# Physicochemical characteristics of habitats in relation to the density of container-breeding mosquitoes in Asom, India

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# ABSTRACT

*Background & objectives:* Container-breeding mosquitoes, especially *Aedes* spp are vectors of diseases such as dengue and chikungunya. The abundance of these disease vectors in an area depends on the availability of container habitats and their physicochemical characteristics. The species composition of container-breeding mosquitoes in Asom, India was studied and the larval density was correlated with the habitat characteristics.

*Methods:* Natural and man-made water-holding containers in Sonitpur district of Asom were surveyed for the presence of mosquito larvae. The percent composition of container-breeding mosquitoes and container index were calculated. The physicochemical characteristics of breeding water such as pH, conductivity, salinity, total dissolved solids, turbidity and dissolved oxygen were measured.

*Results: Aedes albopictus* (93.7%) was the predominant species in the container-breeding habitats whereas *Culex quinquefasciatus* (2.77%), *Armigeres subalbatus* (2.26%), *Ae. aegypti* (0.76%), *Toxorhynchites* sp (0.4%) and *Lutzia* sp (0.11%) were recorded in relatively low numbers. The larval density (mean  $\pm$  SE<sub>mean</sub>) of the container-breeding mosquitoes ranged from 4.4  $\pm$  1.8 to 15.4  $\pm$  8.2, while the container index ranged from 1.58 to 5.68%. The mean ( $\pm$  SE<sub>mean</sub>) pH, conductivity, salinity, total dissolved solids, turbidity, and dissolved oxygen of water in the container habitats were 7.15  $\pm$  0.11; 396.1  $\pm$  58.5  $\mu$ S/cm; 0.24  $\pm$  0.04 ppt; 207.1  $\pm$  30.4 mg/l; 32.3  $\pm$  5.1 NTU; and 1.42  $\pm$  0.12% respectively. The mosquito larval density in the container habitats was having significant negative correlation with the conductivity of breeding water (r = -0.89; p = 0.003). Salinity, total dissolved solids and turbidity of water in the habitats were negatively correlated, whereas pH and dissolved oxygen were positively correlated with the larval density.

*Interpretation & conclusion:* The studies indicated the predominance of *Ae. albopictus* in the container-breeding habitats and reiterated its importance as a potential vector of dengue and chikungunya in the region. The spread of *Ae. aegypti*, the principal vector of dengue, in the semi-urban areas probably through road transport is a matter of public health concern. The use of conductivity of breeding water as an index for the proliferation of container-breeding mosquitoes in the region could be explored further.

Key words Asom; container habitats; mosquito vectors; physicochemical characteristics

#### INTRODUCTION

Mosquitoes are the most important group of insect vectors of human diseases such as malaria, chikungunya, filariasis, dengue and Japanese encephalitis. The aquatic habitats in which they breed include pools, swamps, paddy-fields and water-holding containers. The container habitats have unique ecological properties and these habitats could be natural such as tree holes and leaf axils or artificial such as tyres, plastic cups and water tanks<sup>1</sup>. Aedes aegypti, Ae. albopictus and Culex quinquefasciatus are the most predominant species among the container-breeding mosquitoes<sup>2</sup>. These mosquitoes are important from the public health perspective as they include disease vectors as well as potential biological control agents. Aedes aegypti is the primary vector of dengue<sup>3</sup>

whereas *Ae. albopictus* is the vector of many arboviral diseases including dengue and chikungunya<sup>4–5</sup>. *Culex quinquefasciatus* is the vector of lymphatic filariasis, Japanese encephalitis virus, West Nile virus and the secondary vector of western equine encephalitis virus<sup>6</sup>.

The northeastern region of India, having heavy rainfall and a wide range of breeding habitats is vulnerable to the incidence and transmission of mosquito-borne diseases especially malaria and Japanese encephalitis<sup>7–8</sup>. The species composition of the container-breeding mosquitoes and their habitat characteristics need to be studied in the context of the emergence of dengue and chikungunya in this part of the country. The information on the ecological factors influencing mosquito biology such as the physicochemical properties of breeding water could help in better implementation of the vector management programmes<sup>9</sup>. Hence, the present study was carried out to understand the physicochemical characteristics of the habitats in relation to the density and diversity of container-breeding mosquitoes in the state of Asom, northeastern India.

#### MATERIAL & METHODS

## Study area

The studies on the water-holding containers supporting mosquito breeding were conducted in the semi-urban and the surrounding rural areas at eight sites, namely Kauripathar, Borgang, Biswanath Ghat, Pubjamuguri, Nahoroni, Goroimari, Sirajuli and Puthimari in the Sonitpur district (92° 20' to 93° 45' E and 26° 20' to 27° 05' N) of Asom, India.

# Larval surveys

In each study site, the natural containers such as bamboo stumps, coconut shells, tree-holes, leaf axils as well as the artificial containers such as earthen pots, plastic cups, glass bottles and tyres were examined for the presence of mosquito larvae. The surveys were carried out twice in each of the four seasons, namely pre-monsoon (March–May), monsoon (June–August), post-monsoon (September–November) and winter (December–February) during March 2010 – February 2011. The collected larvae were brought to the laboratory and identified with the help of standard keys<sup>10</sup>.

# Larval density and diversity

The abundance of container-breeding mosquitoes was calculated as, Larval density = (Total number of larvae collected / Total number of positive containers). The container index (CI) was calculated as, CI = (Number of positive containers / Total number of containers inspected) × 100. The species composition of the container-breeding mosquitoes was estimated as, Percent composition = (Number of larvae belonging to the species / Total number of larvae collected) × 100.

# Physicochemical characteristics

Water samples were collected from the containers, which showed the presence of mosquito larvae. The breeding water characteristics were pH, conductivity ( $\mu$ S/cm), salinity (ppt), total dissolved solids (mg/l), turbidity (NTU) and dissolved oxygen (%) were recorded using Orion 5-star portable multiparameter meter (Thermo Scientific).

# Data analysis

The mean larval density among the study sites was

compared using one-way ANOVA. The relationships between the mean values of the habitat characteristics and the larval densities of the container-breeding mosquitoes were derived by Pearson's correlation and linear regression. The statistical analyses were carried out using IBM SPSS 19 statistical software.

## RESULTS

The density of container breeding mosquitoes (mean  $\pm$  $SE_{mean}$ ) was the highest in Sirajuli (15.4 ± 8.2) followed by Borgang (14  $\pm$  8.2), Puthimari (12  $\pm$  7.5) and Kauripathar (11.8  $\pm$  7.5). The densities were lower in Goroimari  $(6.5 \pm 3.3)$ , Pubjamuguri  $(5 \pm 1.5)$ , Biswanath Ghat  $(4.9 \pm 1.5)$  and Nahoroni  $(4.4 \pm 1.8)$  (Fig. 1). However, the larval density did not vary significantly among the study sites (p > 0.05). The container index, which indicates the percentage of positive containers at the study sites ranged from 1.58% in Nahoroni to 5.68% in Borgang. The CI for Kauripathar, Biswanath Ghat, Pubjamuguri, Goroimari, Sirajuli and Puthimari were 4.64, 1.97, 2.14, 2.8, 4.32 and 4.68% respectively. Among the larvae collected from the water-holding containers, 93.7% belonged to Ae. albopictus whereas 2.77% to Cx. quinquefasciatus, 2.26% to Armigeres subalbatus, 0.76% to Ae. aegypti, 0.4% to Toxorhynchites sp and 0.11% to Lutzia spp.

Analysis of the physicochemical characteristics of water in the mosquito breeding containers indicated that the pH ranged from  $6.72 \pm 0.11$  in Kauripathar to  $7.63 \pm 0.18$  in Sirajuli whereas the conductivity ( $\mu$ S/cm) ranged from  $162.9 \pm 22.3$  in Sirajuli to  $616.9 \pm 93.5$  in Nahoroni. The salinity (ppt) was the lowest in Sirajuli ( $0.09 \pm 0.01$ ) and the highest in Kauripathar ( $0.39 \pm 0.11$ ) whereas the total dissolved solids (mg/l) was the highest in Kauripathar



*Fig. 1:* Larval density of container-breeding mosquitoes (mean  $\pm$  SE<sub>mean</sub>) in the study areas in Asom state, India.

Sites surveyed	pH	Conductivity (µS/cm)	Salinity (ppt)	Total dissolved solids (mg/l)	Turbidity (NTU)	Dissolved oxygen (%)
Kauripathar	$6.72 \pm 0.11$	431.4 ± 142.2	$0.39 \pm 0.11$	308.5 ± 89.1	50.1 ± 13.6	$1.23 \pm 0.15$
Borgang	$7.18 \pm 0.11$	$233.5 \pm 38.1$	$0.12 \pm 0.02$	$104.3 \pm 19.4$	$33.8 \pm 18.4$	$1.76 \pm 0.18$
Biswanath Ghat	$6.80 \pm 0.13$	$537.4 \pm 156.5$	$0.28 \pm 0.08$	$263.4 \pm 76.6$	$58.5 \pm 76.6$	$1.46 \pm 0.13$
Pubjamuguri	$6.94 \pm 0.17$	411.6 ± 162.7	$0.34 \pm 0.14$	$130.8 \pm 16.8$	$22.8 \pm 7.08$	$1.26 \pm 0.07$
Nahoroni	$7.37 \pm 0.06$	616.9 ± 93.5	$0.29 \pm 0.05$	$302.3 \pm 45.8$	$28.6 \pm 7.18$	$1.14 \pm 0.07$
Goroimari	$7.18 \pm 0.08$	529.6 ± 116.6	$0.27 \pm 0.06$	$260.3 \pm 56.8$	$20.7 \pm 4.91$	$1.11 \pm 0.06$
Sirajuli	$7.63 \pm 0.18$	$162.9 \pm 22.3$	$0.09 \pm 0.01$	$112.7 \pm 12.1$	$22.7 \pm 3.08$	$1.30 \pm 0.25$
Puthimari	$7.41 \pm 0.19$	$245.7 \pm 34.8$	$0.18 \pm 0.07$	$174.2 \pm 65.6$	$21.3 \pm 8.79$	$2.11 \pm 0.72$

Table 1. Physicochemical characteristics of habitats of container-breeding mosquitoes in Asom state, India

 $(308.5 \pm 89.1)$  and the lowest in Borgang  $(104.3 \pm 19.4)$ . The turbidity of breeding water ranged from  $20.7 \pm 4.91$  in Goroimari to  $58.5 \pm 76.6$  in Biswanath Ghat whereas the dissolved oxygen (%) ranged from  $1.11 \pm 0.06$  in Goroimari to  $2.11 \pm 0.72$  in Puthimari (Table 1).

The correlation between the physicochemical characteristics (mean  $\pm$  SE<sub>mean</sub>) and the larval density of container-breeding mosquitoes indicated that the pH (7.15  $\pm$ 0.11) and the dissolved oxygen (1.42  $\pm$  0.12%) showed

Table 2. Breeding habitat characteristics in relation to the larval density of container-breeding mosquitoes in the study areas of Asom state, India

Physicochemical parameters	Mean ± SE <sub>mean</sub>	Correlation coefficient	<i>p</i> - value
рН	$7.15 \pm 0.11$	0.4	0.33
Conductivity (µS/cm)	396.1 ± 58.5	-0.89	0.003
Salinity (ppt)	$0.24 \pm 0.04$	-0.67	0.07
Total dissolved solids (mg/l)	$207.1 \pm 30.4$	-0.53	0.18
Turbidity (NTU)	$32.3 \pm 5.1$	-0.13	0.75
Dissolved oxygen (%)	$1.42 \pm 0.12$	0.46	0.25



*Fig. 2:* Relationship between the larval density of container-breeding mosquitoes and the conductivity of breeding water (r = -0.89; p = 0.003) in study areas in Asom state, India.

positive correlation with the larval density, the correlation coefficients being 0.4 and 0.46 respectively. Salinity ( $0.24 \pm 0.04$  ppt), total dissolved solids ( $207.1 \pm 30.4$  mg/l) and turbidity ( $32.3 \pm 5.1$  NTU) were negatively correlated with larval abundance in the containers, with correlation coefficients of -0.67, -0.53 and -0.13respectively. However, none of these correlations was significant (p > 0.05) while significant negative correlation was observed between water conductivity ( $396.1 \pm 58.5$ ) and the larval density (r = -0.89; p =0.003) (Table 2). The regression relationship between the larval density (LD) and the water conductivity (WC) was derived as, LD = 18.83 - 0.024 WC ( $R^2 = 0.791$ ) (Fig. 2).

### DISCUSSION

In the case of container-breeding mosquitoes, the selection of containers depends on the volume, size of the water surface and the type of material of which they are made<sup>11</sup>. In the present study, the larval density per container varied from 4.4 to 15.4 and the surveys revealed that Sirajuli and Borgang were having high larval density and container index (CI). A container index of > 20 for *Ae. aegypti* is associated with dengue/dengue haemorrhagic fever in India<sup>12</sup> whereas the range of CI at the study sites was 1.58–5.68. High percentage of breeding sites of *Ae. albopictus*, was recorded in earlier studies in the urban (51.2) and industrial (18.6) areas of Asom<sup>13</sup>.

The predominant species among the container-breeding mosquitoes was *Ae. albopictus*, whereas *Ae. aegypti* constituted <1% of the total larval collections. Apart from *Aedes* mosquitoes, *Cx. quinquefasciatus* and *Ar. subalbatus* were found to be widely distributed in the water-holding containers across the study sites. The larvae of *Toxorhynchites* spp mosquitoes are predatory and are potential biocontrol agents for vector mosquitoes especially *Aedes* spp and *Cx. quinquefasciatus*<sup>14</sup>. The larvae of *Lutzia* spp, which are predatory on *Cx. quinquefasciatus*<sup>15</sup>, were collected from some areas although in very low numbers.

The pH of the breeding water in the present study ranged from 6.72 to 7.63 and showed a positive correlation (r = 0.4) with the larval density. The survival of Ae. aegypti and Cx. quinquefasciatus larvae was found to be the maximum in the pH range of  $6.5-8^{16}$ . Manipulation of the pH of the breeding sites by spraying of biopesticides such as neem oil could be used as a management tool for the container-breeding mosquitoes<sup>9</sup>. Among the physicochemical parameters studied, water conductivity with a range of 162.9-616.9 µS/cm, was observed to have significant negative correlation with the larval density. Negative correlation of the larval density with the water conductivity was also observed in the case of malaria vectors of The Gambia, wherein the conductivity above 2000  $\mu$ S/cm led to significant reductions in the larval density<sup>17</sup>. The salinity of water in the mosquito breeding habitats varied from 0.09 to 0.39 ppt and showed negative correlation with the larval density. Laboratory studies on the selection of oviposition sites by Ae. aegypti revealed that the oviposition decreased with the increase in salinity and there was almost no oviposition above 12%18.

However, the studies on the breeding habitat characteristics of *Ae. albopictus* in Calicut, India indicated positive correlation of the larval density with the total dissolved solids, turbidity, conductivity and salinity whereas negative correlation with the pH<sup>9</sup>.

The breeding habitat characteristics were found to exert a significant influence on the abundance of container-breeding mosquitoes in the study areas in Asom, India. The Asian tiger mosquito Ae. albopictus was widely prevalent at all the sites surveyed, which reiterated its importance as the potential vector of dengue and chikungunya in this region. The spread of Ae. aegypti, the principal vector of dengue, in the region is a matter of public health concern. This species was more prevalent in semi-urban areas with close proximity to roads and vehicle garages indicating the probable role of road transport in its geographical spread. The conductivity of breeding water, which was shown to be highly correlated with the larval density could be used for early warning of the proliferation of disease vectors in an area. However, more studies are needed to validate such relationships in other parts of northeastern India, before these could be utilised for predicting disease outbreaks. Further, these relationships would also help to enhance the efficacy of the ovitraps used for the monitoring of disease vectors by modifying the physicochemical parameters.

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