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Pre-imaginal development of *Aedes aegypti* in drains containing polluted water in urban cities in Sri Lanka

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Abstract

Dengue control in Sri Lanka targets the removal or treatment of fresh water bearing containers that provide breeding sites for *Ae. aegypti*. Health authorities have raised concern that the incidence level of dengue remains high in urban settings due to ignorance of dengue mosquito larval surveillance on polluted water drains. The present study was designed to evaluate the presence of *Ae. aegypti* in the polluted water drains in some urban areas in the country. A preliminary entomological survey was carried out in urban areas from Chilaw to Galle and further studies were carried out in the drains with polluted water that marked the presence of *Ae. aegypti* larvae. Presence of *Ae. aegypti* was recorded in drains containing polluted water in Galle city for the first time in Sri Lanka. The most commonly observed species in the preliminary survey was *Cx. quinquefasciatus* (97.5%) followed by *Cx. gelidus* (0.99%), *Ae. albopictus* (0.62%) and *Ae. aegypti* (0.42%). Water quality characteristics revealed low dissolved oxygen and high conductivity with visible organic pollutants. Constant insecticide pressure and the destruction of the breeding sites through source reduction in vector control campaigns compel the mosquito to adapt to new ecological niches. Therefore, appropriate surveillance and control measures should be implemented by health authorities to control vector mosquitoes and lower the disease burden.

Keywords *Aedes aegypti* · Pre-imaginal stages · Polluted water · Sri Lanka · Water quality

Introduction

Mosquitoes are considered as important insect invaders due to their ability to introduce novel diseases to naïve human populations through their proximity to human habitations. Most medically important mosquito vectors are very well adapted to various human habitats. *Aedes aegypti* is an invasive mosquito that has caused the most human casualties worldwide as the primary vector of viruses that cause Dengue, Yellow Fever, Chikungunya and Zika (Kraemer et al. 2015). *Ae. aegypti* is generally considered to be a clean water breeder and a wide range of water holding containers have been identified as the breeding grounds for the mosquito (Tun-Lin et al. 1995). The most important container types for oviposition and development of larval stages include water storage tanks, tires, small trash items and water containing jars and barrels (Kweka et al. 2018; Murrell

et al. 2011). Although early field investigations have indicated that *Ae. aegypti* is usually absent from water containing decomposing organic matter, recent research findings indicate the presence of *Ae. aegypti* developmental forms in water containing high concentrations of decomposing organic matter (Mackay et al. 2009; Murrell et al. 2011; Nguyen et al. 2014).

In Sri Lanka, Dengue Fever (DF) is endemic and is considered to be the most serious arboviral disease with an increasing number of dengue cases per year. Year 2017 marked the highest number of DF cases with 185,688 cases recorded throughout the island (Epidemiology unit 2019). Due to the dry environmental conditions the number of DF cases reported has decreased during the year 2018. However the monsoon rains impending are expected to escalate the number of reporting DF cases. (Epidemiology unit 2019). Highest number of dengue cases (>45%) has been reported from the urban areas in the western province which is the main commercial province in Sri Lanka.

Dengue control campaigns in the country traditionally focus on source reduction and the use of adulticides and larvicides specially around the households where dengue cases recorded. (Fernando et al. 2018). These campaigns usually

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target the possible breeding places containing clear water, including discarded receptacles, roof gutters and water storage tanks (Sirisena and Noordeen 2014). Despite regular control activities carried out by the health authorities, dengue continues to be a huge burden in the country (Hegoda et al. 2017; Fernando et al. 2018). Recent research findings revealed that the breeding of *Ae. aegypti* in brackish water is a possibility of *Ae. aegypti* adaptation to new habitats in the country (Ramasamy et al. 2011; Jude et al. 2012). Ignorance of the polluted water drains in vector control programs has raised concerns regarding the possibility of *Ae. aegypti* mosquito breeding in these drains. The present study was designed to evaluate the presence of *Ae. aegypti* in the polluted water drains in some urban areas in the country.

Materials and methods

The coastal belt from Chilaw to Galle comprises of urban and suburban areas with high human population density. These areas have reported high DF incidences continuously during the past few years irrespective of the rigorous dengue control campaigns with the use of insecticides and removal of the mosquito breeding places. All the urban and suburban areas have storm water drains. Unauthorized waste disposal into these storm water drains in the form of domestic and small industry waste have resulted in water pollution in these drains. Thus most of the water drains are polluted with organic and synthetic pollutants.

A preliminary entomological survey on water drains was carried out in urban areas from Chilaw to Galle along the coastal belt in Sri Lanka to evaluate the mosquito species present in drains containing polluted water (Fig. 1). The survey included all the major water drains in each city. Specimens of larvae and pupae were collected with the use of standard dippers (WHO 1975) from the study sites. The collected samples were kept in plastic bottles, labeled separately according to the sites and days of collection and were transported to the laboratory. Larvae and pupae were reared in their original water samples and the emerged adults were identified according to the standard mosquito keys (Belkin 1962).

Presence of *Ae. aegypti* was recorded in drains containing polluted water in Galle city, thus further studies were conducted within the Galle city. Collections of mosquito samples were carried out monthly from June 2017 to August 2017 in Galle with the use of standard dippers. The water drains in the city sites were sampled once a month for the presence of *Ae. aegypti* immature stages.

In order to determine the water quality of the drains which contained *Ae. aegypti* pre-imaginal stages in the Galle city, water quality parameters and the selected characteristics were measured during the time of mosquito larvae collection. Water quality (Clear vs turbid), water odor (present or absent) types

of organic matter present and the status of the organic matter present were recorded. Water quality characteristic such as temperature, pH, turbidity, dissolved oxygen, salinity, conductivity (YSI Proplus, USA) were measured.

Results and discussion

We examined 106 individual drains for the preliminary entomological survey, all of which held water at the time they were examined and all the drains yielded mosquito immature stages (Fig. 2). In total 26,901 immature stages were collected representing seven species (Table 1). From the emerged adults predominant species was *Cx. quinquefasciatus* ($n = 26,245$) with a percentage of 97.56% recording from all sites. This was followed by *Cx. gelidus* (0.99%), *Ae. albopictus* (0.62%) and *Ae. aegypti* (0.42%). Other species recorded were *Armigeres sp.*, *Cx. hutchinsoni* and *Ae. vittatus* which accounted for only 0.4% of the specimens collected. Interestingly *Ae. aegypti* larvae were only recorded from the polluted water drains that were sampled in the Galle district..

Only four mosquito species were recorded during the three months field study carried out in the Galle district. *Cx. quinquefasciatus* (97.2%) was predominately recorded from all the polluted drains sampled, with a total of 9594 mosquitoes collected. This was followed by *Ae. aegypti* (1.58%) with 156 mosquitoes and *Ae. vittatus* with 77 mosquitoes (0.78%). *Cx. gelidus* was the only other species recorded from the drains in the Galle city.

Of the examined drains majority of drains contained polluted water at the time of inspection and held turbid water (TDS 520–1280 mg/L), often accompanied with pollutants and domestic waste. Water temperature was in the range of 27 °C – 30 °C. Dissolved oxygen level varied between 0.06 mg/L – 1.8 mg/L and pH of the water collected ranging from 7.1 to 7.9. Salinity and conductivity levels varied between 0.3 ppt – 1.0 ppt and 501.6–2178 $\mu\text{s}/\text{cm}$ respectively. The preferred water quality parameters for *Ae. aegypti* mosquito larvae have been previously studied. These include 85.6–114.9 mg/L turbidity, 5.97–6.53 mg/L dissolved oxygen level, 7.18–7.66 pH and 128.2–177 $\mu\text{s}/\text{cm}$ conductivity levels (Dom et al. 2016). The results of the current study reinforce that *Aedes* mosquito larvae can thrive in a variety of habitats.

Interestingly most drains in the Galle city that held clear water had the presence of *Poecilia reticulata*, thus no immature stages of mosquitoes were found in these drains. The drains containing organic waste were the only ones that recorded the presence of *Ae. aegypti* and interestingly absence of *Poecilia reticulata*. However larvivoracious fish species including *Poecilia reticulata*, *Aplocheilus sp.* and *Oreochromis mossambicus* have been used in organically polluted water in

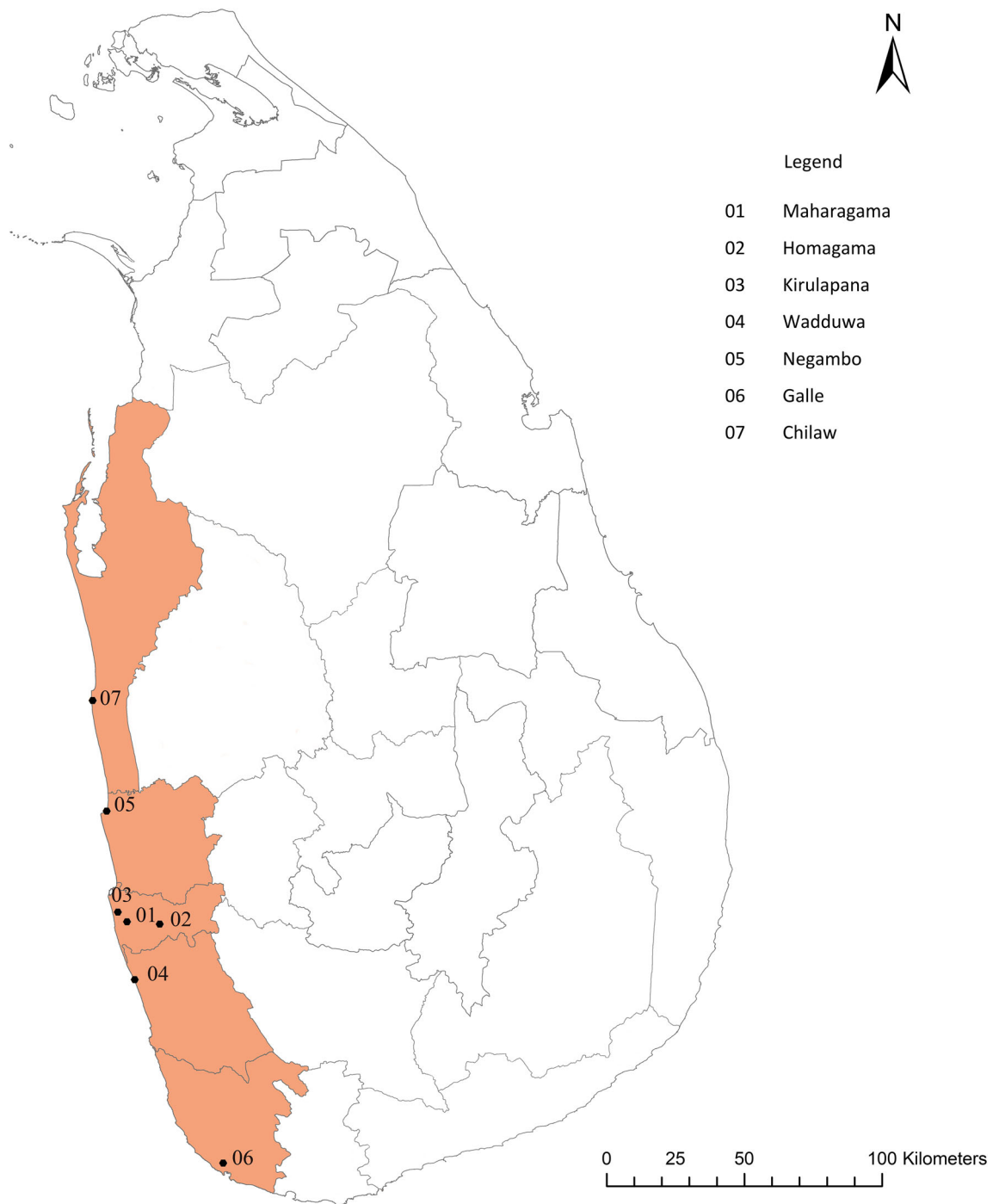


Fig. 1 Map showing the study sites

Sri Lanka (Sirisena and Noordeen 2014) Thus using integrated vector control programs with the inclusion of biological control methods must be implemented.

The present study reveals the presence of *Ae. aegypti* pre-imaginal stages in polluted water drains for the first time in Sri Lanka. This study agrees with the other studies (Barrera et al. 2008; Burke et al. 2010) that have recorded the presence of *Ae. aegypti* in polluted water. Larval surveys that have been carried out in Sri Lanka in potential clean water breeding sites

have reported similar number of larval counts (Weeraratne et al. 2013). These surveys collected mosquito larvae among a variety of breeding places including discarded receptacles and groundwater storage tanks. Our results indicate that drains may serve as an important breeding ground for *Ae. aegypti* mosquitoes thus being instrumental in the transmission of dengue in urban areas. Consequently, drains should be strongly considered for inclusion in the local mosquito control programs.

Fig. 2 Storm water drains where the presence of *Ae. aegypti* larvae was recorded



Other than *Ae. aegypti* two other species of belonging to genus *Aedes*, *Ae. albopictus* and *Ae. vittatus* were present in polluted water drains. Both these species have been identified as potential and secondary vectors of arboviral diseases including dengue, chikungunya and zika (Gratz 2004; Sudeep and Shil 2017). There have been evidences of the capability of *Ae. albopictus* as a vector in transmitting DENV in the absence or low abundance of *Ae. aegypti* (Sirisena and Noordeen 2014). Although the vector status of these mosquitoes needs further investigation, studies have suggested that these mosquitoes play an important role in maintenance and transmission of arboviruses (Gratz 2004; Sudeep and Shil 2017). As noted above, *Culex* was the predominant species in the drains followed by *Aedes* species. Storm sewers and catch basins have been recorded to serve as breeding grounds for *Cx. quinquefasciatus* (Geery and Holub 1989; Su et al. 2003; Hribar et al. 2004; Anderson et al. 2006). Although cases of Filariasis is reported at a minimal rate in Sri Lanka, in the event of the presence of parasite re-emergence of the disease or an outbreak would be inevitable.

A study of population genetics and vector competency of the *Ae. aegypti* mosquitoes emerging from the polluted water drains was not carried out during the present study. An examination of the size differences in adult mosquitoes may also reveal information regarding the longevity, feeding frequency, reproductive capability and flight potential of the mosquito. Thus incorporating these aspects in the future studies may provide information regarding the ability of these mosquitoes to disseminate and transmit the DEN virus.

The risk of disease transmission is positively associated with an increase in vector mosquito density and incidence (Steiger et al. 2016). Mosquitoes particularly *Ae. aegypti* has the ability to adapt to human built environments thus making it an excellent urban exploiter. Constant insecticide pressure and the destruction of the breeding sites through vector control campaigns forces the mosquito to adapt to new ecological niches. Thus, better understanding of the micro-geographical ecology and the behavior of mosquito species is very important in understanding the spread of the disease. Therefore, appropriate surveillance and integrated control measures

Table 1 Species recorded in the studied drains containing polluted water in the urban cities during the preliminary survey

Locality	No. of polluted drains sampled	Number and the species emerged from the collected larva						
		<i>Cx. quinquefasciatus</i>	<i>Cx. gelidus</i>	<i>Cx. hutchinsoni</i>	<i>Ae. aegypti</i>	<i>Ae. albopictus</i>	<i>Ae. vittatus</i>	<i>Armigeres sp.</i>
Galle	21	8668	5	8	112	38	54	25
Chilaw	25	11,569	177	0	0	116	0	11
Kirulapana	24	482	22	0	0	8	0	12
Homagama	11	866	22	0	0	3	0	0
Maharagama	11	466	0	0	0	3	0	0
Wadduwa	19	1058	11	0	0	0	0	0
Negombo	25	3136	29	0	0	0	0	0

should be implemented by health authorities to control these vector mosquitoes.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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