveys have several limitations. Thus ovitraps are used as an alternative method for monitoring dengue vectors. The aim of this study was to determine the effectiveness of Standard Ovitrap (SO), Mosquito larvae Trapping Device (MLTD), Double Sticky Ovitrap (DST) and NPK Fertiliser Trap to monitor the abundance of Aedes mosquitoes. Each ovitrap was placed at four different sampling points and rotated to the next position every one week. Larvae and adult mosquitoes were collected and identified in the laboratory. All four trapping methods successfully collected larvae of Aedes mosquitoes. The mean number of larvae per ovitrap in DST was significantly higher (p < 0.05) compared to SO and MLTD. DST and NPK Fertiliser Trap were capable of capturing adult mosquitoes. Ovitrap Index and the mean number of adults per ovitrap in NPK Fertiliser trap were significantly higher (p < 0.05) as compared to DST. Another set of experiments were conducted to compare NPK Fertiliser traps containing fresh NPK fertiliser solution with those containing stock NPK Fertiliser solution. The fresh solution was prepared fresh while the stock solution was stored for a month before being used in the field. Result shows no significant differences (p > 0.05) between these solutions. Findings from this study conclude that DST is the most effective ovitrap to monitor Aedes larvae, while NPK Fertiliser trap is the most effective ovitrap to trap Aedes adult mosquitoes in the study area.

#### INTRODUCTION

Dengue fever has become a major global public health problem in terms of high morbidity, mortality and economic burden to communities and health services (Juni et al., 2015). Malaysia is among Western Pacific countries that had the greatest burden of dengue (WHO, 2008). According to iDengue website (2018), 6543 dengue cases were reported in 1995 and such cases progressively increased to 83,849 in 2017. Aedes aegypti (Linnaeus) and Aedes albopictus (Skuse) are the main vectors and responsible for the transmission of dengue in Malaysia (Nazri et al., 2013).

Vector surveillance is one of the important aspects in the prevention and control of dengue (Nazri et al., 2013). In Malaysia, vector surveillance is performed through larval surveys that enable calculation of House, Container and Breteau Index (Chang et al., 2011). According to WHO (2009), the aim of larval surveys is to detect the presence of larvae and pupae of Aedes mosquitoes in water-holding containers or fixtures. Mosquito Larvae Trapping Devices (MLTD) does not only function as an autocidal ovitrap but also as a tool for vector surveillance (Ariffin et al., 2009).

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However, the larval surveys used in the vector surveillance are not without disadvantages. Larval surveys are incapable to measure the risk of the spread of dengue and do not show the adult mosquito population in an area (WHO, 2009). Study conducted by Azil *et al.* (2014) reported that the health team faced difficulties to enter private premises for larval surveys. Thus, another effective method is required to overcome the weaknesses of the larval surveys.

Various ovitraps have successfully been produced and tested against *Aedes* mosquitoes for years (Snetselaar *et al.*, 2014). Ovitraps such as Standard Ovitrap and MLTD can be used for sampling of immature stages (larvae and pupae) whereas ovitraps such as Sticky Ovitrap and Double Sticky Ovitrap can be used for sampling of adult mosquitoes. However, the various ovitraps have their own advantages and disadvantages. Thus, comparative studies for the ovitraps are warranted.

The sensitivity of an ovitrap used as surveillance tool to detect the presence of Aedes mosquitoes can be increased with the use of attractant (Focks, 2003). Among the attractant used is the hay infusion. In a study conducted by Roslan et al. (2013) found that ovitrap containing hay infusion is not suitable to be placed in or nearby houses because of its unpleasant smell. Hay infusion is also difficult to produce and be brought to the field (Ritchie, 2001). Earlier studies conducted by Darriet & Corbel (2008), Darriet et al. (2010) and Anderson & Davies (2014) reported that NPK fertiliser has potential as attractant for Aedes mosquitoes. NPK fertiliser which is readily available and its solution, which is easier to prepare, becomes an alternative to hay infusion. However, the study on its effectiveness is scarce and no such study has been carried out in Malaysia.

Besides, no study has been carried out to investigate the effectiveness of aged NPK fertiliser solution in attracting *Aedes* female mosquitoes to lay eggs. This is because the effectiveness of the attractant used in the ovitrap depends on the duration of its storage (Isoe *et al.*, 1995). Study conducted

by Sant'ana (2006) demonstrated that hay infusion (*Panicum maximum*) kept for 15 or 20 days is optimal because a significantly higher number of *Aedes* mosquito eggs were collected compare to control.

This study aimed to determine the most effective ovitrap between Standard Ovitrap (SO), MLTD, Double Sticky Ovitrap (DST) and NPK Fertiliser Trap in monitoring *Aedes* mosquitoes. The results obtained from the study are seen to provide additional information for vector surveillance team to choose the best ovitrap in surveillance and control of dengue vectors. This study is relevant since no comprehensive study comparing these four ovitraps has been conducted in Malaysia and it is the first local study which uses NPK fertiliser as oviposition attractant.

#### MATERIALS AND METHODS

#### Study sites

The selected area for this study was situated west of Kuala Lumpur city centre and in the Titiwangsa Parliamentary area. The selection of the study area was based on several criteria which are a large size area, presence of human settlement and *Aedes* mosquito population. The study area was divided into four main locations for the purpose of sampling which is A, B, C and D. The distance between each sampling points is 100 m. The ecological descriptions of each sampling point's location (A, B, C and D) are given in Table 1.

#### Ovitrap techniques

Standard Ovitrap (SO) consists of a tin container with 7.5 cm internal diameter both at the bottom and the top. This tin container with the height of 10.5 cm is painted in black on the outer wall. Ovistrip is made of hardboard measuring 12 cm X 3.5 cm X 0.3 cm is placed diagonally and each ovitrap was filled with water to a level of 6.5 cm (Service, 1993). Mosquito Larvae Trapping Device (MLTD) is a type of autocidal ovitrap (Azil *et al.*, 2014). This ovitrap consists of a cylinder-shaped plastic container measuring 24 X 13.5 cm with a lid, funnel

Table 1. Ecological descriptions of location of each sampling points

Sampling points	Ecological description
A	<ul> <li>Situated in the hostel block area (Block 6).</li> <li>Spacious and wide area.</li> <li>Located in the area between guard house (main entrance) and Audiology clinic.</li> <li>Big trees and landscape plants planted around the area.</li> <li>The environment is generally clean and well managed.</li> </ul>
В	<ul> <li>Situated next to basketball and tennis court area.</li> <li>Remote area with large drain.</li> <li>Big trees around the area.</li> <li>There is a bush area but the environment is generally clean.</li> </ul>
С	<ul> <li>Situated behind the hostel block area between Block E and Block F.</li> <li>Spacious and wide area with large drain.</li> <li>Big trees and shrubs planted around the area.</li> <li>The environment is generally clean and well managed.</li> </ul>
D	<ul> <li>Situated near to hostel block H and Block J area (near to bus stop).</li> <li>Spacious and open area with large drain.</li> <li>Tall and big trees around the area.</li> <li>The environment is generally clean but with occasional floods after heavy rain.</li> </ul>

and black jacket. About 1.4 L water should be poured into this ovitrap. Dewan Bandaraya Kuala Lumpur (DBKL) is the agency responsible for the introduction of the MLTD (Shaari, 2001).

Double Sticky Ovitrap (DST) is designed and modified from the Sticky Ovitrap. However, it still retains the same principle. DST consists of two dark buckets sitting against each other. The bucket placed on the top has the base removed and placed with gummed paper or plastic on the inner surface. The bucket at the bottom is filled with water (~1.5 L) and punched to allow excess water out. Both the buckets are attached together using "fold-back" clips (Chadee & Ritchie, 2010a). Plant-based pellets were added to attract adult mosquitoes.

### Construction of ovitrap and servicing in the field

One type of ovitrap was specifically designed for this study which is the NPK Fertiliser Trap (Figure 1). The recycled mineral bottle (1.5 L) was sprayed with black paint and dried overnight. Next, the bottle was cut about 10 cm from the top surface (Figure 2a). After being cut, the top surface of the bottle was reversed (Figure 2b) and placed on the top. Next, the middle

part of the trap was punched at the four corners to enable excess water to flow out (Figure 2c). The trap was filled with NPK fertiliser solution (500 ml) before being deployed in the field.

After a week of deployment, each ovitrap was replaced with a new one. Water from each ovitrap was poured into a white tray to collect the mosquito larvae present. In addition, the presence of adult mosquitoes was also examined in the DST and NPK Fertiliser Trap. The larvae and adult mosquitoes which were calculated and recorded were brought to the laboratory for rearing process and species identification.



Figure 1. NPK Fertiliser Trap.



Figure 2. Construction of NPK Fertiliser Trap that was used in sampling larvae and adult mosquitoes.

#### Determination of optimum concentration for NPK fertiliser solution

The fertiliser used in the NPK Fertiliser Trap was the commercial organic fertiliser containing Nitrogen, Phosphorous and Potassium components (ratio 5:5:5). An experiment in the field was first conducted to obtain the optimum concentration of NPK fertiliser solution. A total of five concentration were selected which is 0.05 g/L, 0.1 g/L, 0.25 g/L, 0.5 g/L, and 1.0 g/L. NPK Fertiliser Trap with these five concentrations were placed in different areas for one week. This experiment was conducted seven times and the mean number of *Aedes* larvae were recorded (Table 2).

The optimum concentration of NPK solution selected and used in the NPK Fertiliser Trap was 0.25 g/L for the purpose of this study. A total of 0.25 g NPK fertiliser was weighed and dissolved with 1 L of piped water. The solution was prepared fresh before being used in the field.

### Research design to determine the effectiveness of various ovitraps

The experimental design method of 4X4 Latin Square (Cohran & Cox, 1957) was used to determine the effectiveness of various ovitraps. A total of eight units of ovitrap for each type (SO, MLTD, DST and NPK Fertiliser Trap) were placed in four different location of the sampling point (A, B, C and

D). In each location of the sampling point, the distance between one ovitrap and another was a minimum of 4 m (Williams *et al.*, 2006). These ovitraps were rotated or transferred to other location every week. A complete cycle comprised of four weeks and these experiments were conducted in three replicates. The duration of this study was 3 months which started on 9 November 2015 until 1 February 2016.

## Research design to determine the effectiveness of NPK (fresh) and NPK (stock) solution

The NPK fertiliser (fresh) solution has to be prepared fresh while the NPK fertiliser (stock) solution has to be stored for a month before being used in the field. The experimental design method of 4X4 Latin Square (Cohran & Cox, 1957) was used. Since only two types of ovitrap involved, only two sampling centres were used namely

Table 2. Mean number of *Aedes* larvae collected using different concentrations (g/L) of NPK fertiliser solution

Concentration (g/L)	The mean number of larvae
0.05	44.8
0.1	50.0
0.25	51.57
0.5	43.0
1.0	37.33

A and B. Thus a complete cycle comprised of two weeks (14 March until 28 March 2016) and these experiments were not repeated.

#### Identification of adult mosquitoes

The larvae collected in the field were reared into adults in the insectarium at room temperature from 26°C to 32°C and relative humidity from 65% to 85% before the mosquito species can be identified (Saifur et al., 2012). Beef liver powder was added throughout the breeding period as food for the larvae. The adult mosquitoes collected from the field were immediately identified using EZD/EZ4HD stereomicroscope (Leica Microsystems, Switzerland). Species and sex identification of the adult mosquitoes are based on the taxonomic keys from Jeffery et al. (2012).

#### **Data Analysis**

All data obtained from this study was analysed as follow:

- 1. Ovitrap Index (OI), the percentage of positive ovitrap against the total number of ovitraps deployed at each study site.
- 2. Mean number of larvae/adults per ovitrap.

The significant difference of OI obtained from all trapping methods were conducted using Chi-Square test ( $\chi^2$ ). The one way ANOVA or Kruskal-Wallis test was conducted to analyse the mean number of differences on larvae per ovitrap recovered between four ovitraps. The mean numbers of larvae or adults per ovitrap between two ovitraps were assessed by Student t-test or Mann Whitney test. All level of statistical significance was determined at p  $\leq$  0.05 by using a statistical programme (SPSS v 23.0).

#### RESULT

# Distribution of *Aedes* larvae, Ovitrap Index (%) (OI) and mean number of larvae *Aedes* per ovitrap (Mean ± SEM) obtained from four ovitraps

A total of 29,579 *Aedes* larvae were collected by all four trapping methods. A large number of *Ae. albopictus* larvae were

collected (99.8%) compared to Ae. aegypti larvae (0.2%). All four trapping methods successfully collected Aedes larvae and OI for each technique was more than 95%. The highest OI was obtained from DST (98.9%) followed by NPK Fertiliser Trap (97.7%), MLTD (97.6%) and SO (95.4%). There was no significant difference between OI obtained from all trapping methods,  $\chi^2$  (3, N = 344) = 2.14, p = 0.544. Results of ANOVA showed a significant difference in the mean number of larvae per ovitrap obtained from all four trapping methods, F(3, 44) = 7.76, p < 0.001 (Figure 3). Post-hoc analysis using Tukey HSD showed that the mean number of larvae per ovitrap obtained from DST was significantly higher compared to MLTD and SO but no significant difference compared to NPK Fertiliser Trap. On the other hand, NPK Fertiliser Trap had higher mean number of larvae per ovitrap than MLTD and SO but this was not significantly different.

## Distribution of Aedes adult, Ovitrap Index (%) (OI) and mean number of adult per ovitrap (Mean ± SEM) obtained from two ovitraps

The DST and NPK Fertiliser Trap collected a total of 123 Ae. albopictus adults. Both trapping methods successfully collected Ae. albopictus adults and OI for each techniques ranging from 20% to 50%. OI obtained from NPK Fertiliser Trap was significantly higher,  $\chi^2$  (1, N=174) = 12.68, p < 0.001 compared to DST (Figure 4). NPK

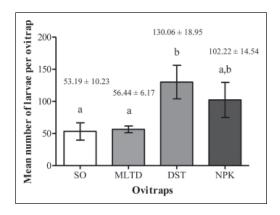


Figure 3. Mean number of Aedes larvae among the four ovitraps. Ovitraps without letters in common are significantly different at p < 0.05.

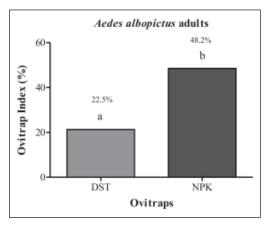


Figure 4. Ovitrap Index (%) between Double Sticky Ovitrap (DST) and NPK Fertiliser Trap (NPK) obtained to monitor Aedes adult mosquitoes in the study area. Ovitraps without letters in common are significantly different at p < 0.001.

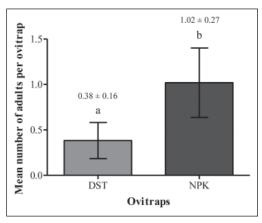


Figure 5. The mean number of adults per ovitrap between Double Sticky Ovitrap (DST) and NPK Fertiliser Trap (NPK). Ovitraps without letters in common are significantly different at p < 0.05.

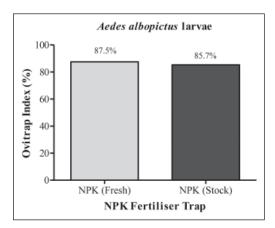


Figure 6. Ovitrap Index (%) between NPK (fresh) and NPK (stock) that were added to the NPK Fertiliser Trap to attract *Aedes* larvae mosquitoes in the study area.

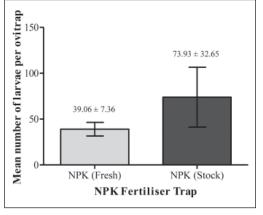


Figure 7. The mean number of larvae (Mean  $\pm$  SEM) obtained from NPK (fresh) and NPK (stock) that were added to the NPK Fertiliser Trap in the study area.

Fertiliser Trap had higher mean number of adults per ovitrap than DST (Figure 5). There was a significant difference between the number of adults per ovitrap obtained from both trapping methods in the study area, U = 28.00, z = -2.55, p < 0.05, two tailed tests.

Ovitrap Index (%) (OI) and mean number of larvae *Aedes* per ovitrap (Mean ± SEM) obtained from NPK (fresh) and NPK (stock)

Both ovitraps that contained NPK (fresh) and NPK (stock) successfully collected *Aedes* larvae mosquitoes and OI for each technique

is more than 85%. OI obtained from NPK Fertiliser Trap that contained NPK (fresh) was higher compared to NPK Fertiliser Trap that contained NPK (stock) solution (Figure 6). There was no significant difference between OI obtained from both solutions (fresh and stock),  $\chi^2$  (1, N=30) = 0.02, p = 1.00. The mean number of larvae collected by NPK (stock) was higher compared to NPK (fresh) (Figure 7). Statistical analysis using Mann-Whitney U test showed there was no significant difference between the number of larvae per ovitrap obtained from both NPK solutions (fresh and stock),

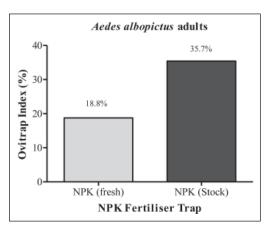


Figure 8. Ovitrap Index (%) between NPK (fresh) and NPK (stock) that were added to the NPK Fertiliser Trap to attract *Aedes* adult mosquitoes in the study area.

U = 103.50, z = -0.35, p = 0.724, two tailed test.

#### Ovitrap Index (%) (OI) and mean number of adult *Aedes* per ovitrap (Mean ± SEM) obtained from NPK (fresh) and NPK (stock)

Both NPK solutions (fresh and stock) successfully collected Ae. albopictus adults and OI for each techniques ranging from 15% to 40%. OI obtained from NPK Fertiliser Trap that contained NPK (stock) was higher compared to NPK Fertiliser Trap that contained NPK (fresh) (Figure 8). There was no significant difference between OI obtained from both solutions (fresh and stock),  $\chi^2$  (1, N = 30) = 1.10, p = 0.417. NPK (stock) collected higher mean number of adults per ovitrap compared to NPK (fresh) (Figure 9). Statistical analysis using Mann-Whitney U test showed there was no significant difference between the number of adults per ovitrap obtained from both NPK solutions (fresh and stock), U = 91.50, z = -1.11, p = 0.269, two tailed test.

#### DISCUSSION

This study was conducted to compare the effectiveness of various ovitrap techniques including Standard Ovitrap (SO), MLTD,

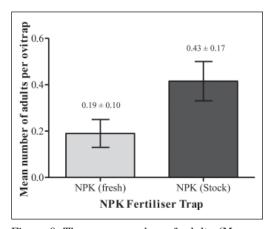


Figure 9. The mean number of adults (Mean  $\pm$  SEM) obtained from NPK (fresh) and NPK (stock) that were added to the NPK Fertiliser Trap in the study area.

Double Sticky Ovitrap (DST) and NPK Fertiliser Trap in monitoring *Aedes* mosquitoes. All ovitraps successfully collected *Aedes* larvae mosquitoes with the Ovitrap Index (OI) more than 95%. The DST recorded the highest OI (98.9%) while SO recorded the lowest OI (95.4%). However no significant difference was found between all ovitraps. Based on the OI, infestation rates of *Aedes* larvae and its population density in the study location were high.

The colours of ovitraps are important in order to attract *Aedes* adult mosquitoes laying their eggs. According to Chua *et al.* (2004), the *Aedes* adult mosquitoes use visual signals to locate suitable areas for oviposition and dark containers (black, blue or red) are preferred. Therefore, the uses of black (SO, MLTD, NPK Fertiliser Trap) and red (DST) ovitraps help in collecting *Aedes* larvae mosquitoes.

Each ovitrap effectively collected Ae. albopictus larvae compared to Ae. aegypti due to the study location which was surrounded by trees and other plants making it a suitable habitat for Ae. albopictus. In contrast, Ae. aegypti commonly breed in man-made containers in houses (Norzahira et al., 2011). The study conducted by Lima-Camara et al. (2006) also suggested that Ae. aegypti adults caught in the city prefer to rest indoors with high density of humans while Ae. albopictus prefers to rest outdoors

nearby vegetation, which explains the lack of Ae. aegypti caught in our traps.

The mean number of Aedes larvae collected by DST was the highest (130.06  $\pm$ 18.95) and significantly different compared to SO (53.19  $\pm$  10.23) and MLTD (56.44  $\pm$ 6.17). Besides colour, the effectiveness of ovitrap is also determined by the size, shape and depth of the water inside the ovitrap (Chua *et al.*, 2004; Reiskind & Zarrabi, 2012). Size and shape of the DST is larger and contains more water enabling it to collect more Aedes larvae compared to SO and MLTD. Panigrahi et al. (2014) concluded that Aedes mosquitoes prefer to lay their eggs in larger containers which are less prone to drying and this will likely increase the survival of the mosquito larvae.

The addition of plant-based pellets to DST as an attractant for adult mosquitoes was also a factor which enabled this ovitrap to collect more *Aedes* larvae. Whilst NPK Fertiliser Trap contains organic solution which stimulates the growth of microorganisms and in turn supplied the food source to the mosquito larvae (Darriet *et al.*, 2012).

Although NPK Fertiliser Trap was able to collect a large number of larvae (102.22  $\pm$  14.54), there was no significant difference compared to the SO and MLTD. This was due to the smaller opening of NPK Fertiliser Trap compared to DST. Some female mosquitoes might not be able to detect the presence of mosquito larvae in that ovitrap due to its small surface opening. According to Wong et al. (2011), the presence of large number of larvae in the breeding area would attract others female mosquito to lay their eggs in that container. However, the Allee effect should be considered here. The Allee effect is indicated when Aedes mosquitoes are more likely to breed in a container containing average numbers of eggs or larvae compared to large numbers or none at all (Williams et al., 2008).

Double Sticky Ovitrap and NPK Fertiliser Trap were also capable to capture female gravid mosquitoes. When the female gravid mosquitoes are captured, some of them will release all their eggs in the ovitrap and this behaviour is called death-stress oviposition. The death-stress oviposition was introduced by Chadee & Ritchie (2010b) who obtained high collection of immatures by DST compared to SO in their study.

Although DST collected the highest mean number of larvae, it has some weaknesses. This ovitrap could become breeding container for Aedes mosquitoes if left unattended for more than seven days (Santos et al., 2003). Thus, monitoring of this ovitrap should be carried out within seven days or less. In contrast, autocidal ovitraps such as MLTD and NPK Fertiliser Trap are invented to kill second generation of adult mosquitoes. Hence, they do not become breeding containers if left for more than seven days. S-methoprene pellets which do not influence the oviposition activities of Aedes mosquitoes, may be used to prevent ovitraps from becoming mosquito breeding grounds (Ritchie & Long, 2003).

The female *Aedes* mosquito practices skip-oviposition behaviour by laying their eggs in several breeding areas (Abreu *et al.*, 2015). Thus, this behaviour could be avoided by using DST and NPK Fertiliser Trap which captured female gravid mosquitoes. As a result, the spread of female mosquito and dengue virus could be prevented. According to Lee *et al.* (2013), female *Aedes* mosquito is known to feed on human blood several times in one gonotrophic cycle. Thus the trapping of gravid female mosquitoes could help in reducing dengue virus transmission.

The decomposing organic matter supplies nutrients for mosquito larvae and is an important chemical signal for female mosquito in searching and selecting breeding areas (Derraik & Slaney, 2004). The chemical released from decomposition of organic matter acts as an attraction and hence stimulates the female gravid mosquito to lay eggs (Ponnusamy *et al.*, 2008). The commercial organic fertiliser solution which contains components of Nitrogen, Phosphorous and Potassium in low ratio selected in this study acts as an attractant for female *Aedes* mosquitoes.

Based on this study, NPK Fertiliser Trap is more capable to capture female *Aedes* mosquito with OI of 48.2% compared to DST with OI of 22.5%. The mean number

of Aedes adult mosquitoes collected by NPK Fertiliser Trap was higher  $(1.02 \pm 0.27)$  compared to DST  $(0.38 \pm 0.16)$ . The OI and mean number of adult mosquitoes for both ovitraps showed significant difference (p < 0.001) and p < 0.05). The finding of this study is supported by Anderson and Davis (2014). They reported that NPK solution was effective to attract Aedes mosquitoes to lay their eggs. In their study, they tested on the NPK solution with the concentration of 20 mg/L of Nitrogen, 11 mg/L of Phosphorus and 28 mg/L of Potassium/Kalium, respectively.

The ammonium compounds ( $\mathrm{NH_4^+}$ ) contained in the NPK solution will be converted into ammonia ( $\mathrm{NH_3}$ ) and hence released into the air in the form of gas (Choudhury & Kennedy, 2005). According to Anderson and Davis (2014), the volatile ammonia compound attracts female *Aedes* mosquitoes to lay eggs in the NPK solution. Study carried out by Marques *et al.* (2013) found that *Aedes* mosquito lays eggs in large number with high ammonium compound in the water. The finding of their study suggested that ammonium compound acted as an oviposition attractant.

In this study, the capability of DST to capture adult Aedes mosquitoes was low as represented by the mean number of adult mosquitoes caught. The external factors such as heavy rainfall could cause damage to the gummed paper of these ovitrap rendering it ineffective to trap adult mosquitoes. This was also supported by Santos et al. (2012) who stated that the effectiveness of the Sticky Ovitrap placed outdoors is largely influenced by environmental factors. As a solution for this problem, transparent plastic strip painted with adhesive was suggested (Ritchie et al., 2003; Williams et al., 2006; Chadee & Ritchie, 2010a).

The NPK Fertiliser Trap invented in this study has several advantages compared to other ovitraps. It is able to collect large numbers of *Aedes* larvae and adult mosquitoes. In addition, this ovitrap uses recycled bottle and can be prepared with a minimal cost. NPK Fertiliser used as an attractant in the ovitrap is easily accessible

and the solution is easy to be prepared. Thus, this ovitrap technique which is of low cost and easy to assemble, should be recommended for the use in the community to foster its utilization in combating Aedes mosquitoes. Its usage as a surveillance tool has been demonstrated in this current study. Nonetheless, improvisation of NPK Fertiliser Trap is needed to explore its potential as an autocidal ovitrap. However, placing NPK Fertiliser Trap outdoors should be carefully done because failure of the ovitrap is likely to occur. NPK Fertiliser Trap may be knocked down as a result of human activities, animals or the environment such as wind. To overcome this failure, detailed explanation should be given to the public regarding its proper use.

This comparative study was conducted to see the effectiveness between fresh and stock NPK Fertiliser solutions added to NPK Fertiliser Trap to monitor Aedes larvae and adult mosquitoes. In monitoring of Aedes larvae stage, the OI for fresh solution was found higher (87.5%) compared to OI for stock solution (85.7%) but mean Aedes larvae was higher for stock solution (73.93  $\pm$  32.65) compared to fresh solution (39.06) ± 7.36). For monitoring of adult Aedes mosquitoes, it was found that both OI and the mean adult Aedes mosquitoes collected by the stock solution was higher (OI: 35.7%; mean adult mosquitoes:  $0.43 \pm 0.17$ ) as compared to fresh solution (OI: 18.8%; mean adult mosquitoes:  $0.19 \pm 0.10$ ).

When statistical tests were conducted on the fresh and stock NPK solutions, no significant differences were found. Thus, the stock solution could be used for commercialisation purposes because it is user friendly and as effective as the fresh solution. However, further studies have to be carried out with prolonged duration of the storage so that the shelf life of the aged NPK solution could be determined.

Also, we compared the four ovitraps in terms of rough estimated costs, maintenance and labour (manpower). Based on the study, it is found that SO and NPK Fertiliser Trap are cheaper than MLTD and DST. The SO and NPK Fertiliser Trap used recycled cans and

mineral bottles. One unit of NPK Fertiliser Trap only requires 0.25g/L of a packet NPK fertiliser (500g) costing only RM10. The DST costs more because it includes several components such as bucket, gummed paper, "fold-back" clips while a unit of MLTD is purchased from DBKL for RM15.

In terms of the maintenance, it is found that the DST and NPK Fertiliser Trap need to be prepared and setup before surveillance is carried out. According to Azil et al. (2014) who conducted interviews with the field workers in Australia noted that Sticky Ovitrap was difficult to handle because of the gummed paper or plastic placed in it. Besides, the procedure in the preparation, maintenance and cleaning of the ovitrap took longer time and higher labour cost. NPK Fertiliser Trap also involves the preparation of NPK fertiliser solution. However, this study proves that NPK fertiliser solution does not need to be fresh and this can save time for ovitrap surveillance. SO and MLTD do not require any preparation beforehand and can be immediately deployed in the field by only filling the ovitrap with water.

The *Aedes* larvae collected by SO and MLTD require rearing in the laboratory before species identification. Thus, the cost for manpower involved is high (Ritchie *et al.*, 2003). On the other hand, the DST and NPK Fertiliser Trap are capable to trap *Aedes* adult mosquitoes. Thus, species identification can be done immediately in the field.

#### CONCLUSION

Based on this study, four ovitraps namely Standard Ovitrap (SO), MLTD, Double Sticky Ovitrap (DST) and NPK Fertiliser Trap were capable to collect *Aedes* larvae mosquitoes in the study location. DST was the most effective due to its capability to collect higher number of *Aedes* larvae mosquitoes compared to other ovitraps. Nonetheless, NPK Fertiliser Trap was more effective compared to DST in collecting and trapping *Aedes* adult mosquitoes in the study location. Comparison shows no significant difference

between the fresh or stock NPK Fertiliser solutions. Therefore, freshly prepared and aged NPK solution can be used for dengue surveillance and vector control purposes. This opens opportunities for further research and product commercialisation.

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