

CHARACTERISTICS OF *Aedes (Stegomyia) albopictus* Skuse (Diptera : Culicidae) BREEDING SITES

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Abstract. We investigated the physicochemical characteristics of *Aedes albopictus* Skuse *viz.* breeding sites, by examining coconut shells, tires and plastic containers in a chikungunya affected area of Calicut, India. The study design was a cross-sectional ecological type. Thirty-four water samples were collected randomly from coconut shells, tires and plastic containers. The larvae were counted and the water was examined. The mean number of larvae per 500 ml was significantly higher in coconut shells than in tires and plastic containers. A negative correlation was observed with pH and a positive correlation was seen with most other variables. Coconut shells were the preferred breeding site for *Ae. albopictus* mosquitoes in our study. The rich organic content, low illumination and small orifice of the coconut shells could be the reasons for this. The higher salinity of the coconut shells did not affect breeding or survival.

Keywords: *Aedes albopictus*, physicochemical characteristic, breeding site

INTRODUCTION

The global resurgence and expanded distribution of vector borne diseases, such as dengue fever and chikungunya infection has generated a renewed interest in the biology and control of *Aedes (Stegomyia)* mosquitoes in recent years. *Aedes aegypti* (Linnaeus) is a principal vector of these diseases in several countries. *Ae. albopictus* also is a vector of disease on many continents, including North America, Europe, Africa, Australia and in the Asia Pacific region (Gartz, 2004; Charrel *et al*, 2007). *Ae. albopictus* is an adaptive

and invasive species co-existing with or displacing *Ae. aegypti* in various regions (Paupy *et al*, 2009). This mosquito usually breeds in natural habitats especially in tree holes, leaf axils, rock pools and similar sites (Hawley, 1988). Widespread deforestation, climate change and increase in global trade has forced this mosquito worldwide to adapt to breeding in domestic and semi-domestic artificial container habitats (Gubler *et al*, 2001; Delatte *et al*, 2008).

Chikungunya virus infections have been increasing in Southeast Asia and India during the last decade (Stock, 2009); thousands of cases have been reported from several southern states of India (Saxena *et al*, 2006). Localized outbreaks of chikungunya virus infection have occurred in many districts of Kerala (NVDCP, 2006);

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the number of cases in Calicut District has increased over the years and a full-scale epidemic occurred in 2009 (Anonymous, 2010). *Ae. albopictus* was shown to be the major vector of chikungunya infection in those areas (Themozhi *et al*, 2007).

Efforts to control *Ae. albopictus* have met with serious setbacks due to lack of resources, an insufficiently trained work force, poor community participation and indiscriminate waste disposal practices of indigenous communities. Control programs are aimed mainly at source reduction activities by village health workers and volunteers who did not achieve an adequate reduction in vector density in the affected areas. A good knowledge of ecological factors influencing mosquito biology, especially in the selection of breeding sites, such as physical and chemical properties of water, could benefit future mosquito management programs. Few reports are available regarding the ecology of this mosquito. Hence, a cross-sectional ecological comparative study was carried out to determine the physical and chemical characteristics of mosquito breeding sites in affected areas of Calicut, Kerala.

MATERIALS AND METHODS

This study was carried out in Calicut City, the capital of Calicut District, situated at 11.25°N longitude and 75.77°E latitude. The topography consists of hills, plains and coastal areas, and has a tropical climate with an average rain fall of 3,000 mm and a temperature ranging from 21°C to 39°C. Chikungunya and dengue virus infections have a seasonal trend coinciding with the southwest monsoon occurring from June to November, due to increases in vector density; the samples were collected during this period. Samples were collected randomly from

coconut shells, discarded plastic objects (eg, cups, bottles and buckets) and discarded tires. Water samples were collected in 500 ml plastic containers, and where the quantity of water was insufficient, a number of breeding sites were pooled to make up the required volume. A total of 34 samples were collected from sites detected around households.

Larvae were collected using a strainer and brought to the laboratory, where *Ae. albopictus* larvae were identified and counted (Barraud, 1934). The water samples were analyzed for temperature, pH, conductivity, salinity, alkalinity, total dissolved solids, turbidity, hardness, phosphate, nitrate, sulfate and calcium using standard APHA (2006) procedures at the Center for Water Resources Development and Management, Calicut, Kerala, India.

Means and standard deviations (SD) were used for summarizing the physicochemical factors. Correlation of physicochemical factors with the number of larvae was done using the Pearson's correlation coefficient test. Differences in the physicochemical factors among different breeding sites were determined using the ANOVA, and when necessary using the Bonferroni test as a post hoc test. A p value ≤ 0.05 was considered statistically significant. SPSS (Chicago, IL) version 10 software was used for analysis.

RESULTS

Table 1 shows the means and standard deviations (SD) for the physicochemical factors at the different breeding sites. Temperature and pH did not vary significantly by breeding site. There were significant differences in conductivity, total dissolved solids, salinity, turbidity, sulphate, alkalinity, hardness and calcium between the coconut breeding sites and

Table 1
Physicochemical patterns of different breeding sites studied (Mean± (SD))

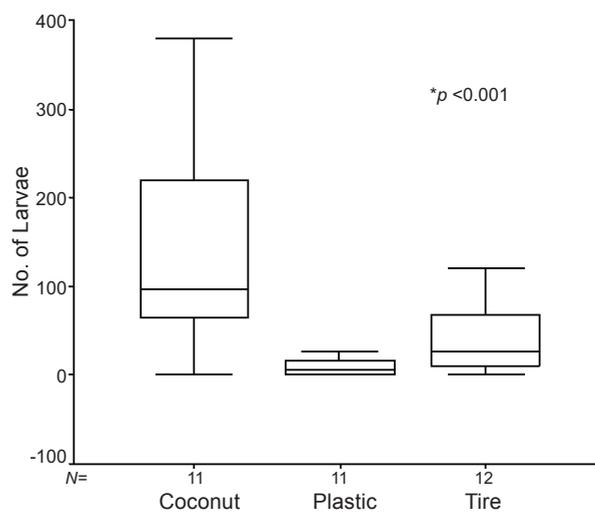
Physicochemical characters	Coconut n=11	Plastic n=11	Tire n=12	p-value ^a
Temperature	26.9 (1.3)	26.5 (0.7)	27.3 (0.7)	0.138
pH	7.3 (0.3)	7.5 (0.3)	7.6 (0.4)	0.033
Conductivity	1,415.9 (795.3)	73.6 (51.7)	109.1 (48.7)	<0.001
Total dissolved solids	962.9 (509.9)	52.4 (36.6)	86.5 (47.1)	<0.001
Salinity	0.6 (0.5)	0.04 (0.02)	0.05 (0.02)	<0.001
Turbidity	215.0 (219.2)	32.9 (43.1)	28.0 (33.3)	0.002
Sulphate	85.6 (100.2)	2.9 (1.7)	7.0 (5.3)	0.002
Nitrate	4.7 (3.0)	1.1 (1.3)	1.3 (1.8)	0.001
Phosphate	10.3 (9.4)	0.02 (0.02)	0.7 (1.6)	<0.001
Alkalinity	804.8 (930.6)	28.4 (25.3)	40.8 (13.4)	0.002
Hardness	563.6 (409.7)	33.5 (27.2)	54.7 (34.7)	<0.001
Calcium	122.7 (58.7)	12.4 (14.0)	14.0 (5.8)	<0.001

^ap-value by ANOVA test

the other container breeding sites investigated. Fig 1 shows the larval density of *Ae. albopictus* in 500 ml water samples collected from different breeding sites. The mean number of *Ae. albopictus* larvae collected from the coconut breeding sites was significantly higher than the plastic and tire breeding sites. The densities of larval breeding in order from greatest to least were coconuts >tires>plastic containers. Table 2 shows the correlation coefficients for *Ae. albopictus* larval density with the various factors investigated. A positive correlation was observed with all parameters except pH.

DISCUSSION

The selection of breeding sites by mosquitoes is a critical factor for mosquito survival and population dynamics and has important implications for mosquito control. Site may be affected by chemical and physical factors: attractants and deterrents. A deterrent causes insects to avoid



*p-value by ANOVA test

Fig 1—*Ae. albopictus* larval abundance in different breeding sites.

that area, inhibiting egg laying. Sharma *et al* (2008) demonstrated the repellent effect of some fatty acid esters on oviposition of mosquitoes. Overposition site selection is important in the life cycle of mosquitoes.

Table 2
Correlation coefficient between
Ae. albopictus larval abundance and
physicochemical factors.

Parameters	Correlation coefficient	p-value
Temperature	0.08	0.652
pH	-0.49	0.003
Conductivity	0.82	<0.001
Total dissolved solids	0.81	<0.001
Salinity	0.73	<0.001
Turbidity	0.48	<0.001
Sulphate	0.86	<0.001
Nitrate	0.42	0.012
Phosphate	0.38	0.033
Alkalinity	0.59	<0.001
Hardness	0.66	<0.001
Calcium	0.77	<0.001

Stimuli for an ovipositional flight are linked with some environmental factors, such as rainfall, relative humidity, temperature and wind speed; the selection of a breeding site involves visual, olfactory, and tactile responses (O'Gower, 1963; Bentely and Jonathan, 1989).

Ae. albopictus exhibits a great deal of specialization in breeding site selection and consequently the distribution of this species is limited by those sites. The spread of *Ae. albopictus* from Asia to the Americas and Europe was a direct consequence of the egg laying behavior of this species (Bonn, 2006). Female mosquitoes have adapted to laying eggs in artificial containers, especially discarded tires, which resist desiccation, the eggs then hatch after being transported in used tires from Asian countries to Western countries (Medlock *et al*, 2006; Straetemans, 2008). It is possible species that breed in containers, both natural and artificial, may rely on

some sort of visual analysis for site location and ensuing oviposition. *Ae. albopictus* females may utilize smell and contact chemoreception for choosing breeding sites and prefer water with low illumination and dark color (Gubler, 1977).

In a study in Delhi (Seghal and Pillar, 1970), *Ae. aegypti* and *Ae. albopictus* were found to favor identical breeding sites in regard to turbidity, pH, alkalinity, chloride and phosphate levels. However, they did not prefer the same water conditions. *Ae. albopictus* preferred less turbid waters with moderate alkalinity whereas *Culex* and *Anopheles* mosquitoes preferred more turbid water with high alkalinity and less oxygen content. The sulphate content was lower in the *Culex* and *Anopheles* breeding sites than the *Aedes* breeding sites, but the chloride content was higher in the *Culex* and *Anopheles* breeding sites and lower in the *Aedes* breeding sites. The phosphate content was low in *Ae. albopictus* breeding sites. The authors concluded mosquitoes exhibited species specific preferences in oviposition in regard to the chemical composition of water.

Various physical and chemical factors of larval habitats contribute to mosquito breeding site selection, such as temperature, pH, ammonia, nitrate, sulphate, phosphate and dissolved solids (Oyewole *et al*, 2009; Oleyemi *et al*, 2010). In our study we found a correlation between factors investigated and larval prevalence. Coconut shells were chosen more often by mosquitoes for breeding sites than other containers, with more *Ae. albopictus* eggs found in these habitats.

Most of the coconut shells were discarded after use or had fallen from a tree and had been gnawed open by a rodent. Coconuts are rich in calcium, potassium, sodium, sulphur and magnesium forming

an ideal breeding ground for *Ae. albopictus*. They have a small orifice and are dark inside, making it an ideal mosquito breeding site.

The pH of water has an impact on mosquitoes, influencing osmoregulation and oxygen transportation (Umar and Donpedro, 2008). In this study of *Ae. albopictus* breeding, an increase in pH resulted in a significant decrease in larval density. The container sites sampled had a comparable pH levels. This demonstrates that *Ae. albopictus* mosquitoes favor a specific pH range for breeding. This could be an important factor in breeding site selection and larval survival. A pH variation outside the range of 7-8 could be used as a tool for management of this vector. Manipulation of the pH of preferred breeding sites, such as coconut shells, tires and plastic containers could be used to control this mosquito where breeding site reduction is not possible due to local factors. Further studies regarding this aspect need to be carried out. Spraying biopesticides, such as neem oil with a pH >8 could be useful for this purpose. Neem oil, a natural product, is environmentally safe and may be less harmful than synthetic insecticides.

Salinity is another important factor that can have a repelling or attracting effect on oviposition. Navarro *et al* (2003) found increasing salinity in the laboratory decreased oviposition. However, our findings showed *Ae. albopictus* was tolerant to variations in salinity, possibly leading to further geographical expansion of this mosquito's breeding sites. Previous studies found *Aedes* prefers to breed in less turbid waters, similar to that found in plastic containers and tires; however, the high turbidity of the coconut shell breeding sites contradicts those findings. The rich organic content of coconut shells

could be a reason for this. Carbonate was not detected in any of the samples we analyzed.

In conclusion, physicochemical characteristics exert a significant influence on mosquito breeding site selection among *Ae. albopictus*. Coconut shells were the preferred breeding site for this mosquito in our study, followed by tires and plastic containers. These preferences could be exploited to develop novel techniques to deter oviposition.

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