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Screening of Five Plant Extracts for Larvicidal Efficacy against Larvae of *Aedes aegypti* (L.) and *Aedes albopictus* (Skuse)

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The adverse effects of prolonged and rampant usage of chemical insecticides in controlling the population of vector arthropod have caused the development of resistance among vector populations as well as non-target organism. Application of plant extracts could be alternative sources for mosquito control. The present study assessed larvicidal activities of methanol extracts of leaf and stem of *Jacaranda mimosifolia* Don (Family: Bignoniaceae), *Melaleuca cajuputi* Powell (Family: Myrtaceae), *Tabebuia chrysantha* (Jacq.) Nicholson (Family: Bignoniaceae), *Tabebuia pallida* (Lindl.) Miers (Family: Bignoniaceae) and *Tabebuia rosea* Toll (Family: Bignoniaceae) against dengue vectors, *Aedes* (Diptera: Culicidae) sp. Among plants tested, *M. cajuputi* exhibited the most effective with the highest mortality against *Ae. aegypti* and *Ae. albopictus*. Leaf extracts showed significantly higher larvicidal effects in relative to stem extracts. The findings also revealed that *Ae. aegypti* is the most susceptible compared to *Ae. albopictus*. LC50 values of *M. cajuputi* leaf extracts were 183.35mg/L and 191.82mg/L against *Ae. aegypti* and *Ae. albopictus* respectively. These studies suggest leaf extracts of *M. cajuputi* have moderate potential as larvicidal against vector larvae of *Aedes* mosquitoes.

Keywords: *Ae.aegypti*; *Ae. albopictus*; larvicidal activities; plant extracts

I. INTRODUCTION

According to the World Health Organization, WHO (2014), mosquito is the prominent vector of all disease-transmitting arthropod, causing millions of death and hundreds of illnesses cases around the world each year. Whilst many scientists struggling in finding a cure for other existing illnesses caused by a mosquito such as dengue fever, yellow fever, malaria and chikungunya, the emerge of Zika virus recently, which transmitted by *Aedes* mosquito, has become into the limelight to make major news headlines. Although Zika virus symptoms are mild, the virus can have drastic and serious implications by causing infection and a congenital neurological disorder called microcephaly in newborns (WHO, 2016). This current development and scenario have created an urge needs to control mosquito population aggressively. The alarming condition is not only restricted to the affected country but also involving many other countries which have

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been dealing with mosquito-borne diseases problem for almost a decade such as Malaysia. In Malaysia, mosquito-borne diseases is a continuously becoming a serious threat and nationwide problem. Dengue fever is common among Malaysians since its early discovery in Penang, Malaysia by Skae (1902), and became a public health problem in the 1970s (Wallace *et al.*, 1980). To date, dengue fever is one of the major health concerns in Malaysia and perceived as a highly contagious health threat with escalating trend of infection (Mia *et al.*, 2013). Therefore, combating mosquito population are essential as one of a proactive approach in controlling dengue transmissions and many other mosquito-borne diseases alike.

For many years, the main strategy in controlling arthropod-borne diseases is mainly dependent on the use of synthetic insecticides (Somboon *et al.*, 2003 and Jirakanjanakit, 2007). However, prolonged use of chemical insecticide has led to resistance among vectors, particularly from endemic countries (Hemingway and Ranson, 2000). It also causes environmental pollution on living organisms and development of insecticide resistance among non-target organisms. In Malaysia, the development of resistance could be due to the heavy use of permethrin formulation against *Ae. aegypti* in early 1996 during dengue control operations (Nazni *et al.*, 1998). For centuries, annoying pests are easily being removed or controlled through the use of cheap chemicals such as organochlorine, organophosphorus and Methyl Carbamate. However, current situations have changed progressively where it is becoming increasingly difficult and requires a high cost. The challenges and problem arise due to the development of resistance among vector, pollution to the environment and long-term toxic effects on animals and humans. This situation has urged the need for alternative insecticides particularly from plant sources (Sukumar *et al.*, 1991; Mulla and Su, 1999 and Shaalan *et al.*, 2005).

Application of plant extracts/natural products as an alternative pesticide control has been known since ancient times (Newman and Cragg, 2010 and El-Wakeil, 2013). In a natural habitat, plants have their own defence mechanisms against pest/insect infestations and pathogens. The mechanisms contain active phytochemicals in their secondary metabolites which are produced to protect the plant from insect herbivores and plant pathogens. The mechanisms involve deterrent or antifeedant activity (Bennet and Wallsgrove, 1944; Luthria *et al.*, 1993; Sukumar *et al.*, 1991; Casida and Quistad, 1998 and Isman, 2000). Jacobson (1982) has classified the active chemical components mechanisms into few types: repellent agent, killer agent, sterility agent, growth regulators agent and as a barrier-eaten (antifeedant). Chemicals released will cause death or act as a repellent agent. Many studies have reported the effectiveness of plant chemical derivatives could potentially play a significant role in contributing to the mosquito control programme (Sukumar *et al.*, 1991; Choochote *et al.*, 2004 and Amer and Mehlhorn, 2006). Hence, natural sources are environmentally safe and biodegradable (Sharma *et al.*, 2006). Ongoing investigation is continuously being carried out by many researchers in searching a natural plant-based insecticide that acts specifically effective and safe for the environment. Apart from finding potential new plants, investigations also include filter-

ing, sorting and development of phytochemicals that have the pesticide active ingredient (Mulla and Su, 1999).

Chemical produced by plants such as rotenone, nicotine, and pyrethrins have long been used in vector-borne disease control activities as well as insect pests. Earliest discovery of a number of beneficial substances in plant extracts was credited to Campbell (1933) for the findings. It recorded that extract of Russia weeds, *Anabasis aphylla* Leonard has a chemical such as nicotine alkaloids, anabasine, metal-anabasine and Lupine can cause the death of *Culex pipiens* L., *Culex territans* Walker and *Culex quinquefasciatus* Say. Despite the proactive study by many researchers in finding bioactive chemical compounds in the potential plant as natural resources, selection of the insect is also essential before a screening test being carried out. There are nearly three million species of insects in the world. More than one million species of insects are crop-eating, and of these, about 700 species worldwide cause damage to man's crops and causing a profound impact on the human health's, livestock animals and pets. Therefore, the choice is based on strong reasons such as the importance of insects to the fields of medicine or agricultural economics.

This study was conducted to screen the potential of five (5) plant extracts as a bioinsecticide source against *Aedes* mosquito larvae. The plant species were *Jacaranda mimosifolia* Don (Family: Bignoniaceae), *Melaleuca cajuputi* Powell (Family: Myrtaceae), *Tabebuia chrysantha* (Jacq.) Nicholson (Family: Bignoniaceae), *Tabebuia pallida* (Lindl.) Miers (Family: Bignoniaceae) and *Tabebuia rosea* Toll (Family: Bignoniaceae). The Jacaranda tree is a subtropical native plant in South America and can live almost everywhere in the world except for extreme weather (Rodd and Stackhouse, 2008). In Malaysia, it is known as 'janda merana' among the locals. Rojas *et al.* (2006) reported its use in medicine as an antimicrobial against certain bacteria such as *Pseudomonas aeruginosa*, *Bacillus cereus*, *Escherichia coli* and *Staphylococcus aureus*. The leaf parts were reported to contain Anthocyanins, Alkaloids, and Phenolic compounds. The *M. cajuputi* or 'tea tree oil' can easily be found in Malaysia especially in mangrove swamps near the coastal area. Among locals, this tea tree oil is known as 'gelam' tree and has various uses as natural medicinal resources. It is known to treat colic, cholera, removing the mucus, treating bronchitis, parasitic worms, relieving toothache (Ruangrungsi and Tontiwat, 1991 and Wee, 1992) and used as a mosquito repellent (Nuyim and Buntawee, 1999).

There are about 100 species of neotropical plants from the genus of Tabebuia (Steyermark et al., 1997). Tabebuia is widely used as a landscaping plant on the roadside because of its colourful and attractive flower. Various species of Bignoniaceae family are also used in traditional treatments for treating diseases associated with fungal infections (Gentry, 1992). Its roots have strong diuretic properties such as antisyphilitic and vermiculite activity (Burkill, 1985). The concentrated extraction of the flower parts and the bark are used in treating abdominal pain and treating diabetic diseases (Burkill, 1985). This study aimed to evaluate the efficacy of methanol extracts of *Jacaranda mimosifolia* Don (Family: Bignoniaceae), *Melaleuca cajuputi* Powell (Family: Myrtaceae), *Tabebuia chrysantha* (Jacq.) Nicholson (Family: Bignoni-

aceae), *Tabebuia pallida* (Lindl.) Miers (Family: Bignoniaceae) and *Tabebuia rosea* Toll (Family: Bignoniaceae) against larvae of dengue vectors, *Aedes* (Diptera: Culicidae) *Aedes aegypti*, and *Aedes albopictus*.

II. MATERIALS AND METHOD

A. Collection and Preparation of Specimen

Plant specimens were obtained from the Botanical Garden Park at the Forest Research Institute of Malaysia (FRIM) ($3^o14'01.64"$N; $101^o37'06.09"$E). Collection of plant specimens were assisted and identified by the staff from the Medicinal Plants Program Division of FRIM. Parts of the plants used were leaves and stems. Collected plant specimens were cleaned and dried at room temperature (34.0^{o}C - 37.5^{o}C) for seven days with relatives humidity (RH%) between 65-85%. Leaves and stem of the specimens were separated, weighed and labelled accordingly before the methanol extraction process. All extraction process was carried out at the Medicinal Plants Program Division laboratory.

B. Methanol Extraction

Methanol extraction was carried out with a cooling immersion method with slight modification (FRIM, 2000). Specimens were transferred into a plastic bottle container (10L), immersed with methanol (1g of a specimen to 10ml of methanol) and shake thoroughly. The mixture was then allowed to stand at room temperature (34.0-37.5^{o}C) for a period of at least 72 hours (three days) with frequent agitation until the soluble matter has dissolved. After 72 hours, the mixture was filtered using Buchner funnel with a filter paper Whatman number 1. The filtrate liquid was then concentrated under reduced pressure using rotavapor EYELA (N-1001S-WD, Japan) at 45^{o}C for eight (8) hours to dry and remove the excessive methanol solvents in the filtrate liquid. The raw and solidified material was transformed into the final products and known as crude methanol extracts. The crude extracts were then transferred into a screw-cap vial, labelled and weighed according to their plant species/part of the plant and stored at 4^{o}C for mosquito bioassay testing.

C. Preparation of Mosquito Larva

In this present study, a laboratory strain of *Ae. aegypti* and *Ae. albopictus* larvae were used. The colony of mosquito larval were continuously reared and properly maintained at 27.0±2.0 ^{o}C and 75-85% relative humidity (RH%) under 12:12 h light and dark cycles throughout the study period in our school insectary. A batch of egg strips was purchased from Vector Control Research Unit, School of Biological Sciences, Universiti Sains Malaysia, Pulau Pinang, Malaysia to maintain the colony. The egg strips were immersed in dechlorinated tap water to initiate larval hatching. Larvae were fed with finely powdered diet food following WHO (2005) guidelines with a slight modification. The larvae population were cultured until late 3^{rd} instar and/or 4^{th} instar for bioassay testing.

D. Preparation of Bioassay Extract Solution

Stock solutions were formulated to a desired target concentration based on the formulation:

1g of extract dissolved in 10ml of methanol, leaving the final solution of 100,000 mg/L (ppm) in 10ml solution. To stabilise the stock solution, 3ml of Tween20 (Sigma) were added (as an emulsifier) to the formulation. The extract of stock solutions was labelled and stored at 4-5 ^{o}C. The serial dilutions of six concentrations 10, 50, 100, 200, 300 and 500 mg/L were prepared each in 250ml from the stock solution using formula M_1V_1=M_2V_2. The proportion of larvae density for bioassay testing is one larva to 25ml of water/solution.

E. Larvicidal Bioassay

Bioassay testing for mosquito larvae was followed WHO (2005) standard guideline. Batches of 25 mosquito larvae were transferred into a paper/plastic cup (300ml) containing serial concentrations that were prepared accordingly. After 24 hours exposure, larval mortality was recorded. An observation was carried out by tapping the cup slowly in the mouth or in the middle. Dead larvae are those that do not respond when they are probed with a needle in the siphon or the cervical region. Moribund larvae are those incapable of rising to the surface or not showing the characteristic diving reaction when the water is disturbed. Moribund larvae are counted and added to dead larvae for calculating mortality rate. The bioassay was carried out at room temperature of 26.0±2.0 ^{o}C and relative humidity (RH%) between 65-85%. Larvae that are survived during the 24-hour observation of the bioassay were transferred into another cup for disposal. Four replicates were set up for each concentration and an equal number of controls were prepared simultaneously, to which 1ml methanol was added to 249ml of distilled water.

F. Data Analysis and Acceptance Criteria

Bioassay testing is valid if mortality in control population is less than 20%. Mortality between 5% and 20% should be corrected according to Abbott's formula:

$$Mortality\ (\%) = \frac{\%\ tested\ mortality - \%\ control\ mortality}{100 - \%\ control\ mortality} \times 100$$

All data obtained were analysed using SPSS 17.0 statistical software program with a confidence interval of 95%. Lethal Concentration (LC) values were calculated using probit analysis formula (Raymond, 1985). The LC50 and LC90 values are concentrations that caused 50% and 90% death of tested populations respectively. Student's t-test was used to analyse the significant difference in mortality rate between two Aedes sp population in response to different plant extracts dose concentrations.

III. RESULTS

The extraction yields of each plant are presented in Table 1. The net weight of plant extracts after the concentrated process (removing the excessive methanol) were between 4.3-5.5g for leaves and 3.6-5.8g for the stems respectively. In general, leaves' part from all plant samples produced higher yields than stems.

Table 2 shows the mortality rate of each plant extracts from leaf and stem against *Aedes* larvae.

Table 1. Yields of plant extract

Plants	Family	Parts used	Dried sample (g)	Extract yields (g)
Jacaranda mimosifolia	Bignoniaceae	leaves	210.0	5.2
		stem	275.8	5.8
Melaleuca cajuputi	Myrtaceae	leaves	225.1	5.5
		stem	285.6	4.3
Tabebuia chrysantha	Bignoniaceae	leaves	205.4	5.0
		stem	281.3	5.1
Tabebuia pallida	Bignoniaceae	leaves	214.1	4.3
		stem	265.9	5.1
Tabebuia rosea	Bignoniaceae	leaves	217.7	4.9
		stem	277.7	3.6

Among plant extracts tested, *M. cajuputi* gave highest toxicity effects against *Ae. aegypti* and *Ae. albopictus* populations. The tested population of *Ae. aegypti*, however, is more susceptible to all plant extracts. Whilst population of *Ae. albopictus* were least affected when exposed to other plants extract except for *M. cajuputi*. In general, it was observed that leaves extract was the most effective as larvicidal compared to stems extract in any case. Leaves extract of *J. mimosifolia*, *M. cajuputi*, *T. chrysantha* and *T. rosea* were showed to possessed larvicidal effects at the concentration as low as 10mg/L with the mortality rate ranging between 1.00±0.50% to 75.00±0.50% respectively. On the other hand, leaf extracts of *T. pallida* started to show its minimal larvicidal effect at the higher concentration of 100mg/L with the mortality rate between 1.00±0.50% to 49.00±0.50%. Larvicidal effect of stem extracts was obviously least effective against *Aedes* species with the mortality rate from 1.00±0.50% to 20.00±0.00%.

The summary of the association between mortality rate of Aedes larvae, plant species, parts used and extract dosage were presented in Figure 1 (a) - (b) and Figure 2 (a) - (b) respectively. In general, the figure shows, dose concentration and parts of plant extracts attributes to the mortality rate of Aedes larvae. It was clearly noted that higher dose indicates higher mortality in Aedes larvae population. In addition to that, extracts from leaves were shown to enhance the number of mortality among tested Aedes population as mentioned earlier in Table 2.

Table 3 shows Lethal Concentration (LC) values of leaf and stem extract of five (5) plant species for its larvicidal activity against *Ae. aegypti* and *Ae. albopictus*. The LC values were analysed and calculated using probit analysis of SPSS computer software. The regression equations indicated that the mortality rate was positively correlated with the plant extracts concentration. LC_{50} values of *J. mi-*

Table 2. Screening of methanol extracts of five plants species against *Ae. aegypti* and *Ae. albopictus*

Plants	Dose (mg/L)	[1]*Ae. aegypti* (n=100) Mortality percentage (%) ± SD		[2]*Ae. albopictus* (n=100) Mortality percentage (%) ± SD	
		*Leaf	#Stem	*Leaf	#Stem
[a]*J. mimosifolia*	10	1.00 ± 0.50	0	0	0
	50	1.00 ± 0.50	1.00 ± 0.50	0	0
	100	3.00 ± 0.50	7.00 ± 0.50	0	0
	200	11.00 ± 0.50	11.00 ± 0.96	0	0
	300	19.00 ± 2.06	12.00 ± 0.00	0	0
	500	28.00 ± 0.00	20.00 ± 0.00	0	0
	Control	0	0	0	0
[b]*M. cajuputi*	10	15.00 ± 0.96	1.00 ± 0.50	12.00 ± 0.00	1.00 ± 0.50
	50	25.00 ± 1.50	3.00 ± 0.50	20.00 ± 0.00	5.00 ± 0.50
	100	33.00 ± 0.50	10.00 ± 1.00	32.00 ± 0.00	8.00 ± 0.00
	200	45.00 ± 1.89	15.00 ± 0.96	47.00 ± 1.26	8.00 ± 0.00
	300	61.00 ± 0.50	16.00 ± 0.00	65.00 ± 0.50	11.00 ± 0.50
	500	75.00 ± 0.50	18.00 ± 0.58	71.00 ± 0.96	13.00 ± 0.96
	Control	0	0	0	0
[c]*T. chrysantha*	10	1.00 ± 0.50	1.00 ± 0.50	0	0
	50	1.00 ± 0.50	1.00 ± 0.50	0	0
	100	11.00 ± 0.96	5.00 ± 0.50	0	0
	200	16.00 ± 0.00	9.00 ± 0.50	0	0
	300	16.00 ± 0.00	9.00 ± 0.96	3.00 ± 0.50	1.00 ± 0.50
	500	19.00 ± 1.26	11.00 ± 0.50	5.00 ± 0.50	5.00 ± 0.50
	Control	0	0	0	0
[d]*T. pallida*	10	0	1.00 ± 0.50	0	0
	50	0	1.00 ± 0.50	0	0
	100	1.00 ± 0.50	1.00 ± 0.50	0	0
	200	13.00 ± 0.50	2.00 ± 0.58	5.00 ± 0.50	3.00 ± 0.50
	300	38.00 ± 1.00	5.00 ± 0.50	20.00 ± 0.00	5.00 ± 0.50
	500	49.00 ± 0.50	8.00 ± 0.00	34.00 ± 0.50	5.00 ± 0.50
	Control	0	0	0	0
[e]*T. rosea*	10	2.00 ± 0.58	0	0	0
	50	5.00 ± 0.50	0	0	0
	100	8.00 ± 0.00	1.00 ± 0.50	0	0
	200	8.00 ± 0.00	1.00 ± 0.50	1.00 ± 0.50	0
	300	11.00 ± 0.50	3.00 ± 0.50	3.00 ± 0.50	1.00 ± 0.58
	500	16.00 ± 0.00	4.00 ± 0.00	5.00 ± 0.50	2.00 ± 0.58
	Control	0	0	0	0

*[1 2 c] *There were significant difference of mortality in leaves extract p < 0.05 ;p = 0.019*
*[# 1 2 c] *There were significant difference of mortality in stems extract p < 0.05; p=0.016*
[# 1 b] *There were significant difference of Ae. aegypti percentage mortality in M. cajuputi p < 0.05; p=0.005*
[# 2 b] *There were significant difference of Ae. albopictus percentage mortality in M. cajuputi p < 0.05; p=0.009*

mosifolia leaf and stem extracts from *Ae. aegypti* were 1321.65mg/L and 2424.42mg/L respectively. Whereas, the LC_{90} were noted more than 5,000 and 20,000 mg/L respectively. There were no LC values calculated for *Ae. albopictus* as there was no mortality recorded during the 24-hour observation period. However, leaf extracts of *M. cajuputi* gave LC_{50} below 200mg/L against *Ae. aegypti* and *Ae. albopictus* with the values were 183.35mg/L and 191.82mg/L respectively. On the other hand, the LC_{90} was more than 1,000mg/L for both *Aedes* species. Whilst the

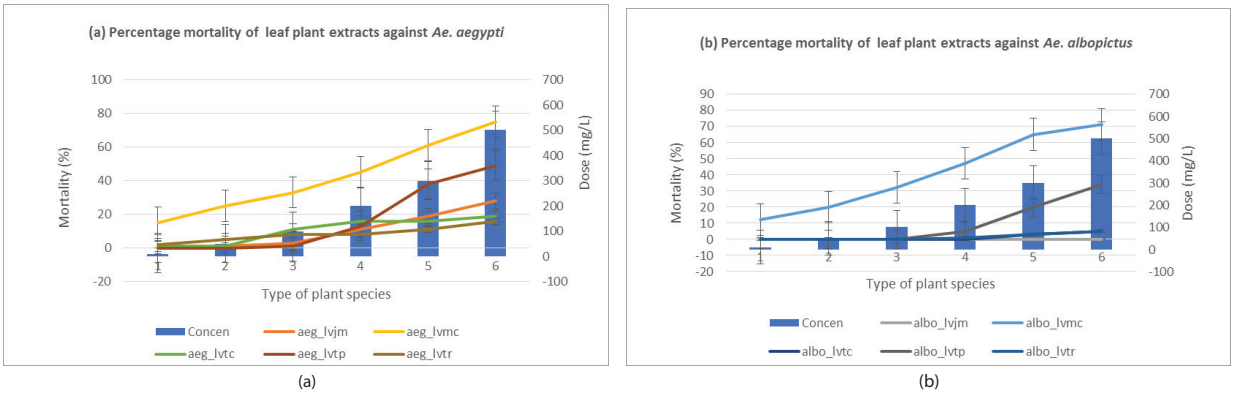

Figure 1. (a) and (b) Larvicidal effect of leaf plant extracts against *Aedes* larvae

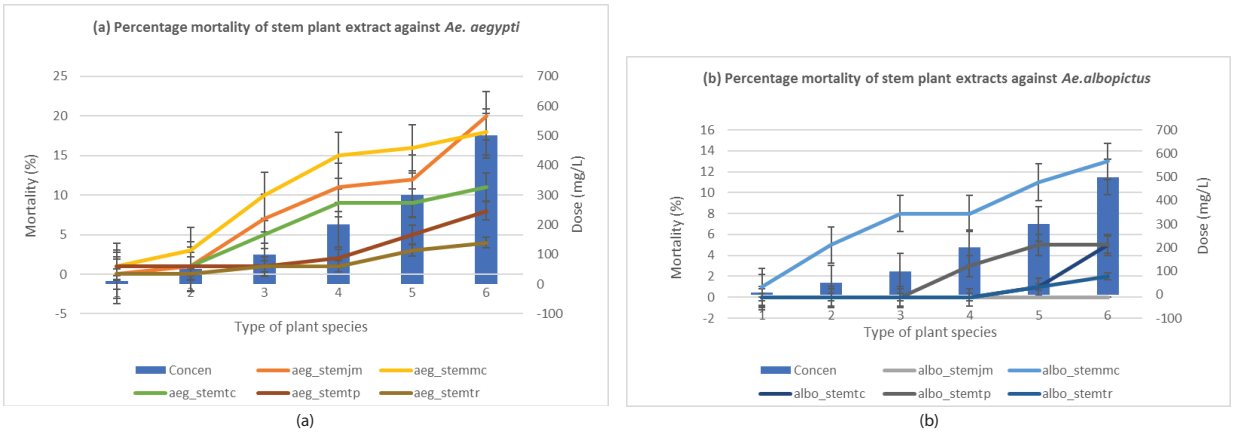

Figure 2. (a) and (b) Larvicidal effect of stem plant extracts against *Aedes* larvae

LC_{50} values of stem extracts were 4568.29mg/L for *Ae. aegypti* and exceeds 30,000mg/L for *Ae. albopictus*. The LC_{90} values were above 10,000mg/L and 30,000mg/L for *Ae. aegypti* and *Ae. albopictus* respectively.

LC_{50} of leaf extracts of *T. chrysantha* against *Ae. aegypti* and *Ae. albopictus* were 3711.26mg/L and 2513.95mg/L respectively. Its LC_{90} values were more than 5000mg/L against *Ae. aegypti* and more than 50,000mg/L for *Ae. albopictus*. LC_{50} and LC_{90} values of stem extracts against *Ae. aegypti* were extremely high with a value of 17882.34mg/L and above 1,000,000mg/L respectively. Whereas, *Ae. albopictus* exhibits higher LC_{50} and LC_{90} values of 3284.82mg/L and 12628.77mg/L respectively. Similarly, leaf extracts of *T. pallida* gave a very weak LC_{50} value of 452.07 mg/L against *Ae. aegypti* and 683.23mg/L against *Ae. albopictus*. On the other hand, the values of LC_{90} for each Aedes species exceed 1,000mg/L. For the stem extracts, the LC_{50} values obtained were 68570.21mg/L and 5377.16mg/L against *Ae. aegypti* and *Ae. albopictus* respectively. Whilst the LC_{90} values of stem extracts exhibits very weakly or inactive against *Ae. aegypti* and *Ae. albopictus*. The values were more than 1,000,000mg/L for *Ae. aegypti* and

41444.95mg/L for *Ae. albopictus*. Leaf extracts of *T. rosea* also exhibit a higher LC_{50} value of 30219.74mg/L and 8868.25mg/L against *Ae. aegypti* and *Ae. albopictus* respectively. Whereas, the LC values of stem extracts were only obtained against *Ae. aegypti* alone because of the insufficient data against *Ae. albopictus*. The LC_{50} and LC_{90} recorded for *Ae. aegypti* were 14455.14mg/L and more than 100,000 mg/L respectively.

IV. DISCUSSION

For almost five decades, the main strategy in controlling vector-borne diseases is by using chemical insecticide (Jirakanjanakit, 2007). Despite its successful approach (Marcombe *et al.*, 2012) the extensive use of synthetic chemical insecticides has resulted in environmental pollution, hazards, and resistance in major vector species (Ranson *et al.*, 2010). In addition to that, the development of resistance is not only limits to the vector species but also towards the non-target organism group. Overcoming this challenge, many researchers nowadays have moving forward focusing on the use of plant extracts in controlling mosquitoes at the larval stage. As broadly defined many plants have defence mechanisms such as mechanical defence and phytochemical defence. Chemical defence includes the ability to produce various chemicals properties naturally, many of which have medicinal, antifeedant, insecticidal, repellent, and growth regulatory properties (War *et al.*, 2012). By manipulating this natural mechanism researchers from any part of the world have become encouraged to explore in the searching of new plant species that can be necessitated the development of a more potent and environmentally friendly insecticide.

The present study aimed to investigate the bioactivity of five (5) species of plants viz *J. mimosifolia*, *T. chrysantha*, *T. pallida*, *T. rosea* from the Bignonaciae family and one (1) from Myrtaceae family, *M. cajuputi* for its larvicidal properties against *Ae. aegypti* and *Ae. albopictus*. In Malaysia, *Ae. aegypti* is the primary vector of dengue fever and chikungunya following the *Ae. albopictus* as a secondary vector. In general, bioassay of the larvicidal effects are said to be good and have potential when the extract causing a high percentage of mortality with a low LC value. The average effective range of larvicidal toxic effects is classified as very weak (inactive), moderate and very good. The toxic effect of $LC_{50} > 200\mu$g/ml is classified as extremely weak (inactive), LC_{50} 20-2003bcg/ml as moderate and $LC_{50} < 203$bcg/ml as very good (Meyer *et al.*, 1982 and Santos *et al.*, 2003). From the results obtained, leaf extracts of *J. mimosifolia*, *T. chrysantha*, *T. pallida* and *T. rosea* exhibited very weak toxic effects with LC_{50} and LC_{90} values were between 400mg/L and above 1,000mg/L against larvae of *Ae. aegypti* and *Ae. albopictus*. *M. cajuputi* extracts can be categorised as moderate larvicidal effects with the LC_{50} value of 180-200mg/L. The effectiveness of a plant extracts as larvicidal is shown to be influenced on several factors including solvents, part of the plant used and exposure period (Hidayatufathi *et al.*, 2003; Komalamisra *et al.*, 2005 and Bagavan *et al.*, 2009). Similar studies were conducted by Hidayatulfathi *et al.* (2003) in Malaysia using methanol extracts of leaves, roots, and flowers of some local plant species. The study found that a mixture of methanol

Table 3. LC_{50} and LC_{90} values of plant extracts against *Ae. aegypti* and *Ae. albopictus*

Plants	Extracts	*Aedes aegypti*			*Aedes albopictus*		
		LC_{50} (CI_{95}) (mg/L)	LC_{90} (CI_{95}) (mg/L)	Regression ± S.E	LC_{50} (CI_{95}) (mg/L)	LC_{90} (CI_{95}) (mg/L)	Regression ± S.E
Jacaranda mimosifolia	Leaf	1321.65 (587.50 - 81652.86)	> 5,000	1.46 ± 0.24	-	-	-
	Stem	2424.42 (1205.15 - 11146.75)	> 20,000	1.23 ± 0.24	-	-	-
Melaleuca cajuputi	Leaf	183.35 (106.17–386.34)	3395.43 (1093.75-63363.66)	1.01 ± 0.11	191.82 (120.95-350.35)	2643.03 (1026.66 –22105.35)	1.13 ± 0.11
	Stem	4568.29 (1728.14 – 41522.56)	> 10,000	0.85 ± 0.17	> 30,000	32377.69 (4679.68- >1,000,000)	0.61 ± 0.17
Tabebuia chrysantha	Leaf	3711.26 (1544.64-25581.71)	9229.45 (2173.93 - >40,000)	0.93 ± 0.19	2513.95 (1043.79 ->1,000,000)	87434.47 (15404.97 ->1,000,000)	2.27 ± 0.93
	Stem	17882.34 (3503.65 - >1,000,000)	>1,000,000	0.76 ± 0.21	3284.82 (1097.67->1,000,000)	12628.77 (2186.43 - >1,000,000)	2.19 ± 1.11
Tabebuia pallida	Leaf	452.07 (396.66 – 538.93)	1218.70 (921.98 – 1871.90)	2.98 ± 0.34	683.23 (553.51 – 974.61)	1992.18 (1291.30 – 4372.99)	2.76 ± 0.42
	Stem	68570.21 (5821.57 - >1,000,000)	>1,000,000	0.72 ± 0.26	5377.16 (1584.60 - >1,000,000)	41444.95 (5361.15 - >1,000,000)	1.45 ± 0.51
Tabebuia rosea	Leaf	30219.74 (4600.92 - >1000000)	>1,000,000	0.59 ± 0.16	8868.25 (1096.94 - >1,000,000)	84186.87 (6835.23 - >1,000,000)	1.31 ± 0.53
	Stem	14455.14 (2262.89 - >1,000,000)	>100,000	1.17 ± 0.51	-	-	-

(-) Not available

extract of *Litsea elliptica* Blume was revealed to be most effective against *Aedes* larvae with LC_{50} values for *Ae. aegypti* were 17.43bcg/ml and 31.113bcg/ml against *Ae. albopictus*.

A study by Bagavan *et al.* (2009) has proved that plant extracts with different solvents could produce various significant results. The ethyl acetate extract of *Rhinocanthus nasutus* Kurz gave a low value of LC_{50} against *Cx. tritaeniorhynchus* of 39.32ppm. Other solvents such as chloroform and methanol gave LC_{50} of 40.46ppm against *Anopheles subpictus* Grassi and 73.27ppm against *Aphis gossypii* Glover respectively. In Thailand, Komalamisra *et al.* (2005) have studied the larvicidal activity of 96 extracts of plant species against *Ae. aegypti* larvae within the first 24 hours and 48 hours. From the findings, it was found that there were very little or almost no mortality was recorded within the first 24 hours. However, mortality was noted when the exposure had completed within 48 hours. Among the plant tested, 44 were classified as effective with the LC_{50} values were <750mg/L). There were six (6) species of plants viz *Rhinacanthus nasutus* (L.) KURZ, *Derris elliptica* Benth, *Homalomena aromatic* Schott, *Trigonostemon reidioides* Kurz, *Stemona tuberose* Lour and *Acorus calamus* (L.) gave LC_{50} values of 16-48mg/L within 48 hours. The other seven (7) plant extracts show moderate larvicidal activity with LC_{50} ranging from 50 to 100mg/L. Whilst 31 other plant extracts gave LC_{50} ranges from 100 to 800mg/L and 52 others showed no larvicidal effects with the LC_{50} value of 1,600mg/L. The results of this study suggested that prolonged exposure duration up to 48 hours, it is most likely the LC value obtained could be lower.

From the results obtained, leaf extracts of *M. cajuputi* gave mortality more than 50% at

the concentration of 500mg/L against *Ae. aegypti* (75%) and *Ae. albopictus* (71%). According to Broussalis *et al.* (1999) results that produced mortality more than 50% is said having insecticidal potential and can be considered for further investigations. Another study conducted by Vaqar *et al.* (2003) have shown that triterpene isolated from the bark of *M. cajuputi* have larvicidal activity against *Ae. albopictus*. The mortality rate was 12.5% at the concentration of 50mg/L. Comparatively, at the same concentration results obtained in our present studies gave higher mortality of 20% against *Ae. albopictus*. At lower concentrations of 100mg/L, the percentage of mortality given by *Ae. aegypti* and *Ae. albopictus* were 33% and 32% respectively exhibiting better larvicidal effects than active compound, triterpene. In brief, it was cleared that extract from the leaf of *M. cajuputi* plant were the most effective as larvicidal against *Ae. aegypti* and *Ae. albopictus*. Other four plant extracts showed extremely weak (inactive) against *Aedes* larvae.

V. CONCLUSION

It was found that among the plants tested, *M. cajuputi* showed to have moderate larvicidal activity and thus has a potential to be explored further. Phytochemicals in plants have various potential insecticides such as larvicidal, adulticidal and also repellents towards vector arthropods such as mosquitoes. Further studies should be continued to evaluate the effectiveness of *M. cajuputi* extracts on dengue vectors in the laboratory as well as in the field. Isolation and identification of the active chemical compounds in the extracts could be done to investigate its potential role in mosquito control agents. It can be utilised in the development of natural insecticides that is environmentally friendly, biodegradable and less toxic to another organism in the ecosystem.

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