Journal of Advances in Microbiology Research

E-ISSN: 2709-944X
P-ISSN: 2709-9431
JRM 2022; 3(1): 61-64

www.microbiojournal.com
Received: 11-01-2022
Accepted: 16-02-2022

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Assessment of larval indices and physicochemical parameters of the breeding habitat of dengue vector in Barnala city, Punjab (India)

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DOI: https://doi.org/10.22271/micro.2022.v3.i1a.181

Abstract
During the cross-sectional immature stage survey carried out to investigate the presence of larvae of *Aedes aegypti* in human habitation in Barnala, Punjab (India), potential breeding sites/containers (With or without larvae) were found in almost all houses surveyed. Larvae of *Aedes aegypti* were identified morphologically using a compound microscope. 32 houses out of 156 houses surveyed, had positive containers (With larvae). 57 containers out of 692 artificial water containers with potential for breeding were found to be positive. The larval indices, house index, 20.52% (95% confidence interval = 17.20–23.70), container index, 8.24% (95% C.I. = 7.28–9.38), and breteau index, 36.54% (95% CI = 34.24–38.66), were found higher than the critical levels. The water containers located away from direct sunlight and with ineffective lids had a higher larval density. TDS, EC, levels of the water in the containers as well as the material of the container had no discernible impact on the presence of *Aedes* larvae, according to the present investigations.

Keywords: *Aedes aegypti*, immature stage, larval indices, containers

1. Introduction
Dengue infection caused by a vector-borne arbovirus of the flavivirus family, is transmitted by the female mosquitoes *Aedes aegypti*, *Aedes albopictus, Aedes scutellaris, and Aedes polynesiensis.* Dengue infection has grown dramatically in recent years and is prevalent in more than 129 countries, with an estimated 96 million clinical cases [1]. It continues to be a significant public health concern in all regions in India [2, 3]. Female of *Aedes sp.* (Diptera: Culicidae) the primary vector of dengue [4] prefers to breed in natural or human-made water collections found in human habitations such as houses and peridomestic areas [5, 6].

The major breeding habitats include abandoned tires, flower pots, discarded plastic or metal containers, desert coolers, swimming pools, plant axils, ant traps, earthen pots, flower pots, drums, concrete tanks, coconut shells, etc. [7, 8], which hold clear and clean water for a long time [9, 10]. Because of their impact on vector bionomics, and vectorhost interactions, climatic and environmental factors have been shown to play a part in dengue transmission dynamics [12]. The physicochemical properties of breeding water affect the development and survival of mosquito larvae [13].

Since there is no specific vaccine and treatment available for dengue [14], it is essential to collect data on vector density, the population at infection risk, and the susceptibility of mosquitoes to insecticides to manage vectors [15, 16] and for the prevention and control of dengue outbreaks [17].

For this monitoring of dengue vector population distribution over time in each region, as well as the larval indices (Container Index (CI), House Index (HI), Breteau Index (BI), and Pupal Index (PI)), have become essential tools.

In 2015, the state of Punjab reported the highest number of dengue cases among all states in India and has been reporting approximately 10,000 dengue cases every year since then[18]. Punjab is facing its worst-ever dengue outbreak with over 18,266 confirmed cases of dengue, including 60 casualties, its highest toll in recent years [19]. There are few reports about preferred breeding habitat and larval indices from other districts of Punjab but no information available in Barnala as per the literature perusal, athough in recent years, a number of dengue cases recorded in Barnala city, This study was, therefore, carried out to determine larval indices, and physicochemical parameters of the breeding habitat of dengue. vector in the municipal area of Barnala, Punjab, India.

2. Materials and Methods

This study was carried out between September - October 2021 in Barnala city of Punjab, India, situated at latitude 30° 23' 0" North and longitude 75° 33' 0" East. Throughout the study period, there were only minor daily and seasonal temperature fluctuations in the typically hot and humid climate. In present study conducted as a cross-sectional survey in the residential area, 156 houses were sampled for larvae, selecting 15-20 randomly from each of the nine areas on basis of ecological and demographic characteristics. All potential indoor and outdoor water-holding habitats/containers in every house were inspected by following the dipping method [20]. pH (using pH strips), TDS, and electrical conductivity (Using portable meters) of water in each positive container, were measured.

The larvae and pupae collected into well-labeled, water-holding bottles were taken to the Biology Laboratory, S.D. College, Barnala, and maintained at 26±2 °C.

Larvae of only *Aedes aegypti* were taken into account and were identified using a compound microscope and the pictorial keys[21]. *Aedes* larvae were identified by observing comb scales on the terminal segment. Comb scales are with a distinct middle denticle and lateral denticles (Pitch-fork shape) in *Aedes aegypti* larvae while with a single denticle (Thorn-like shape) in *Aedes albopictus* larvae [22]. Mosquito larval indices (House index, container index, and breteau index) were calculated by following the guidelines of the World Health Organization [4].

House index (HI) =Number of houses infested Number of houses inspectedX100

Container index (CI) = Number of positive containers Number of containers inspected X 100

Breteau index (BI) =Number of positive containers Number of houses inspected X 100

3. Results and Discussion

In this study, *potential breeding sites* (With or without larvae) were located in almost all houses surveyed. Positive containers (With larvae) were present in 32 houses out of 156, showing a calculated house index of 20.52% (95% confidence interval = 17.20–23.70).

Out of 692 artificial water containers with potential for breeding, 57 containers were found to be positive, leading to a calculated container index of 8.24% (95% C.I. = 7.28–9.38) and breteau index of 36.54% (95% C.I. = 34.24–38.66) (Table 1). Entomological surveillance revealed high larval indices in different localities of the city (Fig.1).

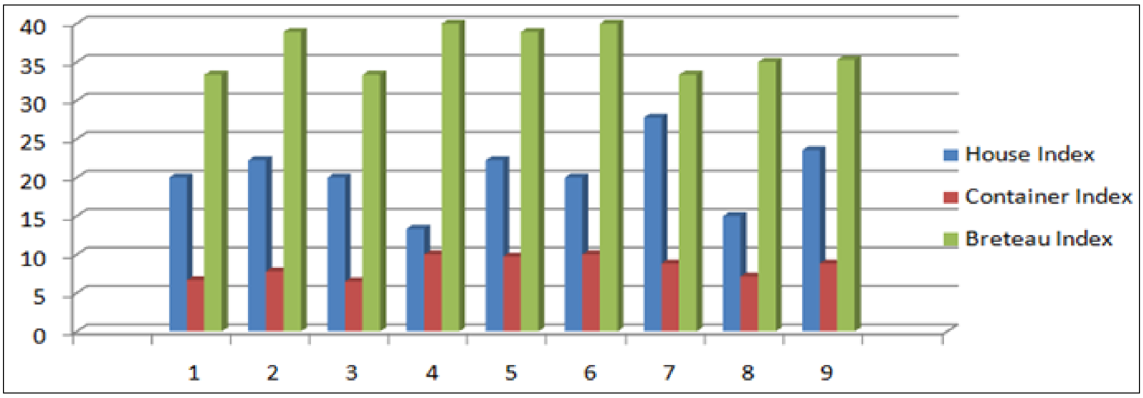

Fig 1: Larval indices in different nine areas studied

Generally, 10% and 5% for the house index and breteau index, respectively, are taken as critical levels, beyond which epidemics are likely to occur;. BI above 50%, represents a very high-risk area, and between 5 and 50%, moderate-risk area [4]. In the present study, the Breteau index (BI) is 36.45%, showing a moderate-risk area.

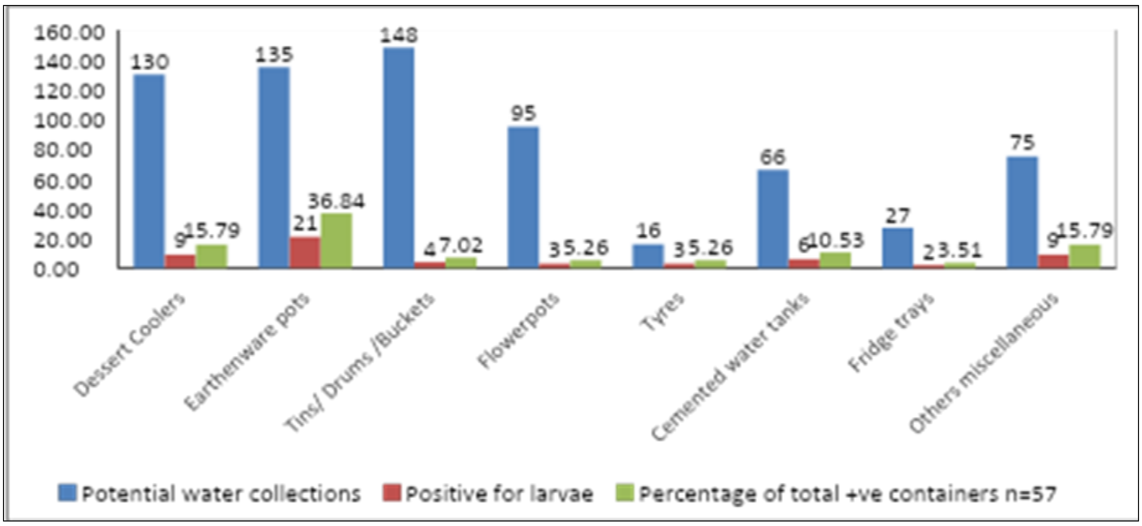

Fig 2: Distribution of different collections with potential for mosquito breeding and proportion of containers with larvae and comparison of percentage of total +ve containers

Breeding was noted in various artificial containers as given in fig.2. Earthenware pots (36.84%) were the most likely containers to harbour mosquitoes, followed by desert coolers (15.79%), cement water tanks (10.53%), and water storage buckets or drums. Used disposables like tins and glasses etc. in peridomestic areas were discovered to contain larvae, among other things. The current findings are consistent with other studies that found that desert coolers, earthen or mud pots, were important *Aedes* breeding habitats [23, 24].

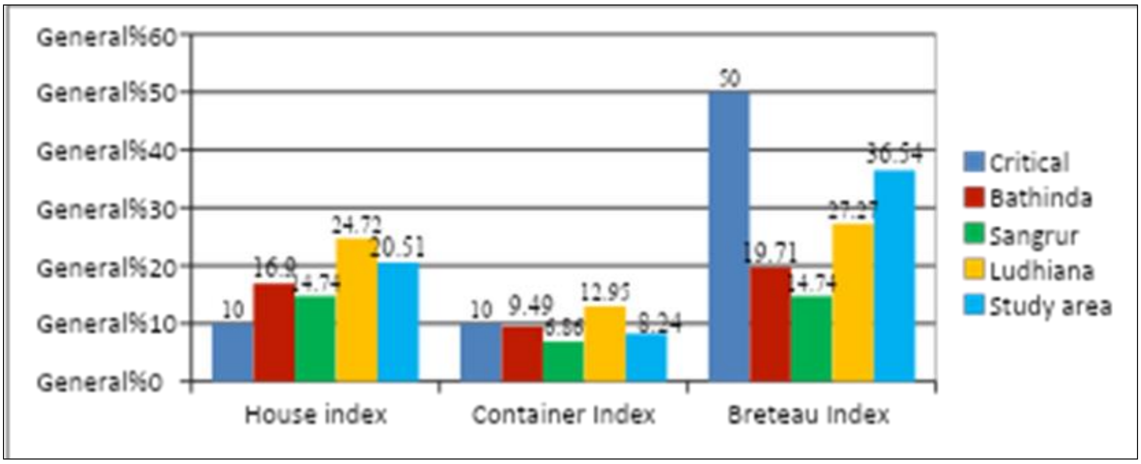

Fig 3: Comparison of larval indices in our study area with critical level and other studies in Punjab

According to [25] Overgaard *et al.*, (2017), the characteristics of the water in containers influence both immature stages' development as well as the oviposition behaviour of female mosquitoes. The present investigation showed an average temperature of 31.02±0.71; an average pH of 7.44±0.58, an average TDS of 217.49±1.35 mg/l, an average EC of 817.3±57.53 µS/cm.

TDS, EC, levels of the water in the containers as well as the material of the container had no discernible impact on the presence of *Aedes* larvae, according to the present investigations. Various studies have shown that the pH of water has a significant correlation with larval density in container-breeding mosquitoes [26, 27, 28]. The location of the water container and lid status were found to influence the density of *Aedes* larvae, in certain areas where people store water in buckets, tins, or drums due to an insufficient or unreliable water supply. In these areas, it was found that the water containers located away from direct sunlight and with ineffective lids had a higher density of larvae when compared to the containers that were uncovered or with effective lid containers. This finding is in agreement with that of [29] Getachew *et al.* (2015), who have reported that nearly every species of mosquito lays eggs in a shaded or less-lit area.

In the present study, positive containers were found more frequently in houses with vegetation or with water storage practices and also in abandoned containers that hold water for a long period, lying in peridomestic areas, possibly due to improper cleaning practices or unmanaged waste accumulation. These findings may enable a more focused approach to vector control in which specific types of water-holding containers would be targeted.

Conclusion

From the present study, it is certain that the area under study (Barnala) has a potential for dengue viral infection, and the characteristics of the breeding habitat affect the larval density. Increasing awareness among people about the proper cleanliness and disinfection of water-holding containers is recommended. The data obtained from this study will serve as a baseline for the control of the dengue vector mosquito. Further studies are recommended to evaluate vast area and the presence of larvae in large tanks in study area and research on virus dynamics within the mosquito must also be considered.

Acknowledgments

The authors declare that there is no conflict of interest. The authors would like to thank the workers from Civil Hospital, Barnala for their support. The authors are also grateful to the family members of the students for their assistance to visit houses in different areas for data collection.

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