

## DENGUE VECTOR DISTRIBUTION AND ABUNDANCE IN LOWER RIO GRANDE VALLEY, TEXAS USA

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**Abstract** Dengue virus has been classified as a worldwide emerging disease. South Texas is potentially at risk for dengue virus introduction due to proximity to Mexico, a country with endemic dengue transmission. Previous research has identified two dengue vectors, *Aedes aegypti* and *Aedes albopictus* in the region. Both species share similar habitats and often compete as larvae. Understanding the distribution and relative abundance of these species will aid in the process of determining risk for dengue virus transmission. We examined the relative abundance of these two species in six sites extending from the Gulf of Mexico inland approximately 60 miles. Three different habitats were surveyed at each field site: a tire store, a cemetery, and a residential location. Mosquitoes eggs were collected weekly using oviposition traps. Temperature and humidity were recorded during each collection. We predicted we would find a greater abundance of *A. aegypti* further away from the shoreline where the humidity was lower. We also predicted we would find greater abundance of *A. aegypti* in tire shops, where there is little cover. Preliminary analysis indicates *A. albopictus* were more common in cemeteries, while *A. aegypti* were more common in tire shops. Both appeared to be equally abundant in residential area. There is no correlation between species abundance and distance from the Gulf of Mexico. These data provide insight into the potential for dengue virus introduction and transmission in the region.

**Key words** Dengue virus, *Aedes albopictus*, *Aedes aegypti*.

### INTRODUCTION

Dengue virus is a vector borne disease that is increasing in prevalence in North and South America (Ehrenkranz et al., 1971; Gubler, 1998; Guzman and Kouri, 2003; Siqueira et al., 2004; World Health Organization, 2009; Jansen and Beebe, 2010). Dengue has recently been classified as an emerging pathogen due to increased prevalence worldwide. Dengue virus historically has been transmitted in Northern Mexico; however there is an extreme disparity between the infections rates of dengue virus and Mexico. From 1980 to 1999, over 60,000 dengue cases were reported in Mexican border states, while Texas only had 64 cases (Reiter et al., 2003). Endemic dengue activity was reported in 2013 in South Texas, suggesting that despite a lack of reported cases in the past, dengue is an ongoing concern (Oliveres, personal communication).

*Aedes aegypti* is the primary vector of dengue, although *Aedes albopictus* has been shown to be capable of transmitting the disease as well (Ibanez-Bernal et al., 1997; Gratz, 2004; Kurane, 2007; Erickson et al., 2010). Both mosquitoes are commonly found in habitats that are associated with human presence and activity. *Aedes aegypti* and *Aedes albopictus* share similar biological characteristics, are both container-breeding mosquitoes and often compete with each other for resources (Sota and Mogi, 1992; O'Meara et al., 1995; Alto et al., 2008; Frances et al., 2011; Richards et al., 2012). The result of their competitive actions influences the distribution of both species. Many studies have been conducted focusing on container breeding mosquito interactions, but the majority of these studies have focused on the Florida environment (Harper and Paulson, 1994; O'Meara et al., 1995; Daugherty et al., 2000; Juliano and Lounibos, 2005; Leisnham and Juliano, 2009).

While studies in Florida, a subtropical region in the United States, have suggested that *A. albopictus* may outcompete *A. aegypti* in more humid areas, they have also suggested that in drier, hotter areas *A. aegypti* may be able to persist (Juliano et al., 2001). South Texas, and specifically the Lower Rio Grande Valley, is a subtropical region as well. However, this region of Texas that is generally hotter and drier than Florida. The Lower Rio Grande Valley is adjacent to Northern Mexico where endemic dengue is present. Understanding vector distribution and potential disease transmission dynamics is of increasing importance in this region.

Our research focused on examining habitat preference between *A. aegypti* and *A. albopictus*. Determining habitat preference can assist in controlling these vectors, possibly limiting or preventing potential outbreaks of dengue virus in the lower Rio Grande Valley. We conducted a field survey for *A. aegypti* and *A. albopictus* to examine habitat preference and abundance based on habitat type, as well as changes in preferences moving closer to the shoreline. We hypothesized we would find a greater abundance of *A. aegypti* further away from the shoreline and in tire shops, and a greater abundance of *A. albopictus* closer to the shoreline and in cemeteries.

## MATERIALS AND METHODS

**Oviposition Trapping.** From May to August 2013 field collections for the mosquitoes *Aedes aegypti* and *Aedes albopictus* were conducted weekly from trapping sites within the cities of McAllen (3 sites), Weslaco (1 site), Mercedes (2 sites), Los Fresnos (2 sites), Laguna Vista (1 site), and Port Isabel (2 site). The cities were chosen based on their relative proximity to the Gulf of Mexico. Trapping sites were classified as cemeteries, tire shops, and residential areas. Cemeteries were well shaded with well-tended lawns but little understory vegetation. Tire shops had little to no shade but a high number of potential oviposition sites in used tires that were scattered around the locations. Residential areas were well shaded, often with many shrubs or bushes, and many hand well-tended lawn. Ten to fourteen oviposition traps were placed in shaded or protected regions in the sites depending on the size of the site. Oviposition traps consisted of a black cups filled with 300mL of deionized water and a scraped wooden tongue depressor (stake) that was secured to the cup with a binder clip. Each week the stakes were collected and replaced. Water still left in cups were inspected for mosquito larvae and replaced. Missing or damaged cups were replaced as needed. Temperature and humidity for each site was recorded at each collection. All stakes and water containing larvae were returned to the laboratory for inspection.

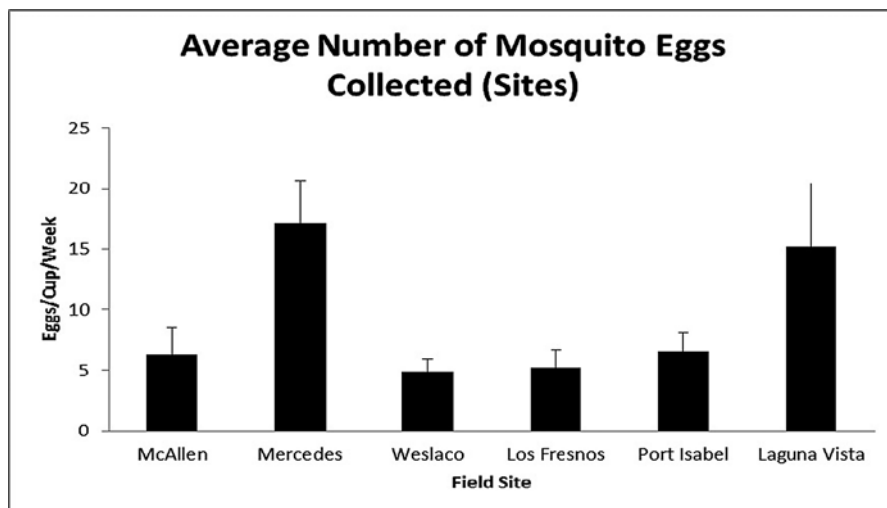
**Egg and Larvae Collection.** All larvae collected from the field sites were counted and recorded. Larvae were placed in rearing pans at low densities (50 larvae per small pan or 150 larvae per large pan). For each wooden stake the total number of unhatched and hatched or damaged eggs was recorded. All stakes containing unhatched eggs were hatched in an aerated 1.0g/L nutrient broth and stayed submerged for 24 hours as described by Vitek and Livdahl (2006). After the eggs were hatched, the larvae were counted and placed in rearing pans at the previously mentioned densities and reared to adulthood. Pupated larvae were transferred into smaller containers, 10 pupae per container, and left to emerge to adults. Adult mosquitoes were frozen and identified to species and recorded as *A. aegypti*, *A. albopictus*, or other. After hatching, wooden stakes were inspected again and total number of hatched, viable, and nonviable eggs were recorded.

## RESULTS AND DISCUSSION

Both vectors species were found in all locations, although there were clear habitat preferences between the two species. The total number of eggs per cup was calculated to use a common measure for analysis. A multiple factor ANOVA was used to analyze the total eggs collected per cup, with site location, habitat type, and week

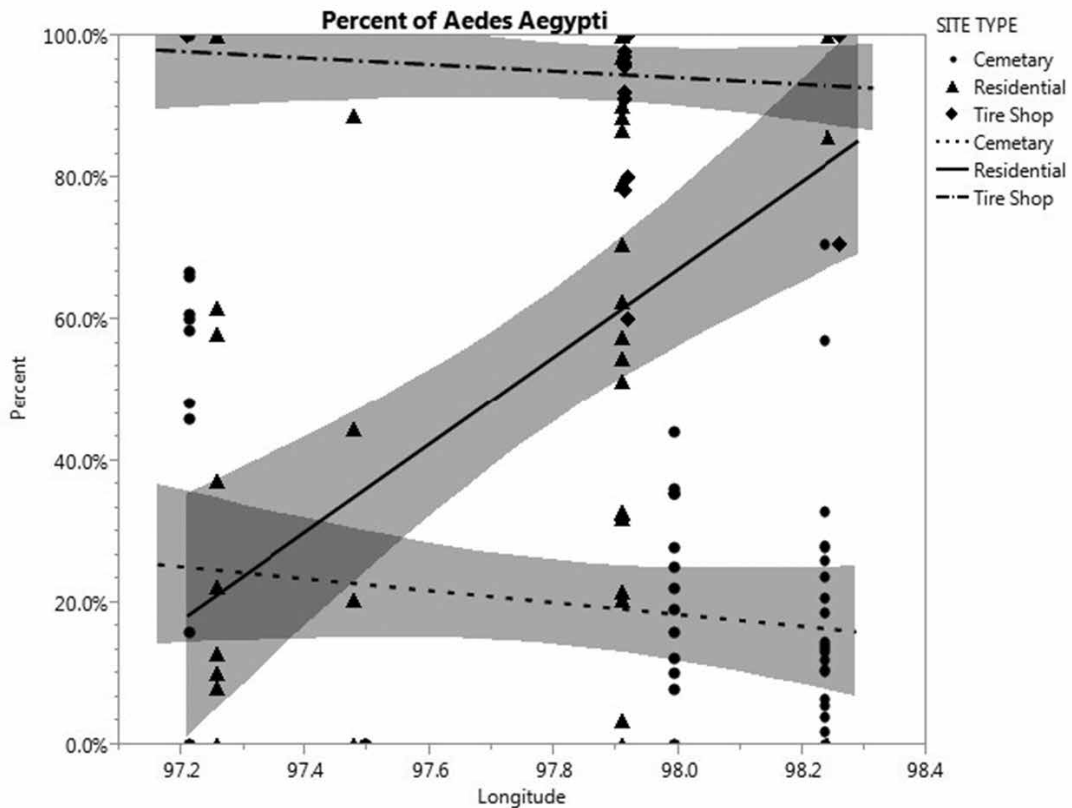
as potential explanatory variables (non-significant interactions were removed). Week was not significant and removed as a potential variable ( $F = 0.923$ ,  $p = 0.339$ ,  $df = 1$ ). Habitat type was significant in the multi-factor ANOVA ( $F = 4.222$ ,  $p = 0.017$ ,  $df = 2$ ), with cemeteries having significantly more mosquitoes than residential or tire shop locations. In addition, a significantly larger number of mosquitoes were collected in two field sites (Figure 1,  $F = 5.732$ ,  $p = 0.00001$ ,  $df = 5$ ) although this was not correlated with distance from the coast.

Eggs that were collected and hatched showed a high level of survivorship to adulthood. Overall, 87.4% of the eggs that were hatched were successfully reared to adulthood, suggesting an estimated abundance of both species based on adult identification would be accurate. The percentage of *A. aegypti* adults to *A. albopictus* adults was calculated from hatched eggs to determine a relative abundance of both species, with week, longitude, and habitat type as potential variables. A multiple factor ANOVA was used to calculate the differences between the potential variables, and non-significant interactions were removed from the analysis. Habitat type was significant ( $F = 93.451$ ,  $p < 0.0001$ ,  $df = 2$ ). A Least Squares Means Students t-test was used to compare sites, and a significantly higher proportion of *A. aegypti* mosquitoes in tire shops and a significantly smaller proportion in cemetery sites. Longitude significantly interacted with habitat type ( $F = 17.918$ ,  $p < 0.0001$ ,  $df = 2$ ). A regression line for longitude using each habitat site was developed, with significant positive linear relationship for residential sites ( $F = 24.1967$ ,  $p < 0.0001$ ,  $r^2 = 0.35$ , Figure 3). The regression lines for cemetery and tire shop sites were non-significant, indicating the relative proportion of *A. aegypti* did not change relative to longitude. Both of these vectors have a short flight range (Reiter et al. 1995, Harrington et al. 2005) suggesting the localized risk estimates based on specific local habitat or conditions may be critical to estimate risk of exposure. These results highlight the role local conditions may play in vector abundance, including the abundance of multiple species. Leisnham and Juliano (2009) reported densities of *A. aegypti* differed among land categories, being more abundant in residential areas compared to industrial and commercial areas, whereas densities of *A. albopictus* did not differ. The change in proportion of *A. aegypti* in residential areas suggests that risk of dengue transmission may vary based on the specific geographic location and conditions rather than overall estimate of mosquito abundance.



**Figure 1.** Average number of eggs per cup per week among field sites.

Sites with different letters are significantly different from each other. Sites arranged furthest from the coast (left) to closest to the coast.



**Figure 2.** Abundance of *Aedes aegypti* by site and longitude. Moving inland, the percentage of *A. aegypti* is significantly different in residential sites, no change in tire shops and cemeteries with longitude (non-significant regression lines). Lines drawn with 95% confidence intervals.

Previous work also suggested a temporal change in relative abundance of *A. aegypti* to *A. albopictus*. While week by itself was not significant in the multi-factor ANOVA ( $F = 0.281$ ,  $p = 0.597$ ,  $df = 1$ ), week did interact significantly with longitude ( $F = 9.815$ ,  $p = 0.0021$ ,  $df = 1$ ). Interestingly, of the six sites, only McAllen (furthest inland) showed any significant relationship between week and percentage of *A. aegypti* ( $F = 6.032$ ,  $p = 0.019$ ,  $r^2 = 0.137$ ). The McAllen area was where the previous collection was conducted, showing the same negative relationship.

The Lower Rio Grande region of South Texas represents an at-risk location that has historically been neglected as a potential focal point of vector-borne disease introduction and transmission. The increased poverty level, including the rural poor often living in substandard housing locally referred to as *colonias*, may represent both an at-risk population as well as a potential reservoir population for diseases like dengue virus and chikungunya virus.

Previous epidemics in South Texas have been linked to corresponding outbreaks in Northern Mexico (Rawling et al., 1998; Brunkard et al., 2007; Brunkard et al., 2008; Adalja et al., 2012). However, the recent identification of 4 locally acquired cases in late 2013, suggest that dengue is more prevalent than previously accepted and maybe a continual thread (Oliveres, personal communication). These results indicate the common presence of both vector species, as well as specific habitat preferences between the two species, further highlight the importance of coordinated research and control efforts to assess and minimize risk of vector-borne disease in the region.

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