Ecological characterization of *Aedes aegypti* larval habitats (Diptera: Culicidae) in artificial water containers in Girardot, Colombia

Diana C. Garcia-Sánchez[1, 2], Gabriel A. Pinilla[2]✉, and Juliana Quintero[1]

[1]*Eje de Salud Pública, Fundación Santa Fe de Bogotá, Bogotá, Colombia*
[2]*Facultad de Ciencias, Departamento de Biología, Universidad Nacional de Colombia, Bogotá, Colombia, gapinillaa@unal.edu.co*

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ABSTRACT: The establishment of habitats for immature *Ae. aegypti* is regulated by biotic and abiotic factors and interactions between these factors. This study aimed to determine the effects of physico-chemical variables and planktonic algae on immature *Ae. aegypti* habitats in 101 water tanks (50 of them containing *Ae. aegypti* pupae and/or larvae) in Girardot, Colombia. Physical data were collected from the water tanks (volume, capacity, material, detritus, and location), along with the physico-chemical variables (temperature, pH, conductivity, redox potential, dissolved oxygen, percentage of oxygen saturation, nitrates, nitrites, and orthophosphates). The richness and abundance of the planktonic organisms were also measured. A chi-square test showed that the occurrence of detritus was greater and the container volume was smaller in the tanks that were positive for larvae. Only Cyanobacteria had a positive correlation with the abundance of immature-stage *Ae. aegypti*. The results could be important for understanding the vector ecology and envisaging its probable control in the domestic water tanks of Girardot. ***Journal of Vector Ecology* 42 (2): 289-297. 2017.**

Keyword Index: Yellow fever mosquito, larval ecology, physico-chemical variables, plankton, concrete water tanks.

INTRODUCTION

Aedes aegypti (L) (Diptera: Culicidae) is one of the main vectors of dengue, chikungunya, and Zika viruses, especially in tropical and subtropical regions. Its biological success may be attributed to its adaptability to urban ecosystems. In fact, it has been determined that the presence of humans in habitats is significantly associated with its invasive status (Juliano and Lounibos 2005). Water reservoirs located inside households are a favorite breeding place for *Ae. aegypti* (Koenraadt et al. 2008). Girardot, a city in central Colombia, has been categorized as hyperendemic for dengue (Padilla et al. 2012). Alcalá et al. (2015) determined that more than 93% of the pupae came from deposits associated with households, including concrete water tanks used for washing and low height tanks located on the floor with a capacity greater than 20 liters.

Several different environmental studies have been conducted on *Ae. aegypti*, including the characterization of the most productive containers, which varies among regions (Focks and Barrera 2006, Romero-Vivas et al. 2007), as well as variance in the genetic analysis of bacterial communities in mosquito container habitats (Ponnusamy et al. 2008) and the modeling of its abundance in ground containers and rainwater tanks under climate change scenarios (Kearney et al. 2009). In relation to food sources, some studies have shown that inputs of detritus are one of the principal energy sources for *Ae. aegypti* larvae in aquatic habitats. It has been revealed that bacterial abundance was reduced by *Ae. triseriatus* larval feeding (Walker et al. 1991). Cyanobacteria seem to have an important role in the diet, as stomach contents of *Ae. aegypti* have high concentrations of these phytoplankton microorganisms. Besides fine detritus, bacteria, and algae, other food sources for *Aedes* species in breeding reservoirs are fungi and protozoans (Merritt et al. 1992, Kaufman et al. 2002, Yee 2016). Otherwise, few studies have attempted to compare the ecological variables that can interfere with the establishment of habitats for the immature stages of this species (Dom et al. 2013). In other culicid species some ecological relationships have been found for *Culex* species. Gardner et al. (2013) proposed ammonia, nitrates, and an area of small shrubs that surround the urban catch basins of Chicago as positive predictors of high larval abundance, whereas pH and area of flowering shrubs seemed to be negatively correlated with larval profusion in ponds located in grassland habitats of Germany. Kroeger et al. (2014) found negative interactions between larvae of *Ae. vexans* and *Culex pipiens* with predatory Cyclopoida.

Studying the ecology of *Ae. aegypti* is of great importance because the selection of a place to lay eggs is an essential component in the life history of mosquitoes (Bentley and Day 1989). Thus, the biotic and abiotic factors that influence the population dynamics of larvae and pupae may determine many characteristics in the life cycle of adult individuals such as longevity, fecundity, body size, and immunological functions. Consequently, these factors can have a direct impact on disease transmission (Telang et al. 2011). Results from this type of study may be very useful as tools to predict the abundance of adults, the transmitters of the disease. In addition, this information could provide input for the definition of more efficient vector control strategies.

Since planktonic algae could be a food source for *Ae. aegypti* (Marten 1986, Thiery et al. 1991, Merritt et al. 1992, Rettich et al. 2001, Bond et al. 2005), this study aimed to identify the plankton characteristics and physico-chemical variables that determine the establishment of habitats for the immature-stages of *Ae. aegypti*. We tested the hypothesis that the density of immature stages of *Ae. aegypti* is directly related to the abundance of microalgae in water tanks because they could be consumed by the larval stages of the vector.

MATERIALS AND METHODS

Study site and study design

The municipality of Girardot, Colombia (4°18' 18" N, 74°48' 06" W) is located in the eastern Andes Mountain Range. It has an area of 129 km^2 and an elevation of 289 m. The municipality shares borders with the Magdalena River to the south and with the Bogotá River to the east. It is 120 km southwest from the Colombian capital, Bogotá. Its average annual temperature is 33.3° C and it has a mean relative humidity of 66% and an average annual rainfall of 821 mm (Alcaldía de Girardot 2015). This municipality has a bimodal climatic pattern with two rainy seasons (March-May and October-November). Its ecological, geographical, and demographic characteristics make this town a hyperendemic area for dengue, with almost 50% of the cases of this disease in the central-west region of the country (Padilla et al. 2012). In 2015 the cases of dengue and chikungunya in Girardot were the highest for the Department of Cundinamarca (503 and 8,905, respectively). Moreover, 1,936 cases of Zika virus were reported in Girardot in the last quarter of 2015 (Rojas et al. 2016).

Study tanks were selected from a group of 2,000 households that had previously taken part in the "Multi-country study of Dengue and Chagas in Latin America and the Caribbean from an Ecological, Biological and Social Approach" (Quintero et al. 2014), a fact that facilitated access to those houses. The main selection criteria were that the water tanks were currently in use for washing clothes, that they were constructed with hard materials (locally known as "albercas"), and that they were located under a roof (shaded) but with availability of indirect sunlight.

The 2,000 households selected for the study were distributed randomly in different parts of the city, and of these, 50 water tanks with the presence of immature-stage *Ae. aegypti* were selected and classified as positive, along with 51 containers without any stage *Ae. aegypti*, that were referred to as negative. Only the containers categorized as "albercas" were considered, because a previous study (Alcalá et al. 2015) showed that in Girardot, these reservoirs contribute more than 90% of the productivity of immature *Ae. aegypti* (larvae and pupae). The volume of water stored in the tanks is constantly restored and only once or twice a year they are washed (Alcalá et al. 2015).

Entomological inspection and physical data of the tanks

From January to May, 2014, entomological inspections were carried out in the 101 chosen reservoirs; each container was sampled only once. For all of the containers, information on the capacity, emptying frequency, volume of stored water, type of detritus (leaves, moss, mud, seeds, fruit), extent of shading, and building material was collected. In the positive water tanks, the abundance of larvae of all instars was estimated and an indirect estimation of the number of pupae was made (Romero-Vivas et al. 2007). The collected larvae and pupae were allowed to emerge and turn into adults and the corresponding taxonomic identification was made (Rueda 2004).

Physico-chemical variables

The temperature, pH, dissolved oxygen, conductivity, and redox potential were measured in situ in every water container with a multiparametric probe (Hq40d, HACH, Loveland, CO). The oxygen saturation percentage was calculated, taking into account the coefficient of solubility for Girardot's atmospheric pressure (0.965) and the solubility of oxygen (7.827 mg/liter) at the average temperature of the reservoirs (28° C). For the nutrients, a sample of 100 ml of water from each reservoir was preserved at 4° C for analysis of nitrate, nitrite, and phosphate concentrations. These samples were filtered and analyzed at the Limnology Laboratory of the Department of Biology at the Universidad Nacional de Colombia using a spectrophotometer (DR2800, HACH, Loveland, CO). In all cases, standardized methodologies for water analysis were followed (APHA et al. 2012).

Plankton sampling

For the plankton examination, 10 liters of water were filtered using a plankton net sampler, 23 μm pores, 20 cm in diameter, and 40 cm in length. Filtration through a net enabled concentration of the 10 liter sample into a final volume of 100 ml. The samples were preserved with a Transeau solution (distilled water-ethylic alcohol-formaldehyde, 6:3:1). At the Limnology Laboratory, subsamples of 30 ml were placed in sedimentation chambers (2 h per ml). The taxonomic determination and quantification of individuals were done with an inverted microscope (UB2001-TR, Advanced Optical, China). The counts were done at 40X magnification. The phytoplankton was characterized to genus using different taxonomic keys (Wehr and Sheath 2003, Bicudo and Menezes 2006, Bellinger and Sigee 2010).

Statistical and numerical analysis

The descriptive analysis included the Shapiro-Wilk normality test, variance comparison tests, and t-tests to evaluate the differences between the averages of each variable. A Principal Component Analysis (PCA) was used to explore which variables had a greater weight on the ordination of the reservoirs. Variables were normalized using division by their standard deviations and the PCA scores were computed utilizing vector products with the original data; the factors whose eigenvalues were greater than unity were extracted. The possible associations between the physico-chemical environment and the abundance of immature-stage *Ae. aegypti* were evaluated with Pearson correlations, with a previous confirmation of normality. A chi-square test was used to analyze the differences between the categorical variables of the two container groups (positive and negative reservoirs). The significance of association between variables was calculated, with p values from the chi-squared distribution and from a permutation test with 9,999 replicates. For phytoplankton, the abundance (organism L^{-1}) was estimated for each taxonomic class and the Shannon diversity (H) and Simpson dominance (D) indices were calculated. The associations among the different classes of phytoplankton and the physico-chemical variables were assessed with a Canonical Correspondence Analysis (CCA), in which environmental variables were plotted as correlations with site scores. All of the analyses were done with STATA 13.0 (Stata Corporation 2013), and PAST 3.06 (Hammer et al. 2001).

RESULTS

Physical characteristics of the tanks

Cement was the predominant material of the tanks (63%). Other materials included bricks, tiles, and stones. The presence of detritus (Figure 1) showed significant differences between the positive and negative mosquito groups of tanks ($p = 0.02$). 63% (32). The water tanks without immature stages had no detritus in the water, whereas 50% (25) of the positive tanks contained mud as the predominant detritus. The water tank capacity was also significantly different between positive and negative tanks with mosquitos ($p = 0.0008$); for the negative reservoirs, the average was 1.025 liters and, for the positive group it was nearly half that figure (675 liters). The water volume stored at the time of sampling was also different ($p = 0.0056$) although the percentage with respect to the storage capacity was similar in both groups of containers (negative 79%, average 807 liters and positive 78%, average 526 liters). The variance comparison test between the tank groups was significant for the water volume (f = 2.83; df = 50.4; p (F > f) = 0.0004). All of the larvae and pupae found in the positive water tanks were identified as *Ae. aegypti*.

Water physico-chemical data

Only phosphates showed statistical differences between the negative and positive water tanks (Table 1). All other variables were very similar in each group of tanks. In general, the water in these reservoirs was oxygenated, warm, neutral and with low nitrogen, but high phosphorus. No correlation was found between the physico-chemical variables and the volume of stored water. In the PCA, redox potential and dissolved oxygen were more influential in the negative mosquito group of tanks (right side of Figure 2). The electrical conductivity was remarkably important to the positive water tanks (left side of Figure 2). The correlation of this variable with component 1 was near -1 ($p = -0.027$). The major influence of the redox potential on the negative tanks was consistent with the slightly higher concentration of oxygen in these tanks. Moreover, the PCA showed a certain positive association between the tanks with the presence of the vector and orthophosphate concentration (bottom of Figure 2). The Pearson correlation of this nutrient with axis 2 was meaningful ($p = 0.045$).

Phytoplankton

In both groups of water containers, three main taxa were found: Bacillariophyceae, Chlorophyceae, and Cyanobacteria. No significant differences were found with the T test in the average of organism abundance from any taxon between the positive and negative water tanks (Bacillariophyceae $p = 0.57$, Chlorophyceae $p = 0.97$, and Cyanobacteria $p = 0.91$) (Table 1), but the variance test of abundance was significantly different for Cyanobacteria (f = 0.46; df = 50.4; p (F < f) = 0.0077) between the two groups of reservoirs with a confidence interval of 95%. The CCA showed some possible associations between Chlorophyceae and the redox potential and temperature; between Bacillariophyceae and the pH; and between Cyanobacteria and the percentage of oxygen saturation and phosphates (Figure 3). A greater abundance of Bacillariophyceae and Chlorophyceae was found in the negative

Table 1. Statistic comparison (T-test) of the physical, physico-chemical, and biotic variables measured in this study (sd = standard deviation).

	Variables measured	Negative group (n:51)	Positive group (n:50)	p value
		Mean (sd)	Mean (sd)	
Physical variables	Detritus occurrence	32	25	0.02*
	Water content (L)	807.3 (604.5)	526.49 (358.96)	0.0056*
	Capacity (L)	1024.8 (571.5)	675.16 (433.83)	0.0008*
Physico-chemical variables	Dissolved Oxygen (mg/L)	6.85 (1.35)	6.42(1.41)	0.1255
	Water temperature (°C)	28.5 (0.82)	28.48 (1.20)	0.9936
	pH	7.32 (0.35)	7.33 (0.29)	0.9245
	Conductivity (µS/cm)	134.7 (15.98)	132.97 (18.56)	0.6186
	Redox (mV)	374.1 (153.0)	352.86 (164.36)	0.5022
Nutrients	Nitrates (mg/L)	0.85 (0 .95)	0.57 (0.83)	0.1236
	Phosphates (mg/L)	0.18 (0.33)	0.33 (0.38)	0.0478*
	Nitrites (mg/L)	0.01 (0.01)	0.01 (0.01)	0.9967
Biological component	Bacillariophyceae (inds/L)	32500 (4651.2)	24200 (3008.2)	0.5714
	Chlorophyceae (inds/L)	12800 (1946.5)	8900 (1457.9)	0.9738
	Cyanobacteria (inds/L)	42000 (7220.7)	51600 (6467.8)	0.9155
	Total algae (inds/L)	87300 (4982.98)	84700 (4563.3)	0.8215
	Immature stages (pupae and/or larvae)	NA	83.4 (72.2)	NA

An asterisk indicates significant differences in the t-test ($p < 0.05$) between the groups.

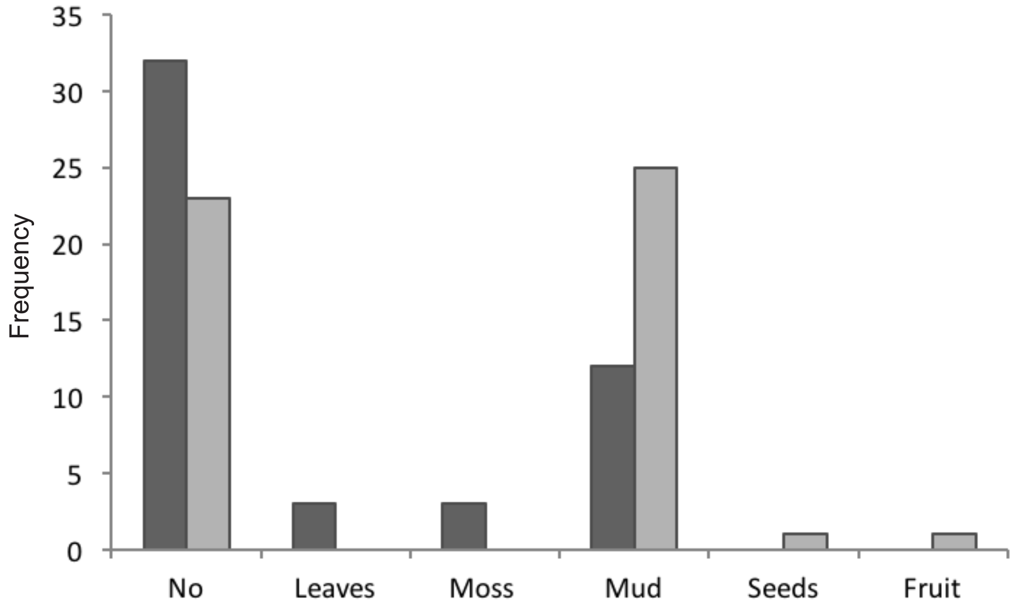

Figure 1. Variation in the type of detritus found in the negative tanks (dark gray) and the water tanks making up the larval habitats (light grey). No = without detritus.

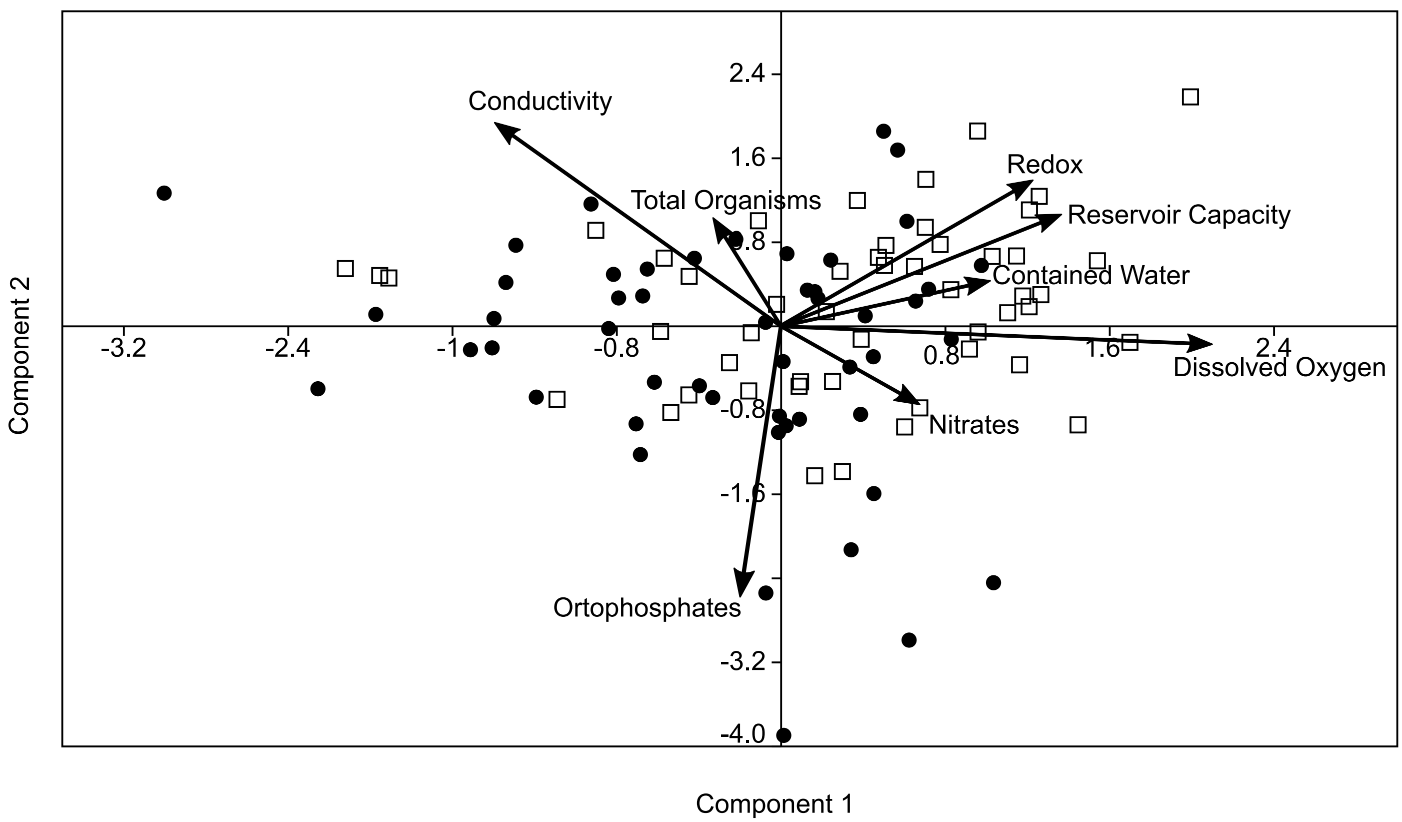

Figure 2. Biplot of the PCA. Physical and chemical variables are represented by the vectors. Black circles correspond to positive water tanks (with *Ae. aegypti* presence) and empty squares to negative water tanks (without *Ae. aegypti*). The explanation percentage for component 1 was 31.8% and, for component 2, it was 22.7%. As the vectors get closer to the axes, their ability increases to explain the ordination. Likewise, the proximity of the points (representing the positive and negative tanks) to the vectors indicates a greater influence of these variables on the corresponding groups of reservoirs.

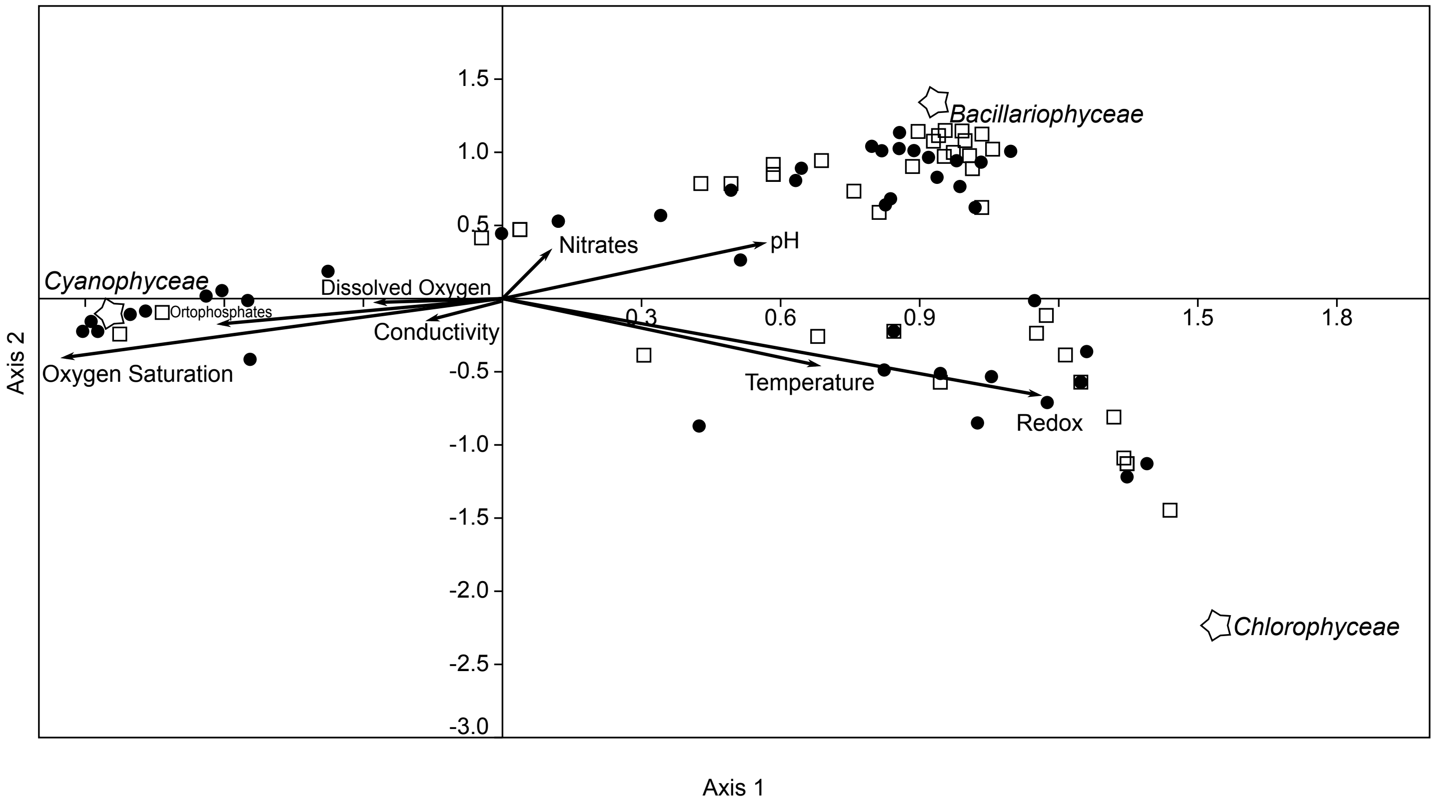

Figure 3. Triplot of the CCA for the physico-chemical variables (vectors) and the taxa of algae (stars) found in the water tanks that were positive (black circles) or negative (empty squares) for *Ae. aegypti*. The explanatory percentage for axis 1 was 94.87% and, for axis 2, it was 5.12%. The oxygen saturation and orthophosphate variables are remarkably close to the Cyanobacteria, especially in positive tanks. Temperature and redox potential are associated with Chlorophyceae, mostly in the negative tanks, while Bacillariophyceae algae appear to be associated with pH, both in the positive and negative reservoirs.

tanks (Figure 4A); however, for both groups, the Cyanobacteria were more abundant (Figure 4A). Dominance was greater for Cyanobacteria in the negative mosquito group of tanks (D = 0.43), whereas the Chlorophyceae were dominant in the larval habitats (D = 0.34) (Figure 4B). The diversity of *Bacillariophyceae* was higher in the *Ae. aegypti* larval habitats (H = 1.69) and Chlorophyceae diversity was greater in the water tanks with no immature stages (H = 1.60) (Figure 4C). With a degree of confidence of 0.05%, the Pearson correlations showed that only Cyanobacteria were positively related to the abundance of immature-stage *Ae. aegypti* (r = 0.26 > r crit 0.05, n = 50). For the other two taxa, there were no correlations (Bacillariophyceae r = -0.0714 and Chlorophyceae r = -0.0064).

The group of larval habitats displayed a higher number of algae genera with respect to the negative mosquito group of tanks (50 vs 44). In the first group, the more abundant genera were, from highest to lowest quantity, *Oscillatoria*, *Dactylococcopsis*, *Nostoc*, *Synedra*, *Scenedesmus*, *Pinnularia*, *Cymbella*, *Meridium*, *Navicula,* and *Dictyosphaerium*, while in the second group the representative genera were *Oscillatoria*, *Synedra*, *Aphanothece*, *Scenedesmus*, *Cymbella*, *Pinnularia*, *Euglena,* and *Anabaena*. The *Oscillatoria*, belonging to Cyanobacteria, had the greatest abundance but this only appeared in a few water tanks in each group (n ≤ 4). *Nostoc* was found only in the positive tanks, where it was one of the more representative organisms. In both groups, there were water containers without phytoplankton organisms (positive 5, negative 6). The only zooplankters found were some rotifers, in very small numbers and in only a few samples from both groups of containers. For this reason, they were not considered for analysis.

DISCUSSION

The establishment of an appropriate habitat for immature of *Ae. aegypti* is regulated by biotic and abiotic factors and interactions between them. Detritus is a determining element in the provision of energy for immature-stage *Ae. aegypti* because it is the primary source of essential nutrients that support the trophic chain of these habitats (Yee and Juliano 2006). Thus, the absence of detritus may limit the establishment of microhabitats since it has been found that dietary restrictions affect survival in a density-dependent way and additionally may determine longevity in adult stages (Focks and Barrera 2006, Joy et al. 2010). In Girardot, a recent study of larval productivity revealed that 92% of all containers with pupae had detritus (Alcalá et al. 2015). It is possible that our larval habitats without detritus, but with the presence of vectors, may have contained particles of fine and dissolved organic matter that did not settle but remained in the water column and were not perceivable to the naked eye. This fact means that there may have been some nutritional sources that sustained the immature stages of *Ae. aegypti* in these no-detritus tanks. More studies are needed in this area to include the assessment of fine particulate and dissolved organic matter.

The percentage of stored water was similar for both groups of reservoirs, but the tanks of the negative mosquito group had

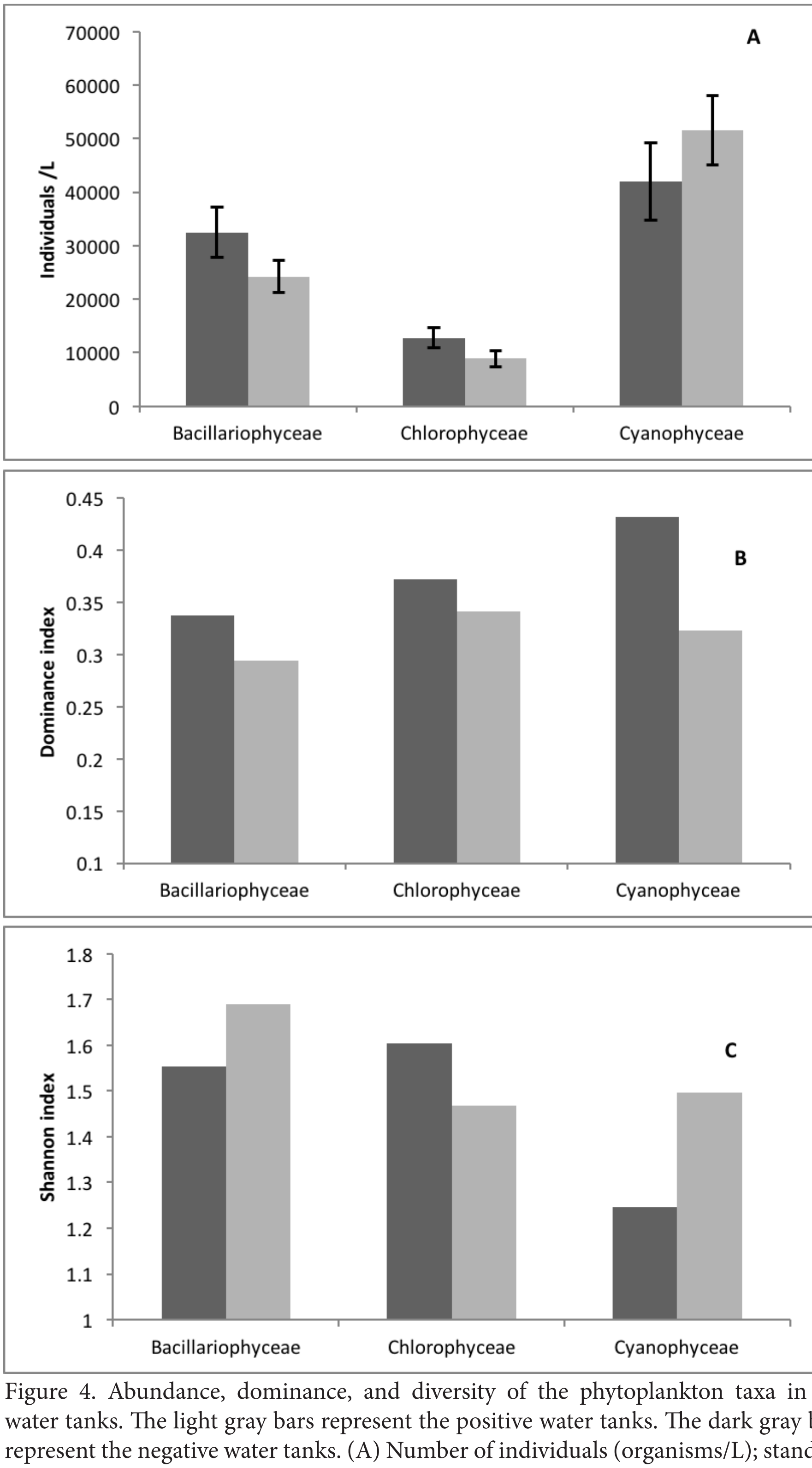

Figure 4. Abundance, dominance, and diversity of the phytoplankton taxa in the water tanks. The light gray bars represent the positive water tanks. The dark gray bars represent the negative water tanks. (A) Number of individuals (organisms/L); standard error is shown. (B) D-Simpson dominance. (C) Shannon's diversity (nats). In B and C, the data correspond to unique values calculated for all water tanks of each category (positive and negative), and, therefore, the standard error cannot be computed.

a much larger capacity. Of the several variables and interactions that could affect the establishment of larval habitat, the influence of tank water volume seems to be important. Prior studies have found that water volume is a determining factor in the selection of a laying site and in the larval development of other mosquito species, such as *Anopheles annulipes* (Mokany and Mokany 2006). This parameter should be analyzed in more detail in forthcoming studies, but nevertheless, the results found in Girardot seem to indicate that *Ae. aegypti* prefers tanks with a smaller volume, perhaps because dissolved and suspended solids are more concentrated, a fact that would be reflected in larger relative detritus concentration. Some authors have shown that most *Aedes* species (e.g., *Ae. aegypti*) are found in smaller water containers because they are preferred by females for oviposition (Sunahara et al. 2002, Dom et al. 2013). Therefore, it should not be unusual that smaller tanks contain *Ae. aegypti* populations. Thus, in addition to detritus, it is likely that females may be responding to the size of the reservoir surface area.

In both groups of water tanks, the temperature was within the optimal range for the larval development of *Ae. aegypti* (from 24 to 35° C). The pH levels were also within the normal limits for drinking water (6.5-8). The similarity of the average for this variable between the positive and negative water tanks confirms previous results in which pH levels had very little effect on the establishment of larval habitats (Stein et al. 2011).

The PCA indicated that conductivity may be an important variable in the environment of a larval habitat. According to the t-test (Table 1), this variable did not show variation between the container groups, but noticeable variations occurred within each group. In the set of positive water containers, the conductivity varied between 50 and 180 $\mu S\ cm^{-1}$, while in the negative mosquito group of tanks the variation was between 107 and 184 $\mu S\ cm^{-1}$. Other studies have shown the incidence of this parameter in larval abundance of anophelines, even more so than any other physico-chemical variable (Edillo et al. 2006, Yee and Juliano 2007). For *Ae. albopictus*, Carvajal et al. (2009) found that conductivity of the species-specific reservoirs was different with respect to the habitats of other species.

The dissolved oxygen measured in the Girardot water tanks is not a limiting factor for *Aedes* sp. High concentrations of this gas may be negatively correlated with the presence of larvae (Mokany and Mokany 2006, Tuten 2011). The similarity in the dissolved oxygen of the positive and negative containers is explained mainly by the location of all reservoirs under the same environmental conditions of temperature and altitude. Nevertheless, slightly lower values of this element in the larval habitats may have corresponded to its intake by phytoplankton and heterotrophic bacteria (not determined in this study).

In controlled experiments, Reiskind et al. (2004) observed that enrichment with nutrients increases the larval abundance of *Culex restuans*. However, larval survival requirements may vary between genera (Johnson et al. 2010). The concentration of orthophosphates was significantly higher in the positive water tanks, but it was high for both groups, as compared with the standardized values for hypereutrophic waters (0.1 mg/liter, Vollenweider and Kerekes 1982). This may have been due to allochthonous inputs of this nutrient from human activities, such as the use of detergents for washing clothes and the introduction of organic matter from the frequent use of water from kitchen chores (García-Betancourt et al. 2015). Previously, the importance of these additions as promoters in the increase of primary productivity in artificial habitats was established (Kesavaraju et al. 2007). Thus, the larger concentration of phosphorus in the positive group can explain the variations in the number of phytoplanktonic individuals, particularly Cyanobacteria, that are increasingly abundant in systems with high phosphate concentrations (Roldán and Ramírez 2008). Therefore, phosphorus could influence the establishment of larval habitats because it promotes the growth of algae and bacteria that can serve as a food source for the vector's immature forms. The larger amount of muddy detritus found in the positive group may have increased phosphates, which would probably account for the differences in the concentration of this nutrient for the two groups of water tanks. The nitrogen concentration for both sets of "albercas" was within the ranges of oligotrophic systems (Esteves 1998). For the positive group, fewer nitrates were found, a fact that may have been due to the consumption of this nutrient by a larger number of microalgae (Rejmánková et al. 1993, Roldán and Ramírez 2008). In addition, in aquatic systems, nitrogen is assimilated by organisms more efficiently than phosphate (Hejzlar et al. 2009). High content of phosphate combined with low availability of inorganic nitrogen is favorable for growth of nitrogen-fixing cyanobacteria.

A larger number of microalgae (mainly Cyanobacteria) in the larval habitats may indicate a larger nutritional supply for immature-stage *Ae. aegypti*. A characterization of the stomach contents of a mosquito population of this species revealed that Cyanobacteria are more frequent than other algal components (Ulloa-García unpublished data), a fact that suggests the importance of this group of algae in the positive reservoirs. Nevertheless, it should not be ignored that other studies have shown that the presence of algae in the gut of *Aedes* and other mosquitoes larvae does not necessarily imply that they are digested; on the contrary, some algae taxa can have negative effects (Marten 1986). In spite of that investigation, the importance of algae in the establishment of larval habitats has been demonstrated in previous studies with other mosquitoes, in which these phototrophic organisms had strong connections with the larvae (Thiery et al. 1991, Vazquez-Martinez et al. 2002). Apparently, the presence and abundance of Cyanobacteria could be a factor that would make it possible to predict the establishment of a larval habitat with a probability greater than 75% in anophelines (Rejmánková et al. 1993). In another study on reservoirs with *Ae. albopictus*, the relative abundance of Cyanobacteria was higher than that of other microalgae (Carvajal et al. 2009). Wang et al. (2011) and Minard et al. (2013) have suggested that Cyanobacteria are associated with the abundance of immature mosquitoes because they are part of their mid-gut flora and, therefore, these algae are not necessarily an independent indicator of habitat suitability. This controversy could be solved with studies on stable isotopes that show if Cyanobacteria are indeed assimilated by the larvae, but independently of the results, clearly, this group of algae seems to have a close relationship with the presence and abundance of these vectors.

According to the proposed hypothesis, in the water reservoirs of the city of Girardot, the Cyanobacteria seem to be important as a possible source of food for the larvae of *Ae. aegypti*. This result may provide new insight and increase our understanding of the factors that affect the local mosquito populations. For example, controlling algal growth and reducing detritus inputs could be additional measures of the vector control. Further research should be considered on the influence of human activities on these artificial breeding sites, because this interaction could directly affect some of these discussed variables through, for example, the addition of organic matter and nutrients.

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