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Cemeteries as sources of *Aedes aegypti* and other mosquito species in southeastern Puerto Rico

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Abstract

Objective: To investigate the presence and abundance of mosquito species in containers found in different types of cemeteries in Puerto Rico to assess their importance and make control recommendations.

Methods: We conducted surveys of containers with water in 16 cemeteries in southeastern Puerto Rico to detect the presence of larvae and pupae of *Aedes aegypti* and other mosquitoes; to identify the most common and productive containers and to study their variation in relation to the type of cemetery.

Results: The most common containers with water were flowerpots, followed in abundance by a variety of discarded containers and open tombs. We found a positive relationship between density of containers with water and rainfall. There was a rich community of mosquito species developing in containers of the inspected cemeteries: nine mosquito species belonging to four genera with *Ae. aegypti* and *Ae. mediovittatus* being the most frequent and abundant. We sampled 13 cement-type cemeteries, 2 mixed and only 1 lawn cemetery, consequently, we could not draw any conclusion regarding container productivity and cemetery type.

Conclusions: Surveyed cemeteries were important sources of *Ae. aegypti* and other mosquitoes in flowerpots, discarded containers and open tombs. We recommend conducting further studies to establish how frequently inspections should occur; and mosquito control by emptying aquatic habitats and larviciding to reduce mosquito productivity and protect workers and visitors from mosquito bites and possible transmission of arboviruses.

Keywords

Aedes aegypti ; cemeteries; flowerpots; mosquito control; Puerto Rico

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INTRODUCTION

Cemeteries can become important sources of *Aedes aegypti* (L.) mosquitoes that transmit dengue, chikungunya and Zika viruses in urban and rural environments. These mosquitoes lay eggs and their larvae develop in water-filled container habitats such as flowerpots and open tombs. Cemeteries also provide adult mosquitoes with nectar from flowers in flowerpots and surrounding vegetation, and visitors or cemetery personnel could be sources of blood for biting mosquitoes [1–4].

Different studies have shown the importance of cemeteries in mosquito production and what variables affect mosquito productivity in these environments. Environmental variables play a major role, but structural and cultural variables are also important. Rainfall, air temperature, humidity, wind speed, vegetation cover, sunlight exposure and content of organic matter of containers are important environmental variables influencing mosquito productivity in cemeteries [5–7].

Cultural traditions also play an important role. In many countries, people visit cemeteries on traditionally important days such as Mother's and Father's Day, Memorial Day, Christmas and Remembrance Day. On these dates, visitors usually bring fresh flowers and gifts that have the potential to become aquatic habitats (bottles, cans, foods and decorations among others). Thus human activity generates aquatic habitats for urban mosquitoes all year long in cemeteries [1,5]. The introduction of flowers, water and containers can be particularly important during the dry season, since these become the only available habitat for mosquito development then, supporting viable population densities [5,8].

The structural characteristics of cemeteries are also important. Cemeteries with intricate grave constructions such as headstones, mausoleums, above- and below-ground burials, and decorative elements such as sculptures and fountains may provide additional aquatic habitats unnoticed by cemetery workers compared to lawn or garden cemeteries that have more homogeneous grave styles (LMO, personal observation). Maintenance frequency in cemeteries is also important since garbage and discarded containers made of different materials often become aquatic habitats after rains. Interestingly, some materials commonly used in cemeteries' flowerpots (e.g. plastic) have been positively associated with the presence of immature mosquito stages [9], while others (e.g. copper) can reduce mosquito larvae survival [10].

Because they can provide a suitable environmental habitat for mosquitoes, cemeteries may play an important role in public health. Some medically relevant mosquito species developing in cemeteries are *Ae. aegypti*, *Ae. albopictus* (Skuse), *Culex quinquefasciatus* Say, *Cx. nigripalpus* (Theobald) and *Culex pipiens* species complex [1,3,4]. These mosquitoes are competent vectors of arboviral and parasitic disease agents such as dengue, Zika, chikungunya, yellow fever, West Nile virus, filariasis and Venezuelan Equine Encephalitis [11].

In Puerto Rico, *Ae. aegypti* is the primary vector of dengue, chikungunya and Zika viruses. The four serotypes of dengue have been endemic in the island since the 1980s [12], and over the last decade three major epidemics have occurred in 2010, 2012 and 2013 [13]. Monthly changes in dengue transmission are positively and significantly associated with monthly fluctuations in temperature and precipitation, and this association varies due to differences in local climate in Puerto Rico [14]. Chikungunya and Zika viruses emerged in Puerto Rico more recently and caused outbreaks in 2014 and 2016, respectively [15–17].

This investigation is the first report on the types of container aquatic habitats and mosquito species inhabiting them in 16 Puerto Rican cemeteries. The main objective was to make recommendations to cemetery managers and public health officials on mosquito and disease control.

METHODS AND MATERIALS

Cemetery inspections and aquatic habitats

Between June 2019 and February 2020, 16 cemeteries in southeast Puerto Rico were inspected for the presence of immature stages of mosquitoes in the municipalities of Caguas, Humacao, Yabucoa, Naguabo and Juncos (Figure 1; Table 1). Visited cemeteries were either private or public (managed by the municipality) and were in urbanised areas according to the United States Census Bureau (except for Oriente Memorial Park in Yabucoa, which is in an area classified as not urban). The inspections at each cemetery occurred on continuous days (except for weekends and holidays) and the duration depended on the size and density of the structures to be inspected: small cemeteries took 1 or 2 days to be fully inspected, while large cemeteries required several weeks (Table 4).

To determine the presence of aquatic container habitats with and without larvae and pupae of mosquitoes, we examined within the entire area of the cemeteries every single burial and structure. These included accumulations of water in tombs (either broken or cracked tombs being used, or open unused tombs), headstones, mausoleums, decorative elements (flowerpots, candleholders, fountains and sculptures), and other water-collecting containers that have the potential to become aquatic habitats of mosquitoes. We used maps to make sure that all areas of the cemetery and all structures were inspected. Containers and structures with water were checked for the presence of mosquito larvae or pupae using turkey basters, mosquito dippers, trays and flashlights. Data were recorded on tablets or cell phones using the ArcGis App Survey ‘123 (Survey 123 for ArcGIS 2019, Environmental Systems Research Institute (ESRI)). Data collected included container type, presence of water, larvae, pupae and its location. Containers were emptied or discarded, and when this was not possible, the granular water-dispersible bacterial larvicide *Bacillus thuringiensis israelensis* was applied (VectoBac® WG, Valent BioSciences, Osage, IA – one teaspoon per container). Once the containers were registered in the app, we used chalk to mark the structure as inspected. It was common to find miscellaneous objects during the inspections such as candles, cups, drums, jars, tarps, wheelbarrows, buckets, decorative water fountains, bromeliads, upside-down plastic tables, broken pipes, paint trays, food containers, bottles, discarded water containers, decorative ceramic and glass figures and cans. For practical reasons we considered these together as ‘other containers’ in our subsequent analysis. Containers were classified into two categories: negative aquatic habitats (containers containing water but no mosquito larvae or pupae) and positive aquatic habitats (containers with larvae or pupae). Dry containers were not recorded.

We classified cemeteries into three categories: lawn, cement and mixed. Lawn cemeteries were those where tombs were covered with grass and tombs were identified by a plate, usually of bronze or marble (Figure 2). In cement or concrete cemeteries tombs and surrounding structures were built with cement or concrete, with scarce vegetation; and mixed cemeteries were those with lawn and cement tomb areas (Figure 2). We estimated cemetery area using Google Earth Pro (version 7.3.3) and used cemetery areas to calculate the densities of positive, negative and total aquatic habitats of mosquitoes.

Rainfall data collection and analysis

We collected rainfall data (mm) to investigate their relationship with container abundance and positivity. Because the cemeteries were visited at different times (but each cemetery was inspected during continuous days – except during weekends or holidays), we calculated average daily precipitation (average of daily rainfall during the days of inspections and during the previous week). Rainfall data were obtained from NOAA (www.ncdc.noaa.gov) and 'Weather Underground' (www.wunderground.com) databases, and from meteorological stations placed and maintained by the Puerto Rico Vector Control Unit. We obtained data from the closest meteorological stations to each cemetery; the distance between these varied between 0.16 km (cemetery Ramon Delgado in Juncos) and 8 km (cemetery Valle de la Paz in Las Piedras). We could not obtain rainfall data for the 'Verde mar' nor 'Historico de Humacao' cemeteries. To determine if there was a relationship between rainfall data (mm) and container abundance and positivity, we conducted a linear regression analysis using GraphPad Prism version 9.1.2 for Windows (GraphPad Software, San Diego, California USA, www.graphpad.com).

Mosquito larval and pupal surveys

After the initial assessment of presence or absence of immatures (conducted between June 2019 and February 2020), we collected specimens to quantify and identify mosquito species in containers and other structures in December 2020 and January 2021. At each cemetery we examined all structures and used turkey basters, trays and flashlights to collect mosquito larvae and pupae. Sampled immatures were transported to the laboratory for posterior identification using taxonomical keys (CDC unpublished; [18–20]).

RESULTS

Of the 16 studied cemeteries, 13 were classified as cement, one as a lawn and two as mixed. We found 9984 containers with water in all cemeteries, of which 45.5% were positive for mosquito larvae or pupae. The most abundant aquatic habitats were flowerpots (negative: 50.3%; positive 42.2%), followed by 'other containers' (negative: 2.9%; positive 2.0%) and tombs (negative: 1.2%; positive 1.2%) (Table 2). In Borinquen Memorial I, Borinquen Memorial II, Verde Mar, Paz Christi and Memorial Park negative flowerpots were more abundant than positive flowerpots; in the Historico de Humacao, El Caimito, Historico de Las Piedras, Verde Paz, Municipal #1, #2 and #3 de Naguabo the opposite occurred, while in the rest of the cemeteries the proportions between positive and negative flowerpots were similar (Table 2). The lawn and two mixed cemeteries were among the group with a higher proportion of negative flowerpots.

The density of containers with water (# of aquatic habitats/m^2 of cemetery area) varied among cemeteries and was low overall (Table 3). The cemetery with the lowest density of positive aquatic habitats was ‘Borinquen Memorial I’ (lawn type cemetery, which also had a low density of negative aquatic habitats). In the two ‘mixed’ type cemeteries we found that ‘Paz Christi’ had the lowest container density and ‘Borinquen Memorial II’ the highest. Among cement type cemeteries, the lowest density of positive aquatic habitats was found in ‘El Caimito’, Las Piedras (this cemetery also had a low density of negative aquatic habitats) whereas ‘Oriente Memorial Park’, Yabucoa had the highest densities of positive and negative containers (Table 3).

Accumulated and average daily precipitation varied among locations and sampling dates, and most sampling occurred during rainy season months (Table 4). There was a positive and significant association between rainfall and number of containers with water ($Y = 0.0035 + 0.0043X$; $R^2 = 0.32$; $F_{1,12} = 5.69$; $p < 0.05$) (Figure 3).

We found immature stages of nine species of mosquitoes in 14 of the 16 cemeteries (Table 5). The cemeteries with the highest mosquito abundances were ‘Municipal #3 de Naguabo’ and ‘Municipal de Yabucoa’. No immature stages were found in Verde Mar nor Oriente Memorial. The most common and abundant species were *Ae. aegypti* and *Aedes mediovittatus* (Coquillett), which were present in 11 cemeteries and represented 84.9% of all the captured immatures (Table 5). Specifically, *Ae. aegypti* was present in 12 of the 14 cemeteries and was the most abundant species in 7 of these 14 cemeteries. *Culex quinquefasciatus* and *Cx. antillummagnorum* Dyar were present in 4 and 5 cemeteries and represented 3.1% and 9.8% of all collected immatures, respectively. The least common species were *Cx. secutor* Theobald (found in two cemeteries) and *Cx. habilitator* Dyar and Knab, *Anopheles albimanus* Weidemann, *Cx. bahamensis* Dyar & Knab *and Uranotaenia* sp. (found in one cemetery). These species represented 2.1% of all immatures collected. Flowerpots were the most common aquatic habitats used by mosquitoes. The species found in flowerpots were *Ae. aegypti*, *Ae. mediovittatus*, *Cx. quinquefasciatus*, *Cx. antillummagnorum and Uranotaenia* spp. while *Cx. bahamensis*, *Cx. habilitator* and *An. albimanus* were found in open tombs.

DISCUSSION

This is the first study describing the immature mosquito fauna and abundance of aquatic habitats in Puerto Rican cemeteries. We studied 16 cemeteries, 13 of which were classified as cement, one as a lawn and two as mixed. The most common aquatic habitat container registered in all inspected cemeteries was flowerpots, followed by a variety of discarded containers and open tombs. Generally, there was a positive relationship between density of containers with water and average daily precipitation per cemetery. We found a rich community of mosquito species developing in containers of the inspected cemeteries, where *Ae. aegypti* and *Ae. mediovittatus* were the most frequent and abundant species.

Flowerpots have been reported as the most abundant aquatic habitat in cemeteries in previous works conducted in Latin American countries (Abe et al., 21; Barrera, 1; [22]). What makes a container more or less likely to become an aquatic habitat for mosquitoes depends on several variables. According to Vezzani and Schweigmann [23] the type of material of the flowerpots is important in predicting the presence and the abundance of mosquito immatures. For example, González et al. [24] and Vezzani & Schweigmann [23] described higher abundances of immature mosquito stages in plastic flowerpots than in metallic or glass flowerpots. Eritja and Herreros [10] described how adding a piece of copper wire to flowerpots reduced *Ae. albopictus* larval and pupae productivity. Shading of containers [5], whether with natural or artificial flowers (in the case of flowerpots), and input of organic matter are important variables influencing the presence and the abundance of immature mosquitoes.

In a considerably smaller proportion than flowerpots (4.9% versus 92.5%, respectively) the second-most common type of aquatic habitats consisted of objects that accumulated water, such as candles in jars, cups, drums, jars, tarps, wheelbarrows, buckets, plants, decorative water fountains, bromeliads, upside-down plastic tables, broken pipes, paint trays, food containers, bottles, discarded water containers, cans and decorative ceramic and glass figures among others. Most of these objects appeared to be left by visitors and in some cases by personnel working in the cemeteries. Finally, the least common aquatic habitat of mosquitoes were tombs (2.4%) that either were opened for future burials or were broken or damaged. However, a visual assessment of immature productivity in tombs is difficult because mosquitoes could colonise inundated tombs through inapparent cracks or crevices. We recommend that cemetery personnel apply larvicides while the tombs are fixed or sealed.

Regarding the density of aquatic habitat per cemetery, we found that although densities observed in this study were variable, they were like those reported in other studies. Abe et al. (2015) reported densities ranging from 0.013 to 0.0235 aquatic habitats/m^2 in Trujillo, Venezuela; Barrera et al [5] estimated a density of 0.3 aquatic habitats/m^2 in the ‘Cementerio del Sur’ in Caracas, Venezuela; Vezzani et al [7] reported densities of 0.1499, 0.1958, 0.1440, 0.2120, 0.0179 aquatic habitats/m^2 in five Buenos Aires cemeteries; and Garcia-Rejon et al [25] obtained densities of 1.12 and 0.7 aquatic habitats/m^2 in Mexico. We also compared the density of aquatic habitats among different types of cemeteries. The lowest density was found in the only lawn cemetery examined, where tombs are underground and the lawn is regularly mowed. Cement-type cemeteries were more heterogeneous than lawn cemeteries, where we found more decorative structures and containers that accumulated water and became aquatic habitats for mosquitoes. Unfortunately, we were able to conduct inspections in only one lawn cemetery, so density between cemetery types could not be compared. An interesting observation was that the only sampled lawn cemetery (Borinquen Memorial I) had few positive aquatic habitats during both the container (August 2019) and immature survey (December 2020). This is a private cemetery with identical metallic flowerpots where the elevated water temperature may prevent mosquito oviposition and development.

Flowerpots were the most common aquatic habitats with the greatest richness of mosquito immature species. Compared to other studies, the richness of the mosquito community in

Puerto Rican cemeteries is high. For example, Devera et al [9] found only two mosquito genera: *Aedes* and *Culex* in Ciudad Bolívar, Venezuela; Castillo et al [22] reported only immature stages of *Ae. aegypti* in the three inspected cemeteries in Trujillo, Venezuela; Garcia-Rejon et al [25] found *Ae. aegypti*, *Cx. quinquefasciatus*, *C. nigripalpus* and *Cx. coronator* Dyar & Knab in Merida, Mexico; and Barrera et al [5] found *Ae. aegypti*, *Cx. quinquefasciatus*, *Cx. corniger* Theobald, *Cx. nigripalpus and Toxorhynchites theobaldi* (Dyar & Knab) in Caracas, Venezuela. The species richness found in this study is comparable to the richness described by Wilke et al [4], who found nine species in Miami, Florida: *Ae. aegypti*, *Ae. albopictus*, *Ae. triseriatus* (Say), *Cx. coronator*, *Cx. quinquefasciatus*, *Cx. interrogator* (Dyar & Knab), *Cx. nigripalpus*, *Wyeomyia mitchelli* (Theobald) *and Wy. vanduzeei* Dyar & Knab.

The two most abundant species in our study were *Ae. aegypti* and *Ae. mediovittatus*. We found *Ae. aegypti* (the most important vector of dengue, chikungunya and Zika virus in Puerto Rico) present in 12 of the 14 cemeteries, and it was the most abundant species in 7 of these 14 cemeteries. Remarkably, *Ae. aegypti* was the only species found in the only lawn type cemetery sampled (Borinquen Memorial I), while in the two mixed-type cemeteries (Borinquen Memorial II and Paz Christi) there were *Ae. aegypti*, *Ae. mediovittatus* and *Cx. antillummagnorum*. The productivity of *Ae. aegypti* in cemeteries has been described previously [5,21,22,25], but whether this productivity in cemeteries plays a role in dengue or Zika outbreaks is unknown. Garcia-Rejon et al [25] reported finding positive pools of *Ae. aegypti* females for dengue (DENV-1) and Zika virus in Merida, Yucatan, Mexico. Given that some of the inspected cemeteries in this study were surrounded by schools, rural communities and urban settlements, it would be important to know how frequently and how far *Ae. aegypti* is dispersing from these cemeteries into inhabited areas. *Ae. mediovittatus* was found in 11 out of the 14 cemeteries. This species was especially abundant in the ‘Cementerio municipal nuevo de Naguabo’. This cemetery is located outside the town and surrounded by vegetation. *Ae. mediovittatus* has been proven to be competent for dengue virus transmission in laboratory settings [26], so its public health relevance should be considered when planning and implementing vector control activities in cemeteries, especially in those cemeteries located in rural areas where arboreal vegetation is common (the native habitat of this tree-hole mosquito).

We expected to find a positive relationship between the number of containers with water or immature mosquitoes and accumulated precipitation, and we did. Even though a positive relationship between rainfall and abundance of immature mosquitoes was expected and has been repeatedly described in the literature, there are multiple additional complexities behind this relationship. The effect of precipitation and human activity on the abundance of *Ae. aegypti* immatures in cemeteries has been previously described by [8], who found that the presence of immature mosquito stages in cemeteries during the dry season was a consequence of the introduction of water and fresh flowers brought by visitors. In Puerto Rico, work by [6] has shown that most immatures sampled were produced in aquatic habitats whose water was managed by humans. So even though we were able to establish a positive relationship between rainfall and positive aquatic habitats in cemeteries (which means control activities should occur more frequently during raining months), there are likely other variables involved that need to be studied. Further studies should investigate the importance of seasonality as well as the influence of habitat variables, such as type of container material, amount of organic matter available in containers, whether the container is shaded, presence of predators and physicochemical properties of the water on mosquito productivity. Additionally, to better understand the relationship between these two variables, it would be ideal to collect meteorological data at each of the sampled cemeteries, instead of collecting these data from stations located kilometres away, as it was the case for cemetery Valle de la Paz in Las Piedras, which was 8 km from the closest meteorological stations.

Although Puerto Rico has environmental health regulations where flowerpots in municipal and private cemeteries must have drainage holes and be filled with sand [27], flowerpots still are the major source of mosquitoes. We observed cement flowerpots with artificial flowers and clogged drainages that were producing mosquitoes. Artificial flowers provide shade, reducing evaporation and hence contribute to immature mosquito productivity. We recommend conducting further studies to establish how frequently inspections should occur and reduce mosquito productivity by unclogging drainage holes and applying larvicides, since it has been demonstrated that the proportion of aquatic habitats producing mosquitoes changes according to seasons and human activities [5].

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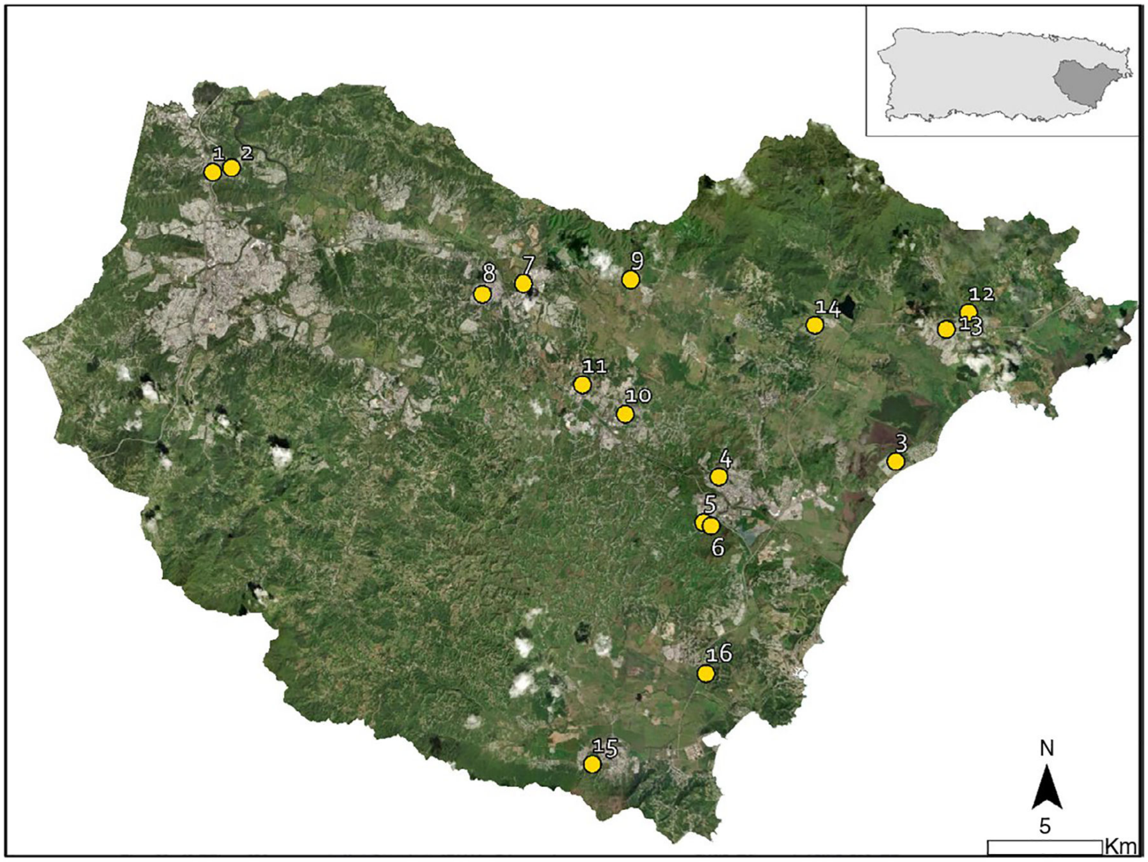

FIGURE 1.

Locations of the cemeteries inspected in southeastern Puerto Rico. 1. Borinquen Memorial I, 2. Borinquen Memorial II, 3. Verde Mar, 4. Historico de Humacao, 5. Paz Christi, 6. Municipal de Humacao, 7. Municipal Ramon Delgado, 8. La Inmaculada, 9. El Caimito, 10. Historico de Las Piedras, 11. Valle de Paz, 12. Municipal #3 de Naguabo, 13. Municipal #1 de Naguabo, 14. Municipal #2 de Naguabo, 15. Municipal de Yabucoa, 16. Oriente Memorial Park

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FIGURE 2.

Types of cemeteries visited in this study. Top: lawn cemetery; middle: cement cemetery; bottom: mixed cemetery

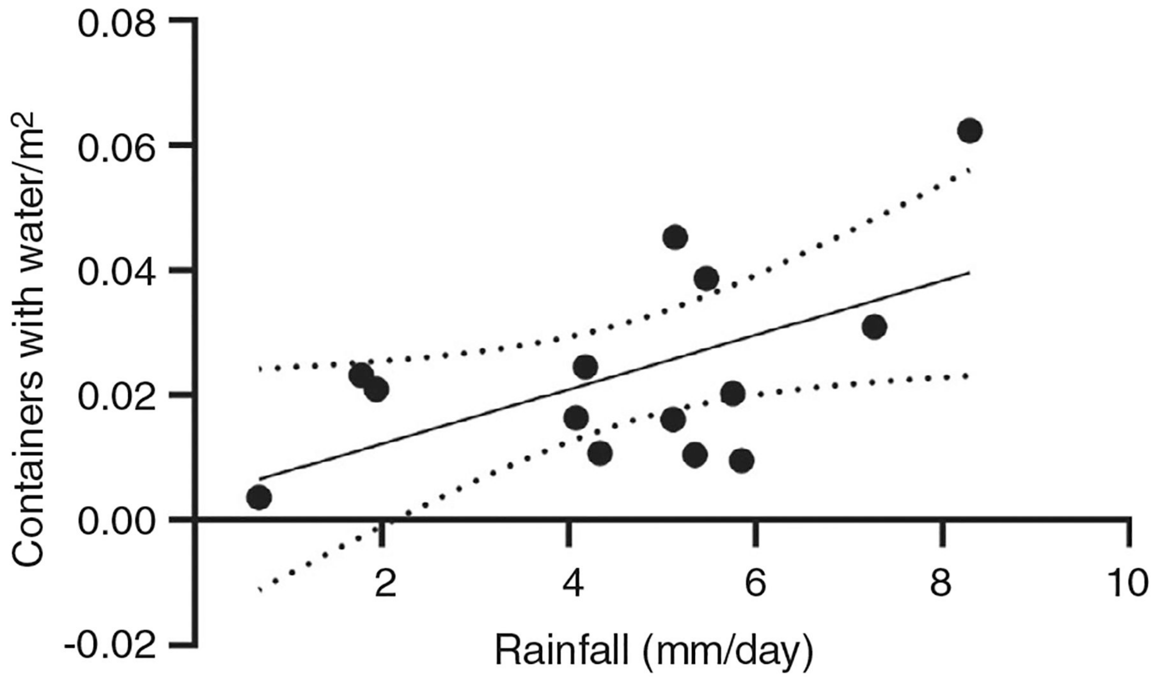

FIGURE 3.

Relationship between the density of containers with water (# of aquatic habitats/m^2) at all the sampled cemeteries and the average daily precipitation (mm) estimated for each cemetery ($Y = 0.0035 + 0.0043X$; $R^2 = 0.32$; $F_{1,12} = 5.69$; $p < 0.05$)

TABLE 1

Cemeteries inspected, locations and areas in southeast Puerto Rico

Cemetery	Municipality	Cemetery coordinates (decimal degrees)		Cemetery area (m^2)
Borinquen Memorial I	Caguas	18.27618	−66.0393	101107.2
Borinquen Memorial II	Caguas	18.27796	−66.0316	60632.8
Verde Mar	Humacao	18.1618	−65.7582	8528.1
Histórico de Humacao	Humacao	18.15597	−65.8312	30455
Paz Christi	Humacao	18.13803	−65.8375	89168.6
Municipal de Humacao	Humacao	18.13669	−65.8344	38789
Municipal Ramón Delgado	Juncos	18.23223	−65.9115	46603.7
La Inmaculada	Juncos	18.22818	−65.9284	16293.4
El Caimito	Las Piedras	18.23364	−65.8672	7685.6
Histórico de Las Piedras	Las Piedras	18.18081	−65.8696	12364.2
Valle de Paz	Las Piedras	18.19246	−65.8873	24355.2
Municipal #3 de Naguabo	Naguabo	18.22028	−65.7281	34239.7
Municipal #1 de Naguabo	Naguabo	18.21371	−65.7373	10443
Municipal #2 de Naguabo	Naguabo	18.21561	−65.7914	8181.8
Municipal de Yabucoa	Yabucoa	18.04289	−65.8836	64088.2
Oriente Memorial Park	Yabucoa	18.07849	−65.8368	15504.6

TABLE 2

Abundance of containers with water (negative and positive for the presence of mosquito immatures) in the studied cemeteries

	Flowerpots		Tombs		Other containers		
Cemetery	Negative	Positive	Negative	Positive	Negative	Positive	Total
Borinquen Memorial I	1048	2	2	0	4	0	1056
Borinquen Memorial II	492	131	15	18	2	2	660
Verde Mar	122	94	6	1	6	6	235
Histórico de Humacao	211	312	21	20	16	27	607
Paz Christi	246	50	5	4	3	0	308
Municipal de Humacao	707	749	15	16	12	12	1511
Municipal Ramón Delgado	178	191	1	1	30	42	443
La Inmaculada	41	44	0	1	1	10	97
El Caimito	19	123	0	3	3	5	153
Histórico de Las Piedras	35	245	3	9	1	6	299
Valle de Paz	260	408	5	16	6	9	704
Municipal #3 de Naguabo	203	408	6	10	11	24	662
Municipal #1 de Naguabo	37	113	7	5	9	3	174
Municipal #2 de Naguabo	56	71	4	2	2	3	138
Municipal de Yabucoa	1309	1237	15	13	186	55	2815
Oriente Memorial Park	65	39	14	0	4	0	122
Total	5029	4217	119	119	296	204	9984

TABLE 3

Densities of containers with water (# of aquatic habitats/m^2) at the 16 inspected cemeteries

Cemetery	Municipality	Positive	Negative	Total	Type of cemetery
Borinquen Memorial I	Caguas	1.978E-05	0.01040	0.01042	Lawn
Borinquen Memorial II	Caguas	0.00369	0.01245	0.01615	Mixed
Verde Mar	Humacao	0.01184	0.01571	0.02756	Cement
Histórico de Humacao	Humacao	0.01179	0.00755	0.01934	Cement
Paz Christi	Humacao	0.00183	0.00888	0.01071	Mixed
Municipal de Humacao	Humacao	0.02022	0.01848	0.03871	Cement
Municipal Ramón Delgado	Juncos	0.00506	0.00452	0.00958	Cement
La Inmaculada	Juncos	0.01368	0.01090	0.02458	Cement
El Caimito	Las Piedras	0.00078	0.00286	0.00364	Cement
Histórico de Las Piedras	Las Piedras	0.02079	0.00243	0.02321	Cement
Valle de Paz	Las Piedras	0.01910	0.01182	0.03092	Cement
Municipal #3 de Naguabo	Naguabo	0.01403	0.00695	0.02097	Cement
Municipal #1 de Naguabo	Naguabo	0.01149	0.00488	0.01637	Cement
Municipal #2 de Naguabo	Naguabo	0.01116	0.00911	0.02027	Cement
Municipal de Yabucoa	Yabucoa	0.02107	0.02426	0.04533	Cement
Oriente Memorial Park	Yabucoa	0.02251	0.03982	0.06233	Cement

TABLE 4

Average daily precipitation (mm) estimated from records from the closest meteorological stations to each cemetery during assessments of positive and negative containers with water

Cemetery	Municipality	Average daily precipitation (mm)	SE	Season	Inspection start date	Inspection end Date
Borinquen Memorial I	Caguas	5.36	2.57	Rainy	5-Aug-19	23-Aug-19
Borinquen Memorial II	Caguas	5.12	2.87	Rainy	26-Aug-19	9-Sep-19
Verde Mar	Humacao	NA	NA	Rainy	11-Jul-19	16-Jul-19
Histórico de Humacao	Humacao	NA	NA	Rainy	17-Jul-19	2-Aug-19
Paz Christi	Humacao	4.34	1.45	Rainy	10-Sep-19	17-Sep-19
Municipal de Humacao	Humacao	5.47	1.39	Rainy	18-Sep-19	28-Oct-19
Municipal Ramón Delgado	Juncos	20.3	0.20	Dry	15-Jan-20	31-Jan-20
La Inmaculada	Juncos	4.18	2.54	Dry	3-Feb-20	4-Feb-20
El Caimito	Las Piedras	0.69	0.28	Rainy	12-Jun-19	14-Jun-19
Histórico de Las Piedras	Las Piedras	1.78	0.98	Rainy	17-Jun-19	20-Jun-19
Valle de Paz	Las Piedras	7.27	2.93	Rainy	28-May-19	11-Jun-19
Municipal #3 de Naguabo	Naguabo	1.94	1.07	Rainy	21-Jun-19	1-Jul-19
Municipal #1 de Naguabo	Naguabo	2.31	1.65	Rainy	2-Jul-19	8-Jul-19
Municipal #2 de Naguabo	Naguabo	5.76	2.37	Rainy	9-Jul-19	10-Jul-19
Municipal de Yabucoa	Yabucoa	5.14	0.81	Rainy	29-Oct-19	13-Jan-20
Oriente Memorial Park	Yabucoa	8.29	2.96	Dry	14-Jan-20	14-Jan-20

Note: We could not obtain rainfall data for the ‘Verde mar’ nor ‘Historico de Humacao’ cemeteries.

TABLE 5

List of species and abundances of mosquito immature stages found in the investigated cemeteries (December 2020–February 2021)

Cemetery	Aquatic habitat	Mosquito species	Number of immatures
Borinquen Memorial I	Flowerpots	*Aedes aegypti*	5
Borinquen Memorial II	Pail	*Aedes aegypti*	3
	Flowerpots	*Aedes aegypti*	98
		Aedes mediovittatus	5
	Pail lid	*Aedes aegypti*	1
		Culex antillummagnorum	3
		Aedes mediovittatus	60
	Cup	*Aedes aegypti*	2
El Caimito	Flowerpots	*Aedes mediovittatus*	9
		Aedes aegypti	4
		Culex quinquefasciatus	28
Historico de Las Piedras	Flowerpots	*Aedes aegypti*	3
		Aedes mediovittatus	73
		Culex antillummagnorum	2
		Culex quinquefasciatus	1
Historico de Humacao	Flowerpots	*Aedes aegypti*	70
		Aedes mediovittatus	8
Paz Christi	Flowerpots	*Aedes aegypti*	8
		Aedes mediovittatus	1
Municipal de Yabucoa	Concrete box	*Aedes mediovittatus*	27
	Flowerpots	*Aedes aegypti*	96
		Aedes mediovittatus	87
	Tomb	*Culex bahamensis*	3
		Culex habilitator	6
		Culex secutor	1
		Culex quinquefasciatus	17
	Plant pot	*Aedes aegypti*	2
Municipal de Humacao	Flowerpots	*Aedes aegypti*	16

Cemetery	Aquatic habitat	Mosquito species	Number of immatures
		Aedes mediovittatus	4
		Culex antillummagnorum	65
	Pail	*Culex antillummagnorum*	23
		Culex quinquefasciatus	10
		Culex secutor	25
Municipal #3 de Naguabo (Nuevo)	Flowerpots	*Aedes aegypti*	50
		Aedes mediovittatus	678
		Culex antillummagnorum	86
		Culex quinquefasciatus	1
Municipal #2 Naguabo (Rio Blanco)	Altar	*Aedes mediovittatus*	5
		Aedes aegypti	9
	Flowerpots	*Aedes aegypti*	103
Municipal #1 Naguabo (Viejo)	Flowerpots	*Aedes aegypti*	67
		Aedes mediovittatus	4
Municipal Ramon Delgado	Flowerpots	*Aedes mediovittatus*	24
La Inmaculada	Flowerpots	*Aedes aegypti*	27
		Uranotaenia spp	2
Valle de Paz	Tomb	*Anopheles albimanus*	1