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# Research Article

# The Effect of Breeding Habitat Characteristics on the Larval Abundance of *Aedes* Vector Mosquitoes (Diptera: Culicidae) in Three Localities, Galle District, Sri Lanka

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Dengue has become a national burden in Sri Lanka, and the understanding of breeding ecology of vectors, *Aedes aegypti* Linnaeus and *Aedes albopictus* Skuse, is the most effective way to control the disease. The present study was undertaken to investigate the relative larval abundance of *Ae. aegypti* and *Ae. albopictus* in different types and characteristics of containers in three selected localities in Galle district, Sri Lanka. Totally, 550 containers were positive for both *Ae. aegypti* and *Ae. albopictus* larvae. *Aedes albopictus* showed the high larval abundance in all studied sites. The larval abundance of artificial containers (90.57%) was high than that of natural containers (9.43%) for both *Aedes* spp. (P < 0.05). The breeding preference for *A. aegypti* was high in tires (rubber) (17.82%), while plastic cups (28.00%) were the most preferable container type for *Ae. albopictus*. Dark color containers than light color containers (P < 0.05) and containers with leaf litter accumulated as detritus (P < 0.05) showed high relative larval abundance for both *Aedes* species. Containers with 50–100 ml volume of water showed the highest relative abundance of both *Ae. aegypti* (29.28%) and *Ae. albopictus* (41.79%) (P > 0.05). The high larval abundance of *Aedes* recorded in ground level containers (1–5 cm) and their abundance decreased significantly with the increasing of height where containers were found (1–20 cm) (P < 0.05). The significantly high relative abundance was observed with the increase of the shady level for *Ae. aegypti* (57.34%) and *Ae. albopictus* (61.32%) (P < 0.05). This knowledge will be helpfull to implement dengue surveillance programs in the area.

### 1. Introduction

Dengue is one of the most prominent mosquito-borne diseases with a greatest public health importance in the Southeast Asian region. Sri Lanka also has a long history of mosquito vector-borne diseases from time to time, and recently, dengue is considered as the major public health hazard in the country. Since the first dengue outbreak was reported from Sri Lanka in 1965, a dramatic increase of the number of dengue cases has been created being a nationwide burden in the country. Galle district is one of high-risk areas in Southern Province of Sri Lanka, which is continuously affected by dengue in past several years [1, 2].

Aedes aegypti Linnaeus and Aedes albopictus Skuse are major vectors of dengue fever (DF) and dengue hemorrhagic fever (DHF). Both species are container-inhabiting mosquitoes [3–6], and Ae. aegypti is the main vector of DF, while Ae. albopictus is the major secondary vector [7]. Both Ae. aegypti and Ae. albopictus are highly anthropophilic daytime biters, and they are dominant in domestic and peridomestic environments [4]. Aedes aegypti is found mainly in urban areas [8], and Ae. albopictus is found mainly in semiurban and rural areas [5, 7, 9]. However, the larvae of both species are found together in the same habitat [10].

The problem with rapid accumulation of discarded containers everywhere increases the difficulties to effective and efficacious *Aedes* vector control activities. Knowing the most productive container habitats and preferable container characteristics for breeding of *Aedes* vectors is most important for the implementation of successful vector control

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strategies. As the same, it will support to reduce unnecessary usage of larvicides and adulticides.

The previous studies have been reported the breeding habitats and seasonal preferences of *Aedes* vector mosquitoes in other countries [3–6] and in Sri Lanka [7]. However, a little is known on the preferable container characteristics for breeding of *Aedes* vectors in Sri Lanka.

Hence, the current study was started with the intention to investigate the most preferable container characteristics for the breeding of *Aedes* vector mosquitoes by carrying out larval surveys in three selected sites in Galle district, Sri Lanka.

#### 2. Materials and Methods

- 2.1. Sites Selection. The study was carried out in randomly selected three sites categorized as urban, semiurban, and rural in Galle District, Sri Lanka. Karapitiya GN (Grama Niladhari) area (N 6°4′ 14.0952″, E 80°13′ 30.576″), one of the urbanized areas in Galle district, was selected as the urban site, Madampagama GN area (N 6°2′ 16.8″, E 80°06′ 71″) was selected as the semiurban site, and Lankagama GN area (N 6°20′ 59.99″, E 80°27′ 59.99″) considered as the rural site (Figure 1).
- 2.2. Sampling Design. Each site was divided into blocks as fifty homes for each block. A single block was selected at random for each site. Each residence within the selected block was sampled, skipping and recording those residences where permission was not granted. All the potential containers in both indoor and two meters area of the outdoor were examined. Nine larval surveys were conducted as three surveys per each site from January to October in 2017.
- 2.3. Sampling Procedure. Containers were defined as anything natural or artificial that in its present state was holding water and was capable of holding water for three or more days. All artificial and natural containers were sampled. Pipettes were used to collect all water from containers, and all larvae were placed in individual 50 ml cups with caps, then they labeled (date, place, indoor or outdoor, and sample number) and transported to the laboratory. The sampling variables were recorded for each container at the site using the type of the container and type of the material such as plastic, woody, plant, clay, cement, ceramic, glass, metal, rubber, and rocky. Inner surface color of the container (black, brown, green, yellow, red, gray, and colorless), water depth in the container, water volume, condition of the location (grass, bare soil, pavement, groundcover, and bush), height to base of the container from ground, sun exposure (full sun, partial sun, and full shade), types of detritus within the water (leaves, dirt, or nothing), and the presence and abundance of each Aedes species with each container characteristics were recorded. All collected larvae were counted and identified using available identification keys up to the species level [11]. The mix breeding containers found with both Aedes species during the study were very low, and hence, those data were excluded.

2.4. Data Analysis. Relative abundance of both Aedes species was calculated for different container characteristics. Data were analyzed using IBM SPSS statistics data editor 20.0 version. The one way ANOVA test was applied to test whether there is a significant difference in larval abundance and container characteristics. The level of significant was at a = 0.05. In addition, the chi-square test was applied to check the association between larval abundance and the habitat types.

#### 3. Results

Totally, 1067 containers (346 containers in urban, 367 containers in semiurban, and 356 containers in rural areas) were examined. Totally, 550 containers were positive for *Ae. aegypti* and *Ae. albopictus*. Among them, 245 containers were positive for the urban site, 210 containers were positive for the semiurban site, and 95 containers were positive for the rural site. In this study, we concerned only these two *Aedes* species (*Ae. aegypti* and *Ae. albopictus*) during the larval survey.

3.1. Comparative Larval Abundance of Aedes spp. among Three Selected Localities. 212 larval mosquitoes of Ae. aegypti and 1434 of Ae. albopictus were recorded from total 5463 in the urban site. 109 specimens of Ae. aegypti and 1606 specimens of Ae. albopictus were recorded from total 3457 specimens of all mosquitoes in the semiurban site. 406 specimens of Ae. albopictus were recorded from 1756 specimens in the rural site. The larval abundance of Ae. aegypti was 3.88% in urban and 3.15% in semiurban. There were no records of Ae. aegypti from the rural site. The larval abundance of Ae. albopictus was 46.46%, 26.25%, and 23.12% in semiurban, urban, and rural sites, respectively (Figure 2).

Aedes aegypti showed comparatively similar larval abundance in both urban and semiurban localities, while indicating significantly low larval abundance in the rural site (ANOVA, P = 0.000). There is a significant association between larval abundance of Ae. aegypti and locality types (chi-square test, P = 0.000).

Figure 2 shows the highest larval abundance of *Ae. albopictus* from the semiurban site. They were commonly distributed in both urban and rural sites, with no significance (ANOVA, P = 0.899). The association of *Ae. albopictus* larval abundance with different localities is significant (chi-square test, P = 0.000).

3.2. Occurrence of Aedes spp. in Different Container Types. The larval density was significantly high in artificial containers (90.57%) than that of natural containers (9.43%) for both Ae. aegypti and Ae. albopictus (chi-square test, P = 0.000 and 0.005, respectively).

Table 1 indicates the occurrence of *Aedes* species in different types of natural and artificial containers recorded from three selected localities.

Among the selected types of containers, the high larval abundance of *Ae. aegypti* was recorded in tires (rubber) (17.82%), plastic cups (17.09%), and the metal cups (9.82%), but not statistically significant between each container types

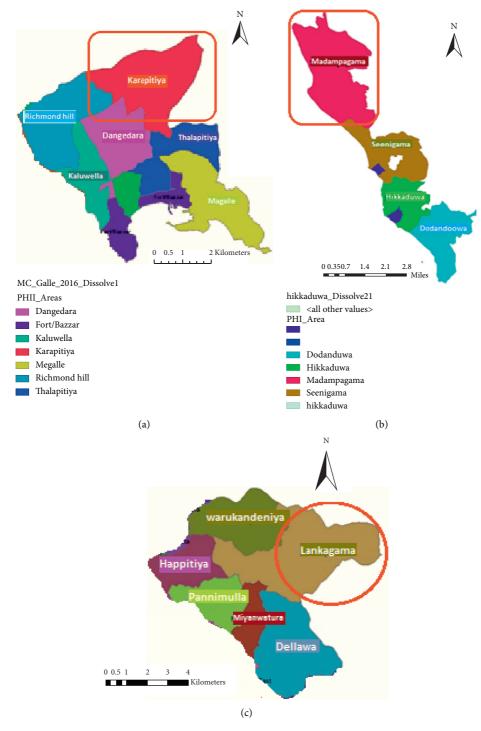


FIGURE 1: The selected sites in the Galle district, Sri Lanka. (a) Urban-Karapitiya. (b) Semiurban-Madampagama. (c) Rural-Lankagama.

(ANOVA, P = 0.123). Among the natural container types, plant axils were the most important for larval abundance of *Ae. aegypti*. The larval abundance in both species was low in rock pools and wood caves (Figure 3).

Plastic cups recorded 28% relative larval abundance for  $Ae.\ albopictus$  as the highly preferable container type, but not statistically significant (ANOVA, P=0.222). The comparative larval abundance of  $Ae.\ albopictus$  was less in tires than

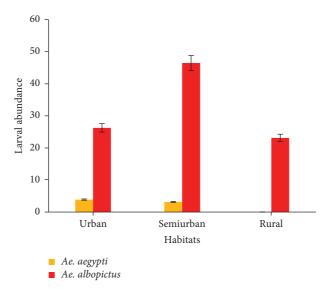


FIGURE 2: Relative abundance of Aedes spp. larvae in three selected habitats.

TABLE 1: Occurrence of Aedes aegypti and Aedes albopictus in different container types.

Type of containers	Type of material	Aedes aegypti	Aedes albopictus
Natural	Animal foot print	_	_
	Burrow pits	_	_
	Clay pots	+	+
	Coconut shells	+	+
	Wood caves	+	+
	Rain water pools	_	_
	Rock pools	+	_
Artificial	Covering items	+	+
	Earth pipes	_	_
	Flower pots	+	+
	Glass boxes	+	+
	Metal cups	+	+
	Ornamental items	+	+
	Pet feeding cups	+	+
	Plant axils	+	+
	Plastic cups	+	+
	Quarry pits	_	_
	Toys	+	+
	Tires	+	+

that of Ae. aegypti. Aedes albopictus also indicated the less larval abundance in wood caves and rock pools (Figure 4).

3.3. Relative Abundance of Aedes Larvae in Containers with Different Colors, in Different Heights, in Different Volumes of Water, and with Different Types of Detritus.

3.3.1. Relative Abundance of AedesLarvae in Containers with Different Colors. Black color containers showed the highest relative abundance for Ae. aegypti (47.98%) compared with containers of other colors, but not statistically significant (ANOVA, P > 0.05). Aedes albopictus (37.32%) also indicated the highest relative larval abundance in black color containers, but not significant (ANOVA, P > 0.05). Brown color containers recorded the second high larval abundance for both Ae. aegypti and Ae.

albopictus. The larval abundance of both Aedes species was significantly high in dark color containers than that of light color containers (yellow and colorless) (t-test, P < 0.05) (Figure 5).

3.3.2. Relative Abundance of AedesLarvae in Containers in Different Heights. Aedes aegypti showed high larval abundance in containers that are located at less than 10 cm heights from the ground. Aedes albopictus indicates a high relative larval abundance in containers found in between 1 and 5 cm heights. Interestingly, figure 6 shows that the both Aedes spp. record their high larval abundance in ground level containers (1-5 cm) and the larval abundance significantly decrease when increasing the height of the containers found (1-20 cm) (ANOVA, P < 0.05) (Figure 6).

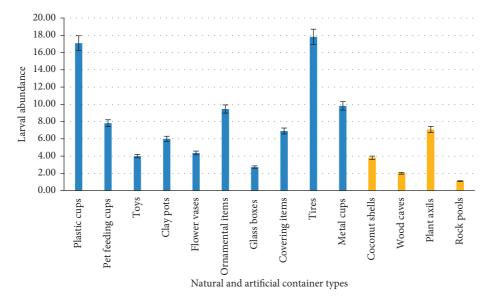


FIGURE 3: Relative abundance of Aedes aegypti larvae in natural containers and artificial containers.

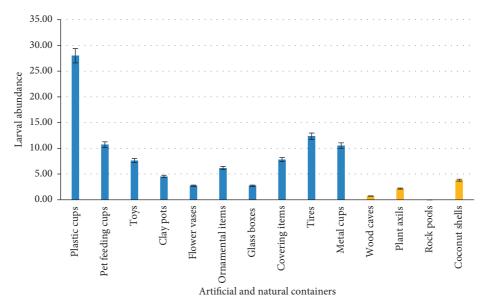


FIGURE 4: Relative abundance of Aedes albopictus larvae in natural containers and artificial containers.

3.3.3. Relative Abundance of AedesLarvae in Containers in Different Volumes of Water. Containers with 50–100 ml volume of water indicated the highest relative larval abundance of both species (29.28% Ae. aegypti and 41.79% Ae. albopictus) (ANOVA, P > 0.05). Both species showed very low larval abundance when the water level becomes very low in containers or when increases more than 150 ml (Figure 7).

3.3.4. Relative Abundance of Aedes Larvae in Containers in Different Shady Levels and Different Types of Detritus. The relative abundance of Ae. aegypti and Ae. albopictus was high in containers located in fully shady places (Figure 8). Their relative abundance increases with high shady level (57.34% Ae. aegypti and 61.32% Ae. albopictus). There is a statistical significance of larval abundance of Ae. aegypti between containers

found in full shade locations and full sun locations (ANOVA, P = 0.049). Aedes albopictus larval abundance is significantly different in containers found from full shady locations and from full sunny locations (ANOVA, P = 0.000).

The different types of detritus occurred in each containers indicated that the larval abundance of both Ae. aegypti and Ae. albopictus was significantly high when leaf litter presents as the detritus in the containers (80.69% Ae. aegypti (ANOVA, P=0.020) and 73.77% Ae. albopictus (ANOVA, P=0.037)). Their larval abundance is very low when sand occurs as the detritus in the container (Figure 9).

#### 4. Discussion

It is well known that Ae. aegypti and Ae. albopictus are main vectors of DF and DHF in Sri Lanka and both are container-

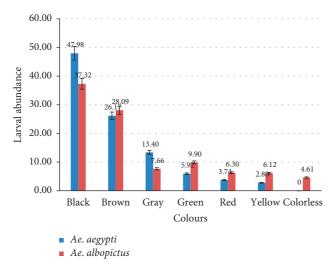


Figure 5: Relative larval abundance of Aedes spp. in containers with different colors.



FIGURE 6: Relative abundance of Aedes spp. larvae in containers with different heights.

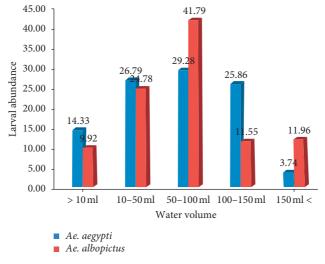


Figure 7: Relative abundance of larvae in containers with different volumes of water.

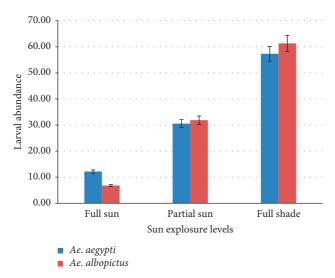


FIGURE 8: Relative abundance of larvae in containers with different shady levels.

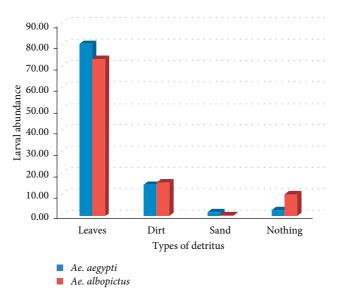


FIGURE 9: Relative abundance of larvae in containers with different types of detritus.

inhabiting mosquitoes that deposit eggs in small type of containers. The present study reveals the effects of container characteristics on the breeding preference of larvae of *Aedes* vector mosquitoes across the selected urban, semiurban, and rural sites in Galle district, Sri Lanka.

Aedes albopictus is the dominant species in all selected sites of the present study. Aedes aegypti is less abundant in urban and semiurban, and there are no records in the rural site. A notable deduction in the population of Ae. aegypti in rural sites has been reported by several previous studies [7, 12–15]. In 2012, Bartlett-Healy et al. further implied that Ae. aegypti is an urban mosquito species while Ae. albopictus prefers semiurban environments and breeds in outdoor natural breeding sites.

In the present study, we excluded the mix breeding sites which found both *Aedes* spp. in same habitat because they were recorded in few numbers.

Compared with the number of positive containers encountered, the numbers of artificial containers are high than that of natural containers, indicating the possible risk of the spread of disease by rapid accumulation of discarded artificial containers in the study area. A study conducted in Central Africa has shown that artificial containers were most frequently encountered than natural containers for both *Ae. aegypti* and *Ae. albopictus* [4]. Furthermore, in 2017, Julien et al. recorded that the predominant breeding sites were industrial containers for *Aedes* vectors in the urban setting in southeastern Côte d'Ivoire.

Among the positive artificial containers recorded, tires (rubber) are the most preferable artificial breeding site for *Ae. aegypti*, while plastic cups are the most preferable artificial breeding site for *Ae. albopictus*. Tires and plastic cups are commonly found artificial discarded materials in all sites during the study. Used tires and plastic containers are the predominantly identified industrial containers [4, 5, 16].

Our study shows that dark color containers have significantly high breeding preference for both *Aedes* spp. than that of light color containers. Dark and cool resting places provide the need of mosquito life cycle [17], black and gray with the species presence for *Ae. albopictus* and *Ae. japonicus* [16]. However, controversy results in the study by [3] presented that *Ae. aegypti* and *Ae. albopictus* larvae were commonly found more in light color containers in Thailand.

Significantly, we found that both *Ae. aegypti* and *Ae. albopictus* record their high larval abundance in ground level containers (1–5 cm) and significantly decrease the larval abundance when increasing the height where containers were found (1–20 cm). As well as, the containers with 50–100 ml water level indicate the highest breeding preference for both *Aedes* species, and their larval abundance becomes very low when the water level increases more than 150 ml. In 2016, Kabirul et al. mentioned that most of *Aedes* species select shallow water depth for their oviposition.

Containers found in shaded areas provide ideal habitats for mosquito larvae. Natural shade not only lowers water temperatures but often provides a bacterial food source for mosquito larvae from fallen leaves and debris that are subsequently collected in containers [18]. The abundance of *Ae. albopictus* larva in shaded or partially shaded areas may be attributed in part to abundant food resources in shaded containers. In the present study, the high relative abundance of *Ae. aegypti* and *Ae. albopictus* was observed in containers of full shady habitats. Containers that expose to direct sunlight show less larval abundance of both *Aedes* mosquitoes.

The study reveals a significant high larval abundance of both *Aedes* species in containers where leaf litter occurs as detritus. Several previous authors addressed that the leaf litter provides an important nutrient source for bacteria, which in turn is the primary food source for mosquito larvae [19, 20], and *Ae. albopictus* prefers to oviposit in containers with leaf litter [16, 21, 22].

The present study provides the most important information on the container characteristics of the breeding preference of *Ae. aegypti* and *Ae. albopictus*, main dengue vectors in Sri Lanka. The study shows that the artificial discarded containers were the most preferred type of breeding habitat in all studied sites, and the accumulation of these containers make serious future health risk for dengue in the study area. Hence, study findings would be helpful for researchers and health authorities to design appropriate vector control measures and to mitigate future dengue outbreaks in the area.

#### 5. Conclusions

Aedes albopictus is the dominant dengue vector species found from all study sites (urban, semiurban, and rural) in the Galle district of Sri Lanka. Artificial containers are more productive for the oviposition of Ae. aegypti and Ae. albopictus than natural containers. Tires are the most preferable artificial breeding site for Ae. aegypti. Aedes albopictus selects plastic cups as the most preferable artificial breeding site. Dark color containers are more favorable that contained

with water between 50 ml and 100 ml volume and locate less than 5 cm height from the ground level for the optimum breeding of *Ae. aegypti* and *Ae. albopictus*. Presence of leaf litter as detritus in the container and the increase of the shady level, positively support to the highest productivity of *Ae. aegypti* and *Ae. albopictus*. The findings of the study help in the effective and efficacious *Aedes* control and management activities in epidemic situations for public health aspects.

## **Data Availability**

The datasets used to support the findings of this study are available from the corresponding author upon request.

#### **Conflicts of Interest**

The authors declare that they have no conflicts of interest.

#### **Authors' Contributions**

Dinithi S. Dissanayake conducted field surveys, data collection, and data entering. Chandana D. Wijekoon analyzed the data and wrote the manuscript. Hemantha C. Wegiriya supervised the research and reviewed the manuscript.

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