A decade of arbovirus emergence in the temperate southern cone of South America: dengue, *Aedes aegypti* and climate dynamics in Córdoba, Argentina

Elizabet L. Estallo[1*¶], Rachel Sippy[2,3¶], Anna M. Stewart-Ibarra[2,4¶], Marta G. Grech[5], Elisabet M. Benitez[1], Francisco F. Ludueña-Almeida[1,6], Mariela Ainete[7], María Frias-Cespedes[7], Michael Robert[8], Moory M. Romero[2,9] and Walter R. Almirón[1]

[1] Instituto de Investigaciones Biológicas y Tecnológicas (IIBYT) CONICET- Universidad Nacional de Córdoba. Centro de Investigaciones Entomológicas de Córdoba, Facultad de Ciencias Exactas, Físicas y Naturales, Universidad Nacional de Córdoba. Ciudad Universitaria, Córdoba Capital, Córdoba, Argentina

[2] Institute for Global Health & Translational Sciences, SUNY Upstate Medical University, Syracuse, NY, USA

[3] Department of Medical Geography, University of Florida, Gainesville, FL, USA

[4] InterAmerican Institute for Global Change Research (IAI), Montevideo, Department of Montevideo, Uruguay

[5] Centro de Investigación Esquel de Montaña y Estepa Patagónica (CIEMEP), CONICET and Universidad Nacional de la Patagonia San Juan Bosco. Facultad de Ciencias Naturales y Ciencias de la Salud, Sede Esquel. Esquel, Chubut, Argentina

[6] Cátedra de Matemática (Cs. Biológicas), Facultad de Ciencias Exactas, Físicas y Naturales, Universidad Nacional de Córdoba, Ciudad Universitaria, Córdoba Capital, Córdoba, Argentina

[7] Ministerio de Salud de la Provincia de Córdoba- Dirección de Epidemiología, Hospital San Roque Viejo, Córdoba Capital, Córdoba, Argentina

[8] Department of Mathematics, Statistics, and Physics, University of the Sciences, Philadelphia, PA, USA

[9] Department of Environmental Studies, State University of New York College of Environmental Science and Forestry (SUNY ESF), Syracuse, NY, USA

*Corresponding author:
Email: eelizabet@gmail.com (ELE)

¶These authors contributed equally to this work.

Abstract

Background: Argentina is located at the southern range of arboviral transmission by *Aedes aegypti* and has experienced a rapid increase in arbovirus transmission in recent years. This study aims to present the study design and findings from the first 9 years of an entomological surveillance work that began in Córdoba following the emergence of dengue one decade ago. We also investigate the temporal dynamics of *Ae. aegypti*, dengue cases, and local climate, and their lagged associations.

Methods: From 2009 to 2017, larval surveys were conducted monthly, from November to May, in 600 randomly selected households distributed across the city. From 2009 to 2013, ovitraps (n=177) were sampled weekly to monitor the oviposition activity of *Ae. aegypti.* Cross correlation analysis was used to identify significant lag periods between climate, entomological and epidemiological variables.

Results: *Aedes aegypti* abundance peaked once annually (from January to March), followed by a peak in autochthonous dengue transmission in April. We identified a notable increase in the proportion of homes with juvenile *Ae. aegypti* (from 5.7% of homes in 2009-10 to 15.4% of homes in 2016-17). The mean number of eggs per ovitrap was positively associated with mean temperature. Monthly juvenile *Ae. aegypti* abundance was not associated with either autochthonous or imported dengue cases. Autochthonous dengue transmission was negatively correlated with lagged temperature and precipitation.

Conclusions: These findings suggest increasing the risk of arbovirus transmission in this temperate region. These results can guide targeted vector control interventions and the development of climate services for the public health sector to reduce the burden of arboviral diseases.

Author summary

There is an increasing risk of arbovirus transmission in temperate regions. Argentina is located at the southern range of dengue virus transmission by the *Aedes aegypti* mosquito, and in the last decade, epidemics of dengue fever have re-emerged. We present the study design and findings from the first 9 years of an entomological surveillance study in Córdoba, Argentina, following the emergence of dengue. We found that *Ae. aegypti* were most abundant from January to March, followed by a peak in local dengue transmission in April. Over the study period, we noted a considerable increase in the proportion of homes with *Ae. aegypti*. We found that vector indices and dengue transmission were associated with local climate conditions (temperature and precipitation). These results highlight the growing risk of arboviral infections and the role of climate in local arbovirus and vector dynamics in the southern cone of South America.

Keywords: *Aedes aegypti,* Argentina, dengue, ovitrap, larval surveys, surveillance, climate

Introduction

Argentina is located at the southern range of arboviral transmission by *Ae. aegypti* and has experienced the emergence of arbovirus transmission in the last decade [1,2]. Dengue (DENV serotypes 1-4), Chikungunya (CHIKV) and Zika viruses (ZIKV) cause febrile illnesses and are transmitted principally by the female *Ae. aegypti*, with *Ae. albopictus* as a secondary vector. No specific therapeutics or vaccines are yet available for ZIKV [3] or CHIKV [4], and there is limited access to the Dengvaxia© DENV vaccine, which has been licensed in Argentina [5]. During the 2015-2016 epidemic, ZIKV was of great concern to the region, due to potential neurological and congenital complications associated with infections [6]. Vector management remains the primary means of preventing and controlling arboviral disease outbreaks.

The earliest outbreaks of dengue fever were recorded in Argentina in 1916, and no cases were reported for 80 years afterwards [7]. *Aedes aegypti* was considered eradicated from Argentina in 1963; nevertheless, in 1986, it was detected in the northeast area [8]. Dengue first re-emerged in 1997 and 2000 in the subtropical northern region of Argentina [9]. Within the last decade, dengue has emerged in native populations in central and southern Argentina [10]. In 2009, Argentina suffered its first major dengue outbreak, with more than 26,000 cases, 3 severe dengue cases, and 5 confirmed deaths; 10 jurisdictions registered autochthonous cases for the first time [11]. Outbreaks occurred in 2013, 2014, and 2015 (8,735 total cases). In 2016, the most important dengue outbreak to date occurred, with 41,233 confirmed autochthonous cases, 2,681 imported cases and 11 deaths nationwide [12]. Imported cases by CHIKV were first reported in 2014, and in 2016, the first autochthonous cases by CHIKV were reported [13]. Since 2015, ZIKV has been confirmed in four provinces of Argentina, with a total of 871 cases (last case reported on May 11, 2019) [14].

A notable aspect of the emergence of dengue in Argentina has been the expansion of the southern distribution of the vector and virus into temperate latitudes of the Americas. Córdoba (31.4°S, 64.2°W), the second largest city in Argentina (population 1.3 million) has registered 1,429 dengue cases and all four dengue virus serotypes since the first outbreak in 2009 through the end of 2018 [2]. *Aedes aegypti* was first detected in Córdoba 14 years prior [15]. Human movement plays a major role in dengue transmission in Córdoba; it is hypothesized that the virus is re-introduced each year to trigger disease transmission. Imported cases have been reported from dengue endemic regions including Brazil, Bolivia, Venezuela, northern Argentina, Colombia, Mexico and Costa Rica [2]. Córdoba is an important place to investigate the emergence of arboviruses, as it is among the southernmost cities in the Western Hemisphere to report autochthonous arbovirus cases.

Aedes aegypti is an urban mosquito vector, living in and around human dwellings, and feeding preferentially on human blood [16]. Arbovirus transmission by the mosquito vector occurs from 18-34°C, due to the constraints of the ambient temperature on vector physiology and life history traits [17]. *Aedes aegypti* juvenile habitat consist of water-bearing containers such as tree holes to man-made cisterns, discarded bottles and tires [18]. Rainfall and drought can both potentially increase the availability of larval habitat, depending on local water storage practices and housing characteristics [19,20].

Our current understanding of *Ae. aegypti* population dynamics in Argentina is largely derived from prior studies on oviposition activity in the northern and central areas of the country. Prior studies in the subtropical northwest region of Argentina revealed a year-round vector population in areas with an annual mean temperature around 20ºC. A low number of eggs was recorded during the winter season, and peak in oviposition was detected during summer months (December to March in the southern hemisphere) [21–24]. Studies from the temperate central areas, like Buenos Aires [25–29] and Córdoba provinces [30–32], found that oviposition discontinued during the winter months due to low temperatures (below 17ºC), with a peak in *Ae. aegypti* during the warm, rainy summer months, from October to April.

In response to the first dengue outbreak in Córdoba City in 2009, the province promulgated the 9666 law, which created the "Master Plan to Fight Dengue", and other diseases transmitted by the same vector [33]. Epidemiological and vector surveillance are the responsibilities of the Epidemiology Area of the Zoonosis Program of the Ministry of Health (MoH) of the province. The MoH began the *Ae. aegypti* surveillance and control using larval surveys and ovitraps during the season of vector activity (October to May) in cooperation with local academic partners. Vector surveillance is used to determine changes in the distribution and density of the vector, to evaluate control programs, obtain relative measurements of the vector population over time, and facilitate appropriate and timely decisions regarding interventions [34]. In Córdoba, vector control is mostly focal around homes with dengue cases, and includes control of adult mosquitoes by indoor and outdoor fumigation, control of larval mosquitoes by *Bacillus thuringiensis israelensis* (BTI) larvicide, and eliminating standing water [7].

Monitoring vector populations, disease cases, and local climate conditions can provide important information for assessing arbovirus transmission risk [20,35,36,37].

However, the linkages between entomological risk, arbovirus risk, and local climate are not always clear when comparing regions with distinct eco-epidemiological contexts (e.g., tropical endemic region versus temperate emergence region) and when different entomological surveillance methodologies are used (e.g., ovitraps, larval surveys, adult trapping). Studies in Puerto Rico, Vietnam, and Trinidad found that vector density measurements were associated with dengue transmission [38–40]; however, studies in Venezuela and Malaysia found no relation [41,42]. Few studies of this nature have been conducted in temperate zones of arbovirus emergence.

To address this gap, this study aims to present the study design and findings from the first 9 years of an entomological surveillance study that began in Córdoba following the dengue emergence one decade ago. We also investigate the temporal dynamics of *Ae. aegypti*, dengue cases, and local climate, and their lagged associations. The results of this study can inform surveillance strategies, which are urgently needed to reduce disease risk in settings of arbovirus emergence.

Methods

Ethics

Entomological data were collected from households by the MoH of Córdoba as part of the routine surveillance program, thus no ethical review or informed consent was required. For this analysis, weekly and monthly entomological data were aggregated to the city-level, with no identifying information for households. Dengue case data at the city-level were extracted from National MoH bulletins and aggregated to weekly and monthly case counts; no identifying information was provided.

Study site

This study was conducted in the city of Córdoba (31.4°S, 64.2°W), which is located in the central region of Argentina (elevation: 360-480 m above sea level) [43]. The city has grown outward from the center to the periphery; agricultural fields with patches of forest surround the urban core [44] (Fig 1). The local climate of Córdoba is warm temperate with hot summers and four seasons (Cwa under the Köppen-Geiger classification system) [45]. On average, the city center is several degrees warmer than the urban periphery, due to dense construction and location in a topographic depression [46]. Summer is a warm, rainy season from November to March (monthly mean temp: 22.7°C, max temp: 37.6°C, min temp: 11.8°C, monthly total rainfall: 123.2mm), and winter is a cool, dry season from June to September (monthly mean temp: 14.8°C, max temp: 30.9°C, min temp: 2.4°C, monthly total rainfall: 36.1mm). *Aedes aegypti* is the only known vector of dengue in Córdoba. The vector does not reproduce (oviposit) during the winter, presumably due to cool temperatures [31]; it is thought that the vectors persist through the cold season as egg.

Fig 1. **Location of Córdoba in South America** (A)**.** Show the lat/long lines, and the 5 sampling quadrants in the city (B).

Field studies were conducted under the *Ae. aegypti* surveillance program of the Department of Epidemiology of the Córdoba Province Ministry of Health in cooperation with the Córdoba Entomological Research Centre (CIEC) of Córdoba National University (UNC) and the Biological and Technological Research Institute (IIByT) of Córdoba National University (UNC) and the National Research Council (CONICET). No formal ethical review or consent procedure was required for this study, as the field operations were conducted as part of routine surveillance activities by the MoH. Entomological samples were collected from households that verbally agreed to participate in the study and were taken to the CIEC laboratory to be processed.

Larval sampling

From 2009 to 2017, the distribution and abundance of *Ae. aegypti* were monitored through monthly larval surveys conducted across the city. Sample periods each year were chosen by the CIEC laboratory team to span the period before, during and after mosquito activity peak in the region [32]. Each month, 600 homes were randomly selected for sampling. The city was divided into 215 quadrants (1.2 km per side), which were distributed across five areas of approximately the same land area: Central, Northeast, Northwest, Southeast, and Southwest (Fig 1). In each quadrant, we randomly selected 6 neighborhoods, and the field technicians randomly selected 20 homes to be inspected per neighborhood.

At each home, an inspector noted all containers inside and outside the home with standing water. Inspectors noted the presence of juvenile mosquitoes and collected specimens for species identification. Whenever possible, all juvenile mosquitoes were collected; for large containers where it was not possible to collect all specimens, an inspector collected three samples using a white dipper (62 ml volume). Specimens were transported to the CIEC laboratory; pupae were reared to adults, and 3rd and 4th instar larvae were preserved in 80% ethanol and were identified using taxonomic keys [47]. First and 2nd instar larvae were reared until reaching the 3rd instar in plastic trays with 500 ml of water from the natural larval habitat or dichlorine water. Each day larvae were fed 0.25 mg of liver powder per larva, and we cleaned the surface of the water using absorbent paper to avoid contamination by fungi and/or bacteria. The juvenile mosquitoes were counted and identified to species. For the purposes of this study, data were aggregated to the city level and we calculated the proportion of households and neighborhoods with juvenile *Ae. aegypti* present during each sampling period.

Ovitraps

The weekly oviposition activity of *Ae. aegypti* was observed from November to May over four years (2009 – 2013) using ovitraps, a sensitive method of detecting the presence of gravid female of *Ae. aegypti* [48]. Ovitraps were placed in randomly selected households (N=177) that were distributed evenly across the city. To assess oviposition during the winter months, a subset of evenly distributed ovitraps (n=40) were selected to continue sampling during June (late autumn-early austral winter seasons) to September 2010 (late winter-early austral spring seasons).

Ovitraps consisted of plastic bottles (350 ml volume, 8 cm diameter x 13 cm height) with filter paper as an oviposition substrate. An attractive infusion (250 ml) was prepared by fermenting dry cut grass with tap water [49]. Each week the traps were inspected and replaced. Traps were transferred to the laboratory and the number of *Ae. aegypti* eggs per trap were counted using a stereomicroscope. For the purposes of this study, ovitrap data were aggregated to the city level. We calculated the proportion of traps that were positive/negative for *Ae. aegypti* eggs and the mean number of eggs per trap during each sampling period.

Climate information

The Argentinian National Meteorological Service provided daily weather data for the study period (rainfall, min/mean/max temperature, min/mean/max relative humidity) from the Observatory station (31.42°S; 64.20°W). This is one of the oldest weather stations in Latin America (in operation since 1871) and the most reliable in Córdoba [50]. We calculated summary monthly values (mean, standard deviation) over the study period, summary values by epidemiologic week, and summary values corresponding with the timing of ovitrap and larval survey collection periods.

Dengue case data

Weekly dengue case reports (January 2009 to December 2017) were extracted from the weekly epidemiological bulletins of the Argentina Health Secretary [51]. Dengue diagnostic procedures and case definitions have been previously described [2]. Data extraction was described in detail previously [2]. Cases include suspected and laboratory confirmed cases aggregated at the city-level and annual incidence was calculated using the total city population for the corresponding year.

Statistical analysis

Statistical analyses were conducted at the city-level using R (version 3.3.3) in RStudio (version 1.0.136), using the packages splines, TSA, geepack, MASS, lubridate, TSimpute [52–58]. Missing values in the ovitrap data (n=10) were imputed via seasonally decomposed structural modeling with Kalman smoothing. Missing values in the larval data (n=20) were imputed via seasonally decomposed random selection. For each climate, mosquito abundance, and dengue incidence variable, we performed a spectral analysis and subsequently tested for the presence of inter-annual variability with a restricted cubic spline using model fit to determine the periodic frequency and the number of knots. Models were fit using a generalized linear model with appropriate distributions for each variable using generalized estimating equations (auto-regressive correlation); best fit was determined using quasi-likelihood information criteria (QIC) as compared to a null model. Residual plots and model assumptions were examined to inform final model selection. Data were analyzed in relation to the collection periods of each respective variable; means and frequencies for these collection periods were calculated for reference.

We assumed a unidirectional temporal relationship between the variables as follows: climate affecting all other variables, *Ae. aegypti* eggs affecting *Ae. aegypti* larvae and dengue incidence, and *Ae. aegypti* larvae variables affecting dengue incidence. Both local and imported cases were included in the analysis: local cases because they would have been acquired within the local climate and mosquito population imported cases because typical vacation periods (e.g. school vacations, holidays) coincide with specific times of year and the climate conditions of those annual occurrences. To examine the correlations between these variables, cross-correlation functions were calculated between differenced monthly summary data for each variable with lags up to two months. We selected this period as it is a biologically plausible period of time that includes the combined time for *Ae. aegypti* egg hatching and larval development to adult mosquitoes.

Results

Aedes aegypti were detected over the entire study period, with the exception of the winter months (June to September), where winter was sampled in 2010 only. Time series of *Ae. aegypti* abundance and dengue cases are shown in Fig 2. Monthly mean temperature, total precipitation, and mean daily relative humidity are shown in Fig 3. Annual (over a season) and monthly summary statistics for *Ae. aegypti* eggs and larval abundance and dengue incidence are shown in Figs 4-9. Summary statistics for all variables are in S1 Table.

Fig 2. Time series of monthly dengue and *Aedes aegypti* abundance. Top: Dengue cases. Center: proportion of homes with juvenile *Aedes aegypti*. Bottom: proportion of ovitraps (N=177) positive for *Aedes aegypti* and the mean number of eggs per ovitrap.

Fig 3. Times series of monthly climate variables. Mean monthly temperatures (°Celsius) are in the top panel (red) (minimum and maximum in dashed lines), total monthly precipitation (mm) is in the central panel (blue) and mean monthly relative humidity (percentage) is in the bottom panel (green) (minimum and maximum in dashed lines).

Fig 4. Seasonality in ovitrap *Aedes aegypti* egg counts and positivity (2009-2013). Box and whisker plots show the median and quartiles. Top: Number of *Aedes aegypti* eggs collected per ovitrap. Bottom: The percent of ovitraps with *Aedes aegypti* eggs.

Fig 5. Annual ovitrap *Aedes aegypti* eggs counts and positivity. Box and whisker plots show the median and quartiles for annual sampling season (November—May). Top:

Number of *Aedes aegypti* eggs collected per ovitrap per annual sampling season. Bottom: The percent of ovitraps with *Aedes aegypti* eggs per annual sampling season.

Fig 6. Seasonality in *Aedes aegypti* larval abundance (2009-2017). Box and whisker plots show the median and quartiles. Top: Percent of homes with water-bearing containers with juvenile *Aedes aegypti*. Bottom: Percent of neighborhoods with water-bearing containers with juvenile *Aedes aegypti*.

Fig 7. Annual *Aedes aegypti* larval abundance. Box and whisker plots show the median and quartiles for annual sampling season (November—May). Top: Percent of homes with water-bearing containers with juvenile *Aedes aegypti*. Bottom: Percent of neighborhoods with water-bearing containers with juvenile *Aedes aegypti*.

Fig 8. Total annual dengue cases. Top: Total annual autochthonous dengue cases (no travel history). Bottom: Total annual imported dengue cases.

Fig 9. Seasonality in dengue cases (2009-2017). Box and whisker plots show the median and quartiles. Top: Counts of autochthonous dengue cases (no travel history). Bottom: Counts of imported dengue cases.

Aedes aegypti egg abundance was highest in January (Fig 4), with a median of 33 eggs per trap and 61% of traps positive for eggs during this month. *Aedes aegypti* egg abundance was highest in the 2009-10 and 2011-12 seasons (Fig 5), with a mean of 21 eggs per ovitrap per month for both seasons, and 36.4% and 41.6% of traps positive for *Ae.*

aegypti eggs in 2009-10 and 2011-12, respectively. Note that ovitraps were used only until 2013 (4 collection seasons).

Aedes aegypti larval abundance was highest in March (residences) and February (neighborhoods) in each year (Fig 6). During these months, 14% of residences and 85% of neighborhoods had containers positive for larvae. There was a notable increase in *Ae. aegypti* larval abundance, with the highest infestation levels detected in 2015-16 and 2016-17 (Fig 7), with 14 and 15.8% of residences and 83.1 and 71.3% of neighborhoods having containers positive for *Ae. aegypti* larvae in 2015-16 and 2016-17, respectively.

Dengue cases were highest during April across all years, with a local transmission season from March to May (mean 65 autochthonous cases and mean 9 imported cases, Fig 8). The number of dengue cases was highest in 2016 (Fig 9), with 689 total autochthonous and 139 total imported cases.

Periodicity

The results of the seasonal analysis for climate, mosquito abundance, and dengue incidence variables are presented in Table 1. All climate variables were found to have a 12-month periodicity with nonlinear interannual variability, except for minimum relative humidity, which had no interannual variability, and maximum relative humidity, which had no periodicity nor interannual variability (*i.e.* temporal trend).

Table 1. Seasonality and Interannual Variability of Data.

Variable	Frequency of Season	Period Frequency	Mean Timing of Seasonal Peak	Shape of Interannual Variability

Climate	Minimum Temperature	12 months	annual	early June	non-linear
	Mean Temperature	12 months	annual	late May	non-linear
	Maximum Temperature	12 months	annual	early May	non-linear
	Total Precipitation	12 months	annual	early June	non-linear
	Minimum Relative Humidity	12 months	annual	early August	none
	Mean Relative Humidity	12 months	annual	late August	non-linear
	Maximum Relative Humidity	none	na	na	none
Eggs	% Positive Traps	27 periods[A]	~annual	na	none
	Mean Number of Eggs per Trap	27, 53 periods[A]	~annual, every 2 years	na	non-linear
Larvae	Positive Homes	none	na	na	none
	Positive Neighborhoods	5 & 50 periods[B]	~annual, every 10 years	na	non-linear
Dengue Incidence	Autochthonous	24 & 45.5 periods[C]	~twice per year, ~annual	na	non-linear
	Imported	60.2 & 120.7 periods[C]	~every 1.5 years, ~every 3 years	na	non-linear

For each variable, the seasonal frequency and presence of interannual variability for the best-fit model is reported, with period frequency included for those variables with partial-year collection periods. For variables with an annual seasonality, the timing of the seasonal peak is also reported.

na=not applicable

[A]On average, collection periods were every 7 days and occurred 26 times in a collection season (November—May).

[B]On average, collection periods were every 39 days and occurred six times in a collection season (September—May)

[C]On average, surveillance frequency was 8 days and occurred 39 times in a year

Aedes aegypti eggs were collected from ovitraps every 7 days on average; an average of 26 times within each collection season. Both *Ae. aegypti* egg abundance variables (trap positivity and mean number of eggs) exhibited a peak about once each year on average, and the mean number of eggs had an additional peak occurring every 2 years on average. Trap positivity had no interannual variability while the mean number of eggs had nonlinear interannual variability.

Over the study period, on average, *Ae. aegypti* larvae were collected every 39 days, six times per collection season. *Aedes aegypti* larvae abundance variables (proportion of positive homes, proportion of positive neighborhoods) were found to have different seasonal patterns: there was no periodicity nor interannual variability for the proportion of positive homes, but the proportion of positive neighborhoods peaked once per year, as well as every 10 years.

Dengue surveillance reports of autochthonous and imported cases occurred every 8 days on average; an average of 39 times per year. Autochthonous dengue incidence peaked approximately twice per year and annually. Imported dengue incidence peaked approximately every 1.5 and 3 years. There was no interannual variability in autochthonous or imported dengue incidence.

Cross correlation analysis

The results of the cross-correlation between *Ae. aegypti* eggs and larval abundance, climate, and dengue incidence variables are shown in Table 2. *Aedes aegypti* egg abundance (mean number of eggs per ovitrap) was positively correlated with mean temperature in the same month. *Aedes aegypti* larval abundance (percent of neighborhoods with positive larval traps) was positively correlated with minimum temperature in the same month, while the percentage of residences with positive larval traps was not correlated with any climate variable. Autochthonous dengue incidence was negatively correlated with mean and maximum temperature at lag 2, and negatively correlated with total precipitation in the same month, with no correlation to any relative humidity variables. Imported dengue incidence was negatively correlated with mean temperature at lag 2, positively correlated with minimum relative humidity at lag 1, and correlated with total precipitation in the same month (positively) and at lag 1 (negatively). *Aedes aegypti* egg abundance was not correlated with autochthonous nor imported dengue incidence. *Aedes aegypti* egg abundance was positively correlated with the percent of neighborhoods with positive larval traps at lag 1. *Aedes aegypti* larvae abundance (percent of neighborhoods with positive larval traps) was negatively correlated with autochthonous dengue incidence at lag 1; both percent of neighborhoods and percent of residences with positive larval traps were positively correlated with imported dengue incidence in the same month.

Table 2. Cross-Correlation Between Climate, Mosquito Abundance, and Dengue Cases for 2009—2017.

Pairing	Variable A	Variable B	Correlated Lags	Direction of Correlation
Climate & Egg Abundance	Min Temp	Mean Number of Eggs	none	
	Mean Temp		0 months	positive
	Max Temp		none	
	Min Rel Humidity		none	
	Mean Rel Humidity		none	
	Max Rel Humidity		none	
	Total Precipitation		none	
Climate & Larvae Abundance	Min Temp	% Positive Homes	none	
	Mean Temp		none	
	Max Temp		none	
	Min Rel Humidity		none	
	Mean Rel Humidity		none	

	Max Rel Humidity		none	
	Total Precipitation		none	
	Min Temp	% Positive Neighborhoods	0 months	positive
	Mean Temp		none	
	Max Temp		none	
	Min Rel Humidity		none	
	Mean Rel Humidity		none	
	Max Rel Humidity		none	
	Total Precipitation		none	
Climate & Dengue Incidence	Min Temp	Autochtonous Dengue Incidence	none	
	Mean Temp		2 months	negative
	Max Temp		2 months	negative
	Min Rel Humidity		none	
	Mean Rel Humidity		none	
	Max Rel Humidity		none	
	Total Precipitation		0 months	negative
	Min Temp	Imported Dengue Incidence	none	
	Mean Temp		2 months	negative
	Max Temp		none	
	Min Rel Humidity		1 months	positive
	Mean Rel Humidity		none	
	Max Rel Humidity		none	
	Total Precipitation		0, 1 months	positive, negative
Mosquito Egg & Dengue Incidence	Mean Eggs/ovitrap	Autochtonous Dengue Incidence	none	
		Imported Dengue Incidence	none	
Mosquito Egg & Larvae Abundance	Mean Eggs/ovitrap	% Positive Homes	none	
		% Positive Neighborhoods	1 months	positive
Mosquito Larvae Abundance & Dengue Incidence	Positive Homes	Autochtonous Dengue Incidence	none	
	Positive Neighborhoods		1 months	negative
	Positive Homes	Imported Dengue Incidence	0 months	positive
	Positive Neighborhoods		0 months	positive

Using the cross-correlation function, we examined the correlation between pairs of variables, with variable A hypothesized to occur 0 to 2 months before variable B.

Discussion

Here we describe the seasonal and interannual patterns of climate, *Ae. aegypti* eggs and larval abundance, and dengue transmission in the temperate city of Córdoba, Argentina, one decade after the first epidemic of dengue. We identified a notable increase in the proportion of homes with juvenile *Ae. aegypti* (from 5.7% of homes in 2009-10 to

15.4% of homes in 2016-17), suggesting an increased risk of arbovirus transmission in the region.

Climate is a critical driver of *Ae. aegypti* eggs and larval abundance and dengue incidence. Long-term and seasonal patterns of dengue vary around the globe, with some locations experiencing stable transmission throughout the year and other experiencing single or multiple dengue seasons within each year [59–63]. We found that the temperatures of Córdoba were warm enough to support survival and breeding of *Ae. aegypti* populations from October to April. We found that *Ae. aegypti* eggs and larval abundance peaked once annually (from January to March), followed by a peak in autochthonous dengue transmission in April. Prior studies from Córdoba found that *Ae. aegypti* egg-to-adult survival began at mean minimum ambient temperatures greater than 13°C [32], suggesting that *Ae. aegypti* populations in Córdoba are not likely to survive from May to September, when temperatures drop below this threshold. Thus, these findings indicate that the temporal window for a stable *Ae. aegypti* population in Córdoba is very short, limiting the period of potential autochthonous dengue transmission. Indeed, Estallo *et al*. [11] showed that low autumnal temperatures were an important factor limiting the spread of dengue during the first dengue outbreak in 2009.

Ambient temperatures were associated with vector abundance, as expected from the thermal biology of ectothermic mosquito vectors [64]. We found that the mean number of eggs per ovitrap was positively associated with mean temperature; the number of eggs laid per female of *Ae. aegypti* is closely linked to temperature [17]. As we would expect from thermal limits of egg-to-adult survival [17], larval abundance (neighborhoods) was correlated with minimum temperature. Whether a particular abundance measure is best correlated to minimum or mean temperature likely has to do with how well these particular measures represent the day-to-day temperature-mosquito dynamics that are occurring within Córdoba. Our data represent monthly summaries that vary in their ability to precisely detect the complex relations between climate and mosquitos. The vector activity in the temperate Córdoba city begins after low winter temperatures. Larvae probably hatched from eggs laid during the previous season. In Córdoba vector activity decreases at low autumnal temperatures until adults die. Grech *et al*. [65] found that, for Córdoba city, once the ambient temperature has become greater than the thermal threshold (11.11ºC), 93.7 degree-days are necessary to for larval-pupa development and adult emergence to be completed.

While we would not expect imported dengue cases to be affected by local *Ae. aegypti* populations, imported cases also peak annually in March. This pattern could be caused by human movement within and outside the country, as this is a significant risk factor for dengue emergence in a non-endemic city like Córdoba [11]. For example, during 2009 dengue outbreak in the city, the introduction of DENV was associated with outbreaks in the neighboring countries of Bolivia, Brazil and Paraguay at the end of 2008, and also with dengue transmission in the Northern provinces of Argentina, where 92% of dengue cases occurred [11]. It is likely that human movement into Córdoba from neighboring areas corresponds with summer breaks in school schedules in Argentina (December-February) before university classes begin each year in March. An estimated 60% of the ~132,000 university students in the National University of Córdoba originate from other provinces or countries [66,67]. Therefore, cases of dengue could be imported by students who travel between Córdoba and northern provinces and people vacationing in tropical dengue endemic countries. Additionally, cases could be imported by migrant workers traveling to and from endemic countries, like Bolivia and Paraguay.

Autochthonous dengue transmission was negatively correlated with several variables such as temperature and precipitation, which was somewhat unexpected. As temperatures declined towards the later part of the season of vector activity (end of summer), dengue transmission increased. As previously indicated, this may be related to the timing of the importation of dengue virus from people traveling from neighboring endemic regions.Transmission peaked in April, when mean daily temperatures were 18ºC (mean min temp = 13.6ºC, mean max temp = 25.5ºC). Prior studies reported that arbovirus transmission by *Ae. aegypti* declined to zero below 17.8ºC (lower thermal limit) [17]. We hypothesize that local *Ae. aegypti* populations have adapted to cooler climate conditions, while at the same time local climate conditions have warmed to permit arbovirus transmission at the lower thermal limit [64].

We found that monthly juvenile *Ae. aegypti* abundance was not associated with either autochthonous or imported dengue cases, confirming prior studies in Córdoba [11]. Several possible explanations are as follows: (1) surveillance of juvenile vectors did not capture the entomological risk presented by adults of *Ae. aegypti;* (2) spatial variation in entomological risk leads to localized hotspots of transmission risk that are not captured in the aggregated city-level data; and (3) in a zone of emergence, a high proportion of the population is immunologically naive to dengue, thus transmission depends on the timing of the introduction of the virus and may be sustained with low vector densities. Prior studies also found that dengue transmission was not associated with vector densities. In Brazil, dengue transmission was most closely related to the movement of viremic humans rather than vector densities [68].

As indicated by the appearance and continued transmission of dengue in Córdoba, the city, along with other parts of temperate Argentina, are actively experiencing the emergence of dengue [1,2]. Emergence of dengue in temperate regions has been occurring with more frequency in the past two decades [69–71] as climate becomes increasingly favorable for *Ae. aegypti* in Argentina, and increase their distribution and active periods of the year. Our study highlighted significant relationships between temperature and mosquito indices, indicating that climate is potentially playing an important role in the emergence of dengue in Córdoba; however, our study was not conclusive on the environmental drivers of vector activity and dengue transmission in the city and highlights the need to conduct further studies investigating emergence. In particular, studies of human movement, including origins, destinations, and timing, throughout South America will be critical to understanding patterns of emergence of dengue and other arboviruses in Córdoba. Further, our study, independently and combined with future studies, will be helpful in understanding dengue emergence on a more global scale. The data presented here is one of the most comprehensive databases describing dengue emergence and drivers thereof that is currently available and our study is among few such studies investigating arbovirus emergence in a temperate climate as it is actively occurring.

Our results can be used to guide surveillance and control efforts in this region. Based on *Ae. aegypti* egg abundance variables (trap positivity and mean number of eggs) exhibiting a peak once per collection season, we suggest public health authorities to make vector management and educational campaigns before the occurrence of peaks to prevent the spread of larvae. Juvenile vector surveillance is a useful tool to detect temporal variation, and we recommend the maintenance of surveillance through ovitraps and larval surveys. Alternative surveillance and control strategies, such as novel adult traps, should also be explored [72,73]. Our results also suggest the potential to develop climate services to support the public health sector [74]. For example, if seasonal climate forecasts are shared on a regular basis with the health sector, this information can potentially be used to predict when there will be an increase in vector abundance and disease transmission risk, as done demonstrated in dengue endemic regions [19,75]. Climate services, such as climate driven early warning systems to predict arbovirus transmission risk, can leverage the existing robust entomological and climate surveillance and monitoring systems.

Empirical studies such as this, from the lower thermal range of arbovirus transmission, can be used to improve models that investigate arbovirus dynamics in temperate zones of emergence, predict future outbreaks, and explore potential control measures. Understanding seasonal variability in vector populations and dengue transmission that is potentially driven by variability in temperature, precipitation, and/or humidity is crucial to understanding periods of the year in which risk of dengue transmission is highest [59,76,77]. For example, our study indicates that meteorological variables such as mean and maximum temperature and minimum relative humidity are significantly correlated with dengue transmission at one- or two-month lags (Table 2). By combining this information with known mechanistic relationships between meteorological variables and mosquito population and dengue transmission parameters, models can help to predict transmission or windows of risk of transmission utilizing meteorological indices. This, in turn, can help inform mathematical models for studying the potential impacts of vector and disease control measures so that the most appropriate control measures can be implemented at optimal times to have the greatest impact on reducing dengue transmission. Furthermore, well-characterized relationships between vector dynamics and arbovirus transmission and current typical meteorological conditions are important for providing baseline information for mathematical models that can be utilized to better understand how changes in climate, such as increased average temperatures and extreme deviations in precipitation patterns, may alter the potential for arbovirus emergence and spread in the future [64,78,79].

It is important to note that we do not expect that the correlations between climate or mosquito abundance and imported dengue case incidence to be causal in any way. Cross-correlation analyses do not adjust for potential confounding and we expect that the correlation found is due to the coincidental timing of Argentina vacation periods and peak larval activity. One limitation of our work is irregular sampling in the surveillance data, creating gaps in some periods, though the sampling design was strong and carefully designed by the team of researchers at the CIEC.

Conclusions

This longitudinal study provides insights into the complex dynamics of arbovirus transmission and vector populations in a temperate region of arbovirus emergence. Studies such as this provide critical local-level information to guide public health interventions. Our findings suggest that Córdoba is well suited for arbovirus disease transmission, given the stable and abundant vector populations. It is possible that the region may shift from epidemic to endemic dengue transmission if measures are not taken to reduce vector populations and disease transmission.

Conflict of interest

The authors report no conflicts of interest.

Acknowledgements

The authors wish to acknowledge the United States Embassy in Argentina and Fulbright Commission as well as the Department of Epidemiology of the Córdoba Province Ministry of Health. AMSI and MAR were support in Córdoba Argentina by the USA Zika program of the United States Embassy in Argentina administrated by Fulbright commission. ELE, MGG, and WRA is a member of the Consejo de Investigaciones Cientificas y Tecnologicas (CONICET) from Argentina, EMB is a PhD Student with scholarship support from CONICET.

References

1. Masuh H. Re-emergence of dengue in Argentina: Historical development and future challenges. Dengue Bull. 2008; 32: 44-54.
2. Arbovirus emergence in the temperate city of Córdoba, Argentina, 2009–2018.
3. Robert MA, Tinunin DT, Benitez EM, Ludueña-Almeida F, Romero M, Stewart-Ibarra AM, et al. Arbovirus emergence in the temperate city of Córdoba, Argentina, 2009-2018. Scientific Data. 2019; 6:276. Available from: https://doi.org/10.1038/s41597-019-0295-z.
4. Cohen J. The race for a Zika vaccine is on. Science. 2016; 351: 543–544.
5. Smalley C, Erasmus JH, Chesson CB, Beasley DW. Status of research and development of vaccines for chikungunya. Vaccine. 2016; 34: 2976–2981.
6. Vannice KS, Durbin A, Hombach J. Status of vaccine research and development of vaccines for dengue. Vaccine. 2016; 34: 2934–2938.
7. WHO. Zika virus. World Health Organization [Cited 2016 Feb 24]. Available from: http://www.who.int/mediacentre/factsheets/zika/en/.
8. Plan nacional para la prevención y el control del dengue y la fiebre amarilla; 2009 [cited 2018 Apr 25]. Available from: http://www.msal.gob.ar/images/stories/cofesa/2009/acta-02-09/anexo-5-resumen-plan-dengue-02-09.pdf.
9. Curto S, Boffi R, Carbajo AE, Plastina R, Schweigmann N. Reinfestación del territorio Argentino por *Aedes aegypti*. Distribución geográfica (1994-1999). In: Salomón OD, editor. Actualizaciones en Artropodología Sanitaria Argentina. Buenos Aires: Fundación Mundo Sano; 2002. pp. 127–137.

10. Avilés G, Rangeón G, Vorndam V, Briones A, Baroni P, Enria D, et al. Dengue reemergence in Argentina. Emerg Infect Dis. 1999; 5: 575.
11. Vezzani D, Carbajo AE. *Aedes aegypti*, *Aedes albopictus*, and dengue in Argentina: current knowledge and future directions. Mem Inst Oswaldo Cruz. 2008; 103: 66–74.
12. Estallo EL, Carbajo AE, Grech MG, Frías-Céspedes M, López L, Lanfri MA, et al. Spatio-temporal dynamics of dengue 2009 outbreak in Córdoba City, Argentina. Acta Trop. 2014; 136: 129-136.
13. Dirección nacional de epidemiología y análisis de la situación de salud. Boletín integrado de vigilancia: No. 341, SE 52; 2016 [cited 2019 Apr 17]. Available from: https://www.argentina.gob.ar/sites/default/files/boletin_integrado_vigilancia_n341-se52.pdf.
14. Dirección nacional de epidemiología y análisis de la situación de salud. Boletin Integrado de Vigilancia: No. 327, SE 37; 2016 [cited 2018 Jul 10]. Available from: http://www.msal.gob.ar/images/stories/boletines/Boletin-Integrado-De-Vigilancia-N327-SE37.pdf.
15. PAHO. PLISA Health Information Platform for the Americas. Pan American Health Organization / World Health Organization; 2019 [cited 2019 Oct 8]. Available from: http://www.paho.org/data/index.php/en/?option=com_content&view=article&id=524&Itemid=.
16. Almirón WR, Ludueña-Almeida F. *Aedes aegypti* (Diptera: Culicidae) en Córdoba, Argentina. Rev Soc Entomol Argent. 1998; 57: 27-28.

17. WHO. Dengue: guidelines for diagnosis, treatment, prevention and control. World Health Organization; 2009 [cited 2018 Apr 28]. Available from: https://apps.who.int/iris/handle/10665/44188
18. Mordecai EA, Cohen JM, Evans MV, Gudapati P, Johnson LR, Lippi CA, et al. Detecting the impact of temperature on transmission of Zika, dengue, and chikungunya using mechanistic models. PLoS Negl Trop Dis. 2017; 11: e0005568.
19. Manrique-Saide P, Che-Mendoza A, Rizzo N, Arana B, Pilger D, Lenhart A, et al. Operational guide for assessing the productivity of *Aedes aegypti* breeding sites. Geneva Switz WHO-TDR. 2011; pp. 1–30.
20. Lowe R, Gasparrini A, Van Meerbeeck CJ, Lippi CA, Mahon R, Trotman AR, et al. Nonlinear and delayed impacts of climate on dengue risk in Barbados: A modelling study. PLoS Med. 2018; 15: e1002613.
21. Stewart Ibarra AM, Ryan SJ, Beltrán E, Mejía R, Silva M, Muñoz Á. Dengue Vector Dynamics (*Aedes aegypti*) Influenced by Climate and Social Factors in Ecuador: Implications for Targeted Control. PLoS One. 2013; 8: e78263.
22. Micieli MV, Campos RE. Oviposition activity and seasonal pattern of a population of *Aedes* (*Stegomyia*) *aegypti* (L.) (Diptera: Culicidae) in subtropical Argentina. Mem Inst Oswaldo Cruz. 2003; 98: 659–63.
23. Estallo EL, Benitez EM, Lanfri MA, Scavuzzo CM, Almirón WR. MODIS environmental data to assess Chikungunya, Dengue, and Zika diseases through *Aedes* (*Stegomia*) *aegypti* oviposition activity estimation. IEEE J Sel Top Appl Earth Obs Remote Sens. 2016; 9: 5461–5466.

24. Estallo EL, Más G, Vergara-Cid C, Lanfri MA, Ludueña-Almeida F, Scavuzzo CM, et al. Spatial patterns of high *Aedes aegypti* oviposition activity in northwestern Argentina. PLoS One. 2013; 8: e54167.

25. Estallo EL, Ludueña-Almeida FF, Introini MV, Zaidenberg M, Almirón WR. Weather variability associated with *Aedes* (*Stegomyia*) *aegypti* (Dengue vector) Oviposition dynamics in northwestern Argentina. PLoS One. 2015; 10: e0127820.

26. Campos RE, Maciá A. Observaciones biológicas de una población natural de *Aedes aegypti* (Diptera: Culicidae) en la provincia de Buenos Aires, Argentina. Rev Soc Entomol Argent. 1996; 55: 67–72.

27. Carbajo AE, Gómez SM, Curto SI, Schweigmann NJ. Variación espacio-temporal del riesgo de transmisión de dengue en la Ciudad de Buenos Aires. Med B Aires. 2004; 64: 231–234.

28. Carbajo AE, Curto SI, Schweigmann NJ. Spatial distribution pattern of oviposition in the mosquito *Aedes aegypti* in relation to urbanization in Buenos Aires: southern fringe bionomics of an introduced vector. Med Vet Entomol. 2006; 20: 209–218.

29. Vezzani D, Velázquez SM, Schweigmann N. Seasonal pattern of abundance of *Aedes aegypti* (Diptera: Culicidae) in Buenos Aires city, Argentina. Mem Inst Oswaldo Cruz. 2004; 99: 351–356.

30. Micieli MV, García JJ, Achinelly MF, Martí GA. Dinámica poblacional de los estadios inmaduros del vector del dengue *Aedes aegypti* (Diptera: Culicidae): un estudio longitudinal (1996-2000). Rev Biol Trop. 2006; 54: 979–983.

31. Avilés G, Cecchini R, Harrington ME, Cichero J, Asis R, Rios C. *Aedes aegypti* in Córdoba province, Argentina. J Am Mosq Control Assoc Mosq News. 1997; 13: 255–258.

32. Grech MG. Bioecología de culícidos (Diptera) urbanos de importancia sanitaria de la ciudad de Córdoba, Córdoba (Argentina). PhD. Thesis in Biological Sciences, National University of Córdoba. 2013. Available from: https://catalogo.biblio.unc.edu.ar/Record/exactas.25342/Description#tabnav

33. Domínguez MC, Ludueña Almeida FF, Almirón WR. Dinámica poblacional de *Aedes aegypti* (Diptera: Culicidae) en Córdoba capital. Rev Soc Entomol Argent. 2000; 59: 41–50.

34. Constitución de la provincia de Córdoba; 2001 [cited 2018 May 3]. Available from: http://www.infoleg.gob.ar/basehome/ConstituciondeCordoba.htm.

35. Focks DA. A review of entomological sampling methods and indicators for dengue vectors. Geneva World Health Organization; 2003 [cited 2013 May 7]. Available from: http://203.90.70.117/PDS_DOCS/B0219.pdf#page=233.

36. Barrera R, Navarro JC, Mora JD, Dominguez D, Gonzalez J. Public service deficiencies and *Aedes aegypti* breeding sites in Venezuela. Bull Pan Am Health Organ. 1995; 29: 193–205.

37. Arunachalam N, Tana S, Espino F, Kittayapong P, Abeyewickrem W, Wai KT, et al. Eco-bio-social determinants of dengue vector breeding: a multicountry study in urban and periurban Asia. Bull World Health Organ. 2010; 88: 173–184.

38. Benitez EM, Estallo EL, Grech M, Frías-Céspedes M, Almirón WR, Ludueña-Almeida FF. Temporal models using environmental variables to predict *Aedes*

aegypti oviposition activity in a temperate region of Argentina. BioRxiv [Preprint]. 2019 bioRxiv 816421[posted 2019 Oct 23; cited 2019 Oct 30]: [29 p.]. Available from: https://www.biorxiv.org/content/10.1101/816421v1 doi: 10.1101/816421

39. Chadee DD. Dengue cases and *Aedes aegypti* indices in Trinidad, West Indies. Acta Trop. 2009; 112: 174–180.
40. Barrera R, Amador M, MacKay AJ. Population dynamics of *Aedes aegypti* and Dengue as influenced by weather and human behavior in San Juan, Puerto Rico. PLoS Negl Trop Dis. 2011; 5: e1378.
41. Pham H, Doan H, Phan T, Minh NT. Ecological factors associated with dengue fever in a central highlands Province, Vietnam. BMC Infect Dis. 2011; 11: 172.
42. Sulaiman S, Pawanchee ZA, Arifin Z, Wahab A. Relationship between Breteau and House indices and cases of dengue/dengue hemorrhagic fever in Kuala Lumpur, Malaysia. J Am Mosq Control Assoc. 1996; 12: 494-496.
43. Barrera R, Delgado N, Jiménez M, Valero S. Eco-epidemiological factors associated with hyperendemic dengue haemorrhagic fever in Maracay City, Venezuela. Dengue Bull. 2002; 26: 84–95.
44. INDEC. Censo Nacional de Población, Hogares y Viviendas 2010; 2010 [cited 2019 Apr 18]. Available from: https://www.indec.gob.ar/censos_provinciales.asp?id_tema_1=2&id_tema_2=41&id_tema_3=135&p=14&d=999&t=0&s=0&c=2010.
45. Maristany A, Abadía L, Angiolini S, Pacharoni A, Pardina M. Estudio del fenómeno de la isla de calor en la ciudad de Córdoba-Resultados preliminares. Av Energ Renov Medio Ambient. 2008; 12: 11–69.

46. Peel MC, Finlayson BL, McMahon TA. Updated world map of the Köppen-Geiger climate classification. Hydrol Earth Syst Sci Discuss. 2007; 4: 439–473.

47. Asar ML, Estallo EL, Benítez EM, Di Benedetto HM, Ludueña-Almeida FF. Un estudio sobre el efecto Isla de Calor Urbano en la ciudad de Córdoba, Argentina. EIDIPA+UNC 2019. Tercer encuentro interdisciplinario de investigaciones en problemáticas ambientales de la Universidad Nacional de Córdoba.

48. Darsie Jr RF. Mosquitoes of Argentina. I. Keys for identification of adult females and fourth stage larvae in English and Spanish (Diptera, Culicidae). Mosq Syst. 1985; 17: 153-253.

49. Fay RW, Eliason DA. A preferred oviposition site as a surveillance method for *Aedes aegypti*. Mosq News. 1966; 26: 531–535.

50. Reiter P, Amador MA, Colon N. Enhancement of the CDC ovitrap with hay infusions for daily monitoring of *Aedes aegypti* populations. J Am Mosq Control Assoc. 1991; 7: 52-55.

51. de la Casa A, Nasello O. Breakpoints in annual rainfall trends in Córdoba, Argentina. Atmos Res. 2010; 95: 419–427.

52. Ministerio de Salud de Nación. Boletines Epidemiologicos; 2009-2019 [cited 2018 Apr 28]. Available from: https://www.argentina.gob.ar/salud/epidemiologia/boletinesepidemiologicos.

53. RStudio Team (2015). RStudio: Integrated Development for R. RStudio, Inc., Boston, MA URL http://www.rstudio.com/.

54. Moritz S, Bartz-Beielstein T. ImputeTS: time series missing value imputation in R. R J. 2017; 9: 207–218.

55. Grolemund G, Wickham H. Dates and times made easy with lubridate. J Stat Softw. 2011; 40: 1–25.

56. Bates MD, Venables B. Team MRC. Package 'splines.' R Version. 2011.

57. Chan K-S, Ripley B. TSA: time series analysis. R package version 1.01; 2012 [cited 2018]. Available from: https://CRAN.R-project.org/package=TSA

58. Halekoh U, Højsgaard S, Yan J. The R package geepack for generalized estimating equations. J Stat Softw. 2006; 15: 1–11.

59. Venables WN, Ripley BD. Modern applied statistics with S-PLUS. 4th ed. New York: Springer Science & Business Media; 2013.

60. Sippy R, Herrera D, Gaus D, Gangnon RE, Patz JA, Osorio JE. Seasonal patterns of dengue fever in rural Ecuador: 2009-2016. PLoS Negl Trop Dis. 2019; 13: e0007360.

61. Stoddard ST, Wearing HJ, Reiner RC, Morrison AC, Astete H, Vilcarromero S, et al. Long-term and seasonal dynamics of Dengue in Iquitos, Perú. PLoS Negl Trop Dis. 2014; 8: e3003.

62. Eastin MD, Delmelle E, Casas I, Wexler J, Self C. Intra-and interseasonal autoregressive prediction of dengue outbreaks using local weather and regional climate for a tropical environment in Colombia. Am J Trop Med Hyg. 2014; 91: 598–610.

63. Stewart-Ibarra AM, Lowe R. Climate and non-climate drivers of dengue epidemics in southern coastal Ecuador. Am J Trop Med Hyg. 2013; 88: 971–981.

64. Stewart Ibarra AM, Munoz AG, Ryan SJ, Borbor MJ, Ayala EB, Finkelstein JL, et al. Spatiotemporal clustering, climate periodicity, and social-ecological risk factors

for dengue during an outbreak in Machala, Ecuador, in 2010. BMC Infect Dis. 2014; 14: 610.

65. Mordecai EA, Caldwell JM, Grossman MK, Lippi CA, Johnson LR, Neira M, et al. Thermal biology of mosquito-borne disease. Ecol Lett. 2019; 22: 1690–708.
66. Grech MG, Sartor PD, Almirón WR, Ludueña-Almeida FF. Effect of temperature on life history traits during immature development of *Aedes aegypti* and *Culex quinquefasciatus* (Diptera: Culicidae) from Córdoba city, Argentina. Acta Trop. 2015; 146: 1-6.
67. Centenario de la Reforma Universitaria. La UNC en cifras. [cited 2019 Oct 8]. Available from: https://centenariodelareforma.unc.edu.ar/la-unc-en-cifras/.
68. Telediario Digital. Población estudiantil: bajo la lupa de la UNC. [cited 2019 Oct 8]. Available from: https://www.telediariodigital.net/2014/11/poblacion-estudiantil-bajo-la-lupa-de-la-unc/.
69. Honório N, Codeço C, Alves F, Magalhães M, Lourencco-de-Oliveira R. Temporal distribution of *Aedes aegypti* in different districts of Rio de Janeiro, Brazil, measured by two types of traps. J Med Entomol. 2009; 46: 1001–1014.
70. Messina JP, Brady OJ, Scott TW, Zou C, Pigott DM, Duda KA, et al. Global spread of dengue virus types: mapping the 70 year history. Trends Microbiol. 2014; 22: 138–46.
71. Radke EG, Gregory CJ, Kintziger KW, Sauber-Schatz EK, Hunsperger EA, Gallagher GR, et al. Dengue outbreak in key west, Florida, USA, 2009. Emerg Infect Dis. 2012; 18: 135-137.

72. Lourenço J, Recker M. The 2012 Madeira Dengue outbreak: epidemiological determinants and future epidemic potential. PLoS Negl Trop Dis. 2014; 8: e3083.

73. Ritchie SA, Long S, Smith G, Pyke A, Knox TB. Entomological investigations in a focus of dengue transmission in Cairns, Queensland, Australia, by using the sticky ovitraps. J Med Entomol. 2004; 41: 1–4.

74. Barrera R, Amador M, Acevedo V, Caban B, Felix G, Mackay AJ. Use of the CDC autocidal gravid ovitrap to control and prevent outbreaks of *Aedes aegypti* (Diptera: Culicidae). J Med Entomol. 2014; 51: 145–154.

75. Global framework for climate services. Health Exemplar to the user interface platform of the Global Framework for Climate Services. Geneva, Switzerland: World Meteorological Association; 2014 [cited 2018 Nov 18]. Available from: http://www.gfcs-climate.org/sites/default/files/Priority-Areas/Health/GFCS-HEALTH-EXEMPLAR-FINAL-14152_en.pdf.

76. Lowe R, Stewart-Ibarra AM, Petrova D, García-Díez M, Borbor-Cordova MJ, Mejía R, et al. Climate services for health: predicting the evolution of the 2016 dengue season in Machala, Ecuador. Lancet Planet Health. 2017; 1: e142–e151.

77. Huber JH, Childs ML, Caldwell JM, Mordecai EA. Seasonal temperature variation influences climate suitability for dengue, chikungunya, and Zika transmission. PLoS Negl Trop Dis. 2018; 12: e0006451.

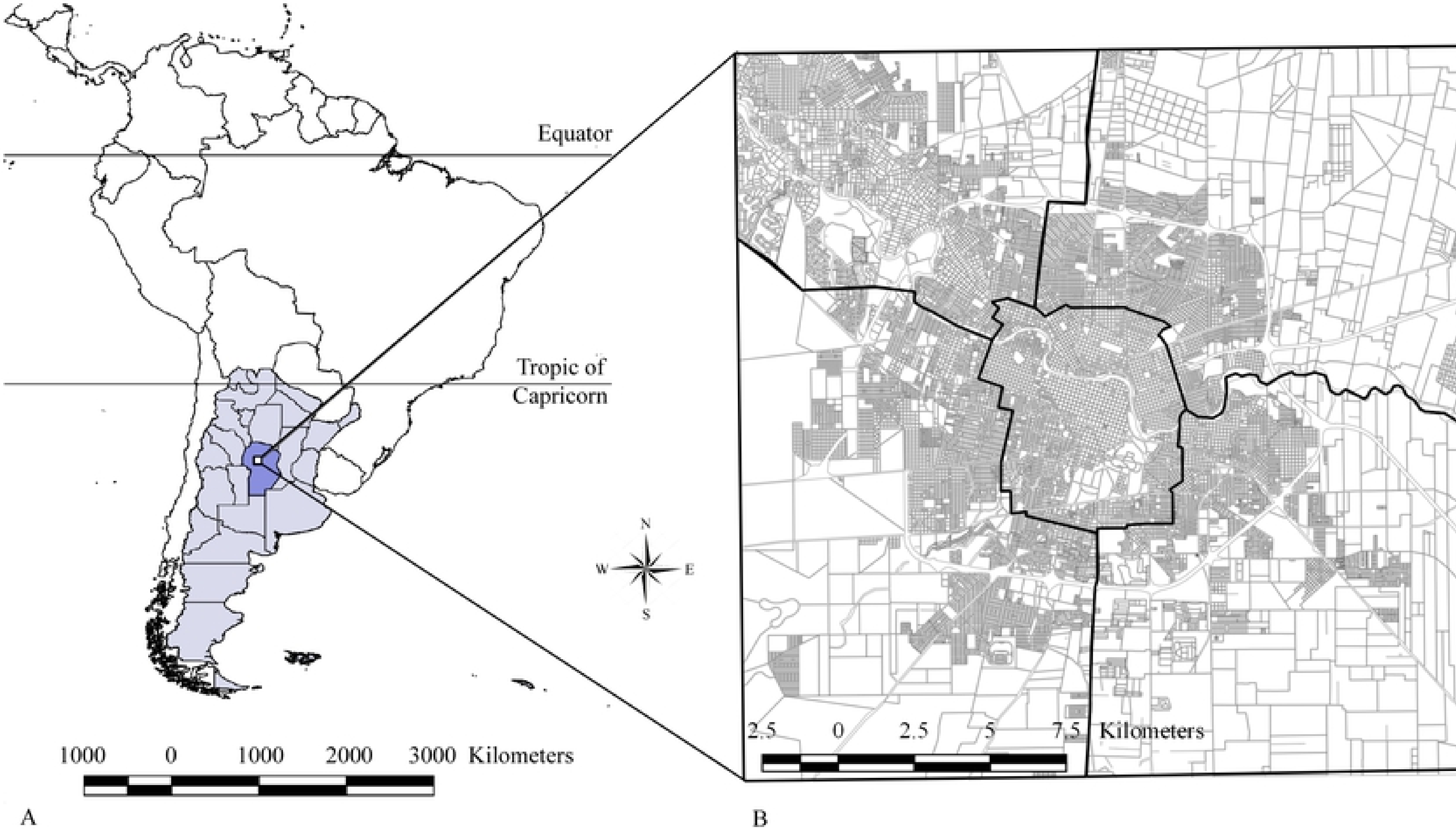
78. Robert MA, Christofferson RC, Silva NJ, Vasquez C, Mores CN, Wearing HJ. Modeling mosquito-borne disease spread in US urbanized areas: The case of Dengue in Miami. PLoS One. 2016; 11: e0161365.

79. Butterworth MK, Morin CW, Comrie AC. An analysis of the potential impact of climate change on dengue transmission in the southeastern United States. Environ Health Perspect. 2016; 125: 579–585.

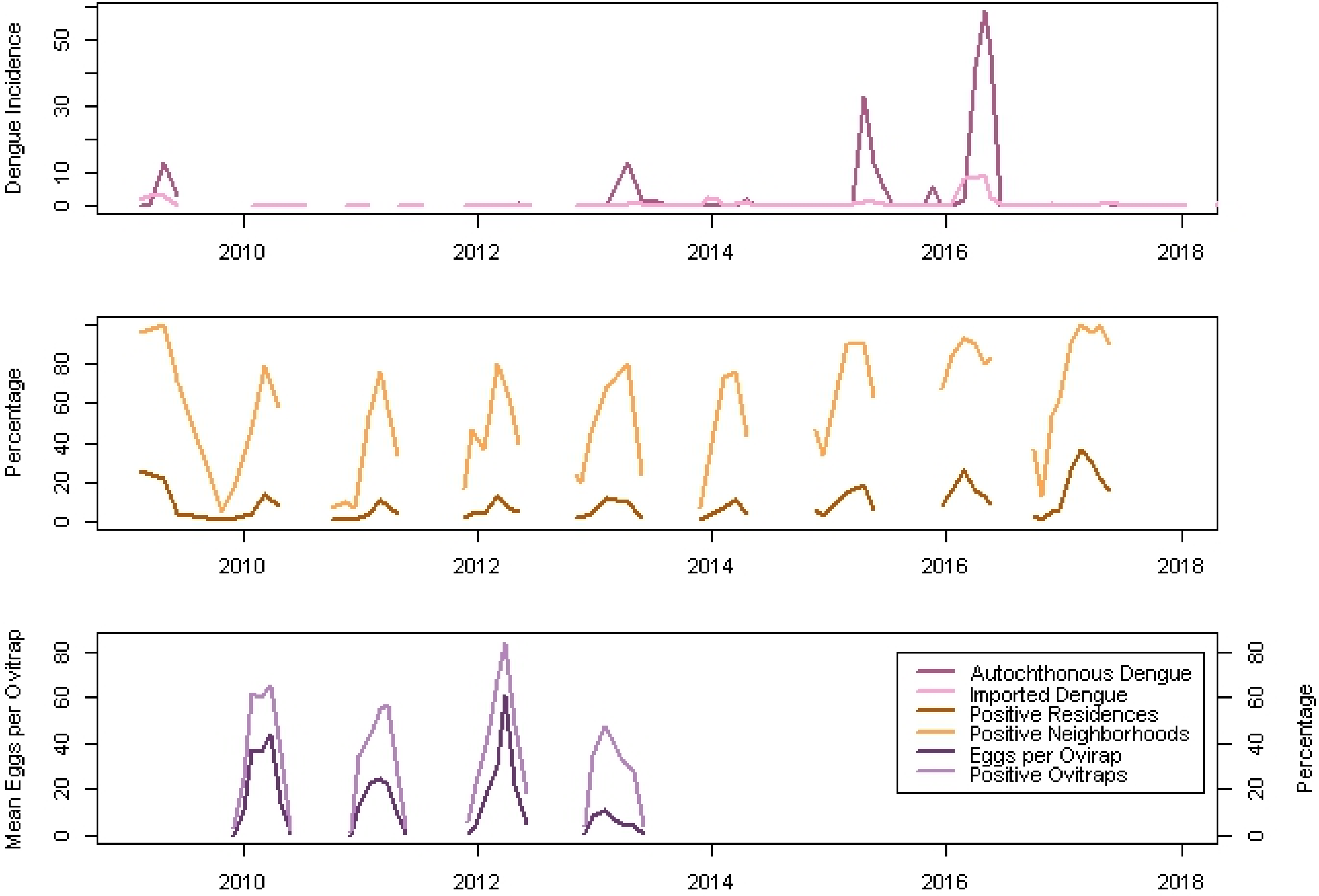
80. Ryan SJ, Carlson CJ, Mordecai EA, Johnson LR. Global expansion and redistribution of *Aedes*-borne virus transmission risk with climate change. PLoS Negl Trop Dis. 2019; 13: e0007213.

Supplemental Table 1. Summary Statistics of Climate, *Aedes aegypti*, and Dengue Measures. Mean and range by year are given for climate, *Aedes aegypti*, and dengue measures used in this study.

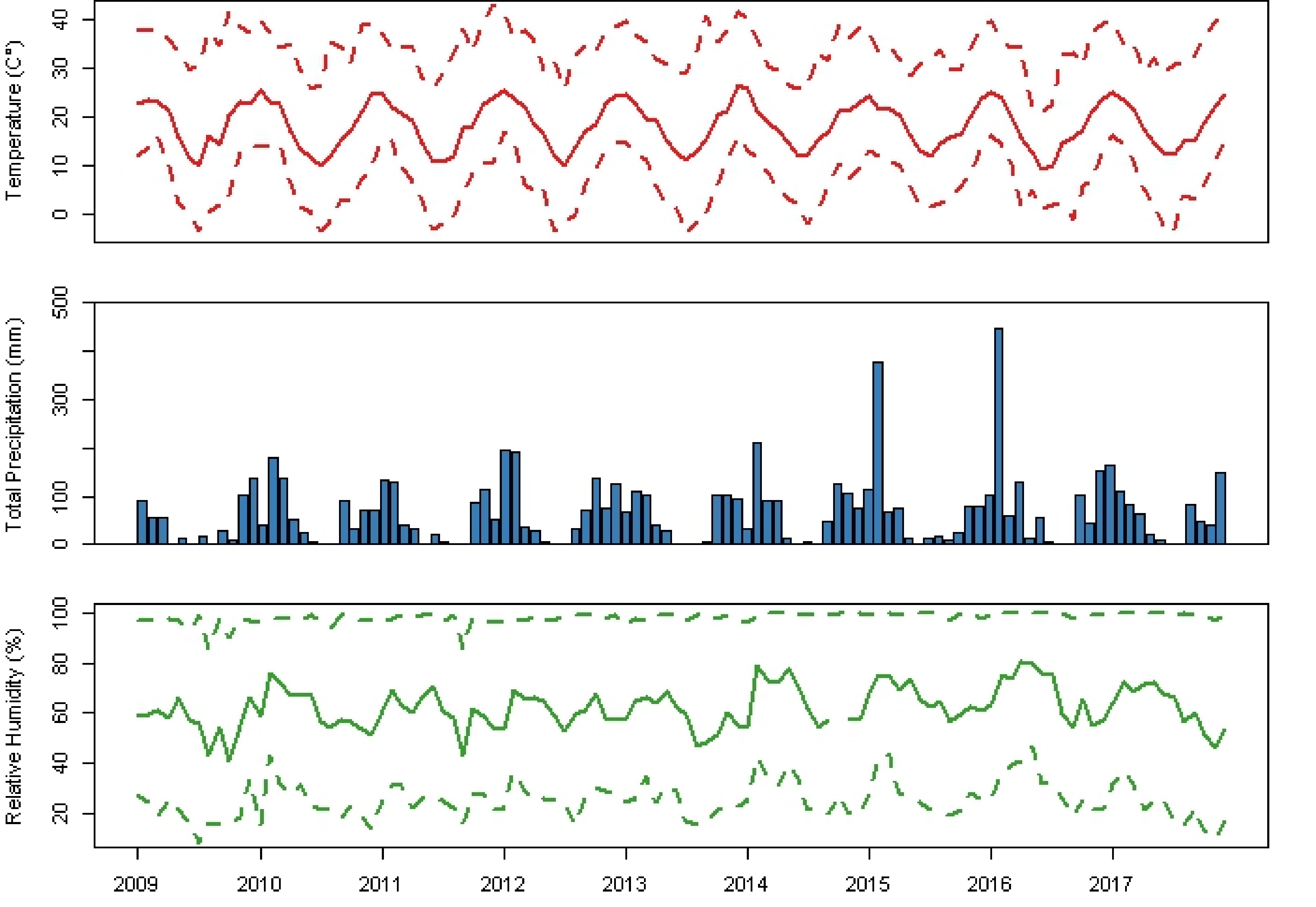
Supplemental Figure 1. Location of 177 sites where ovitraps were placed to collect *Aedes aegypti* eggs in the city of Córdoba, from November 2009 to October 2013.


Equator
Tropic of
Capricorn
1000 0 1000 2000 3000 Kilometers
A
N
W
E
S
2.5 0 2.5 5 7.5 Kilometers
B

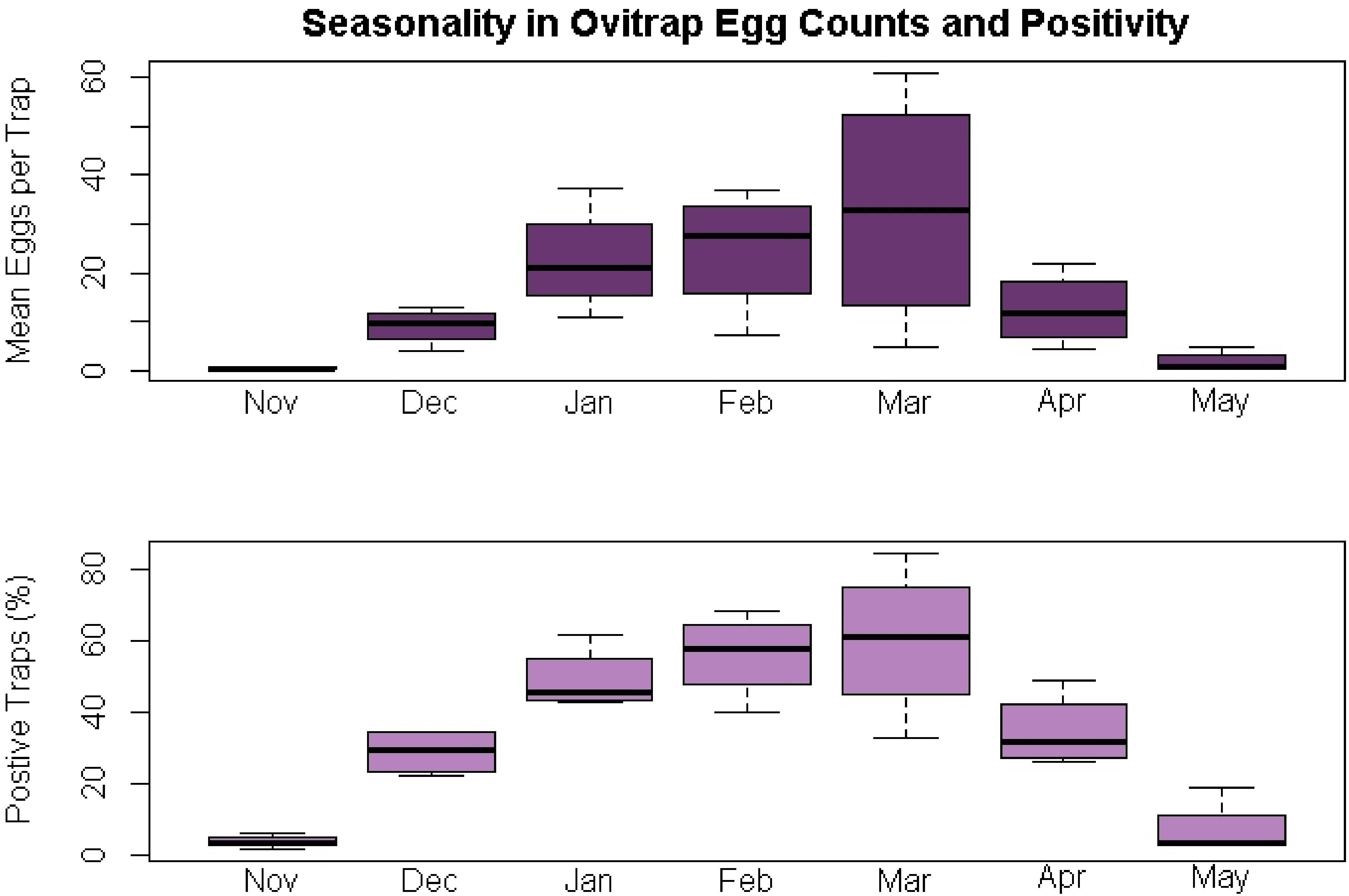
Figure


Dengue Incidence
0
10
30
50
2010
2012
2014
2016
2018
Percentage
0
20
40
60
80
2010
2012
2014
2016
2018
Mean Eggs per Ovitrap
0
20
40
60
80
Autochthonous Dengue
Imported Dengue
Positive Residences
Positive Neighborhoods
Eggs per Ovirap
Positive Ovitraps
0
20
40
60
80
Percentage
2010
2012
2014
2016
2018

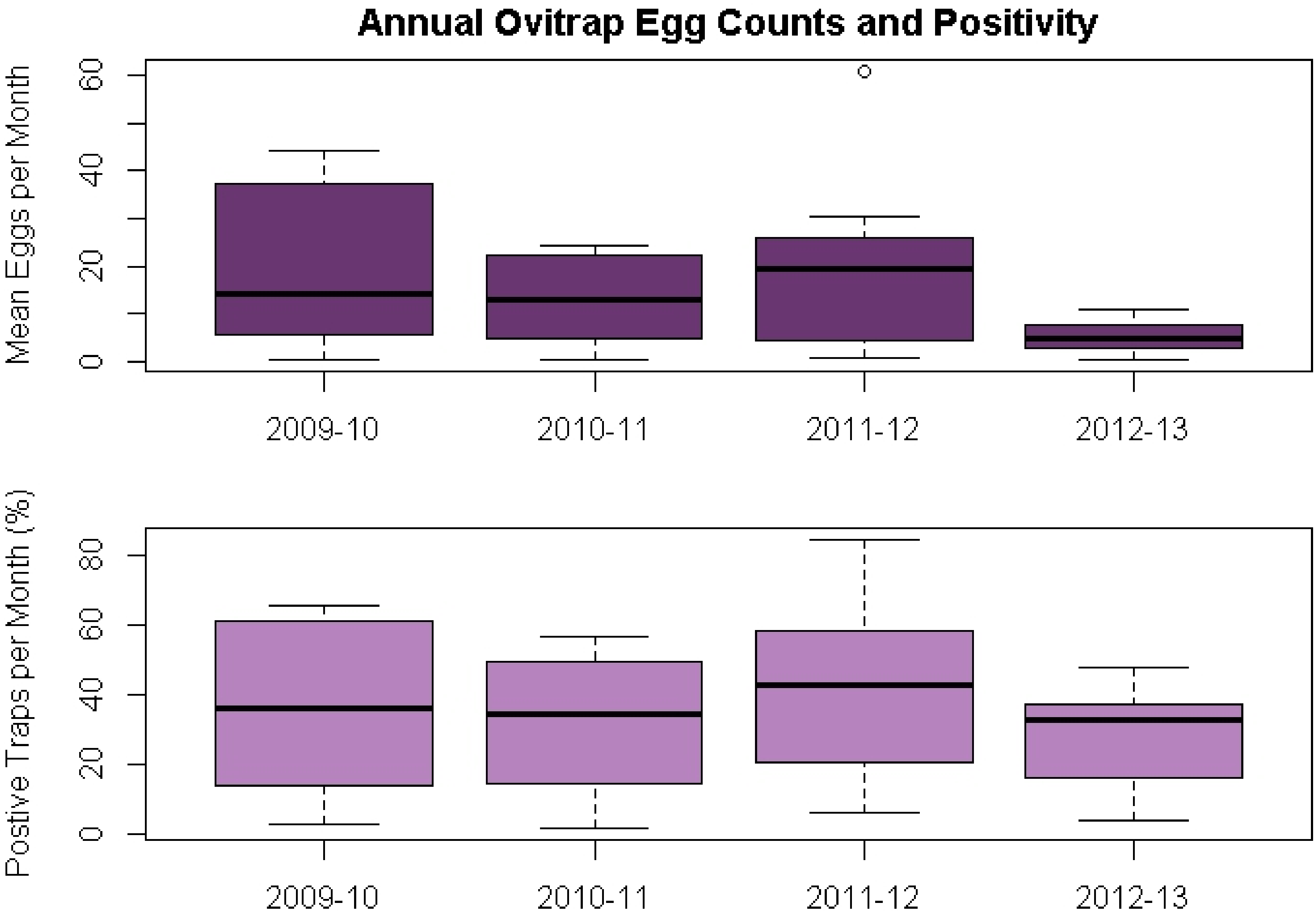
Figure


Temperature (C°)
0
10
20
30
40
Total Precipitation (mm)
0
100
300
500
Relative Humidity (%)
20
40
60
80
100
2009
2010
2011
2012
2013
2014
2015
2016
2017

Figure

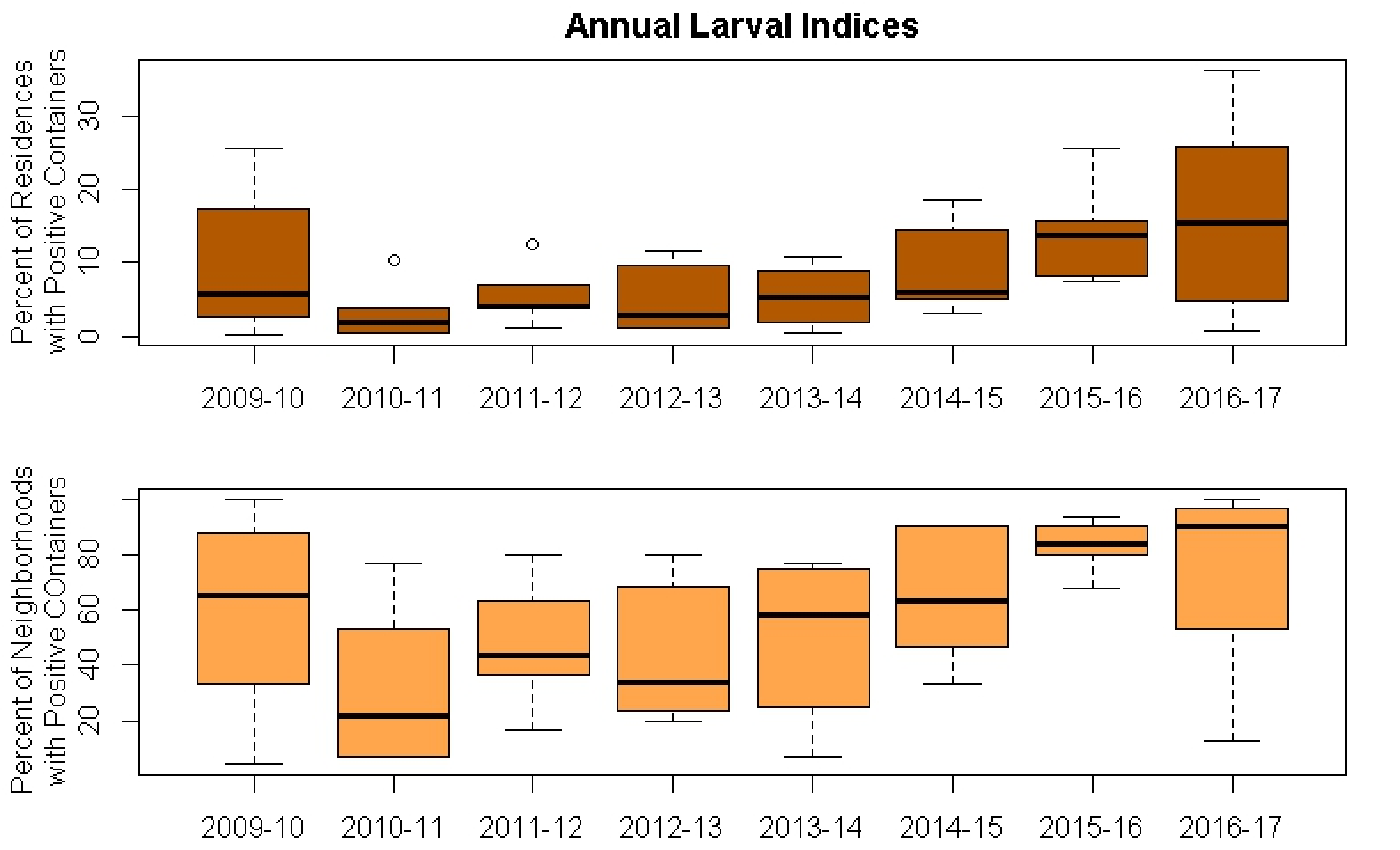

Seasonality in Ovitrap Egg Counts and Positivity
Mean Eggs per Trap
0
20
40
60
Nov
Dec
Jan
Feb
Mar
Apr
May
Postive Traps (%)
0
20
40
60
80
Nov
Dec
Jan
Feb
Mar
Apr
May

Figure

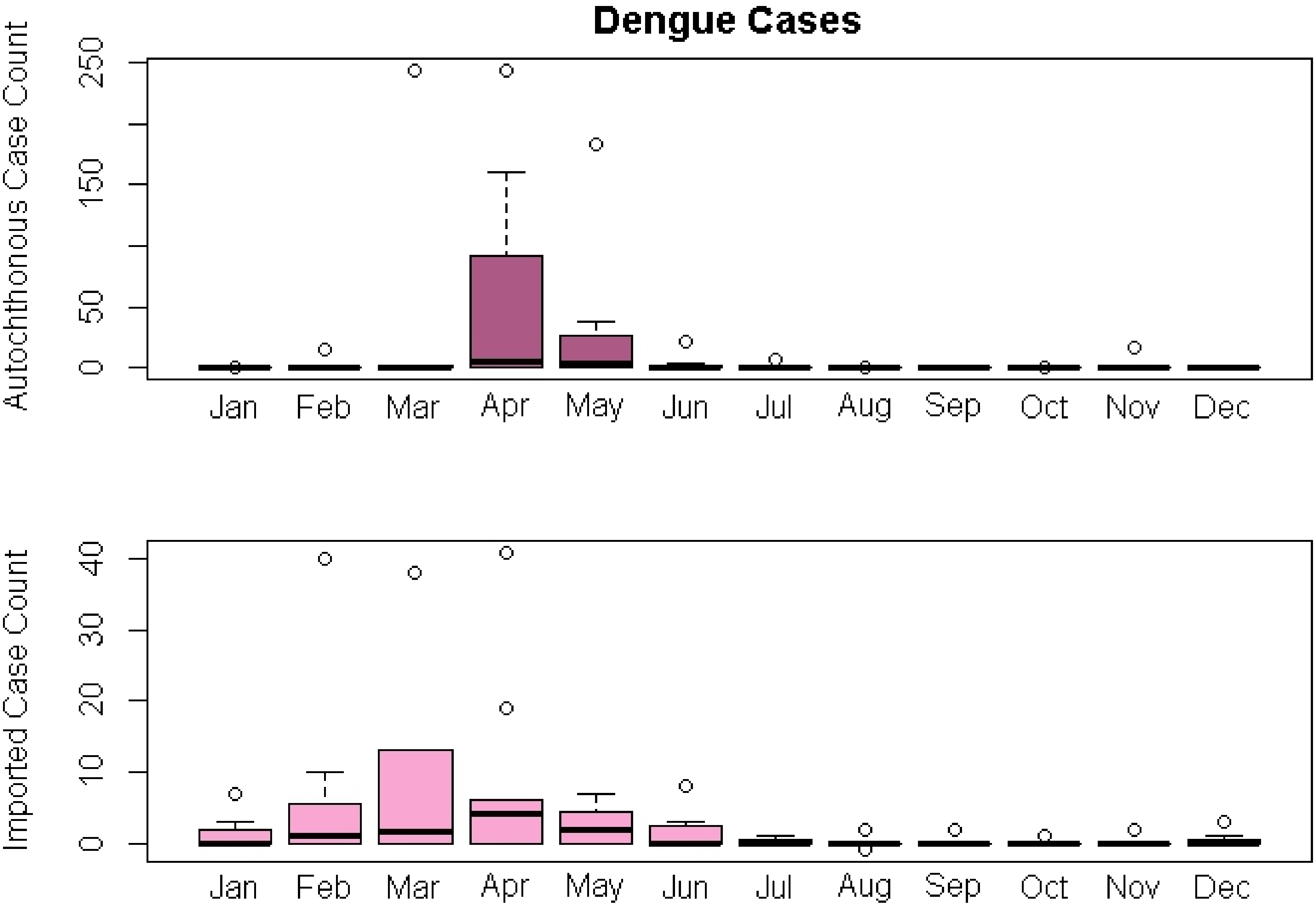


Annual Ovitrap Egg Counts and Positivity
Mean Eggs per Month
0
20
40
60
2009-10
2010-11
2011-12
2012-13
Postive Traps per Month (%)
0
20
40
60
80
2009-10
2010-11
2011-12
2012-13

Figure

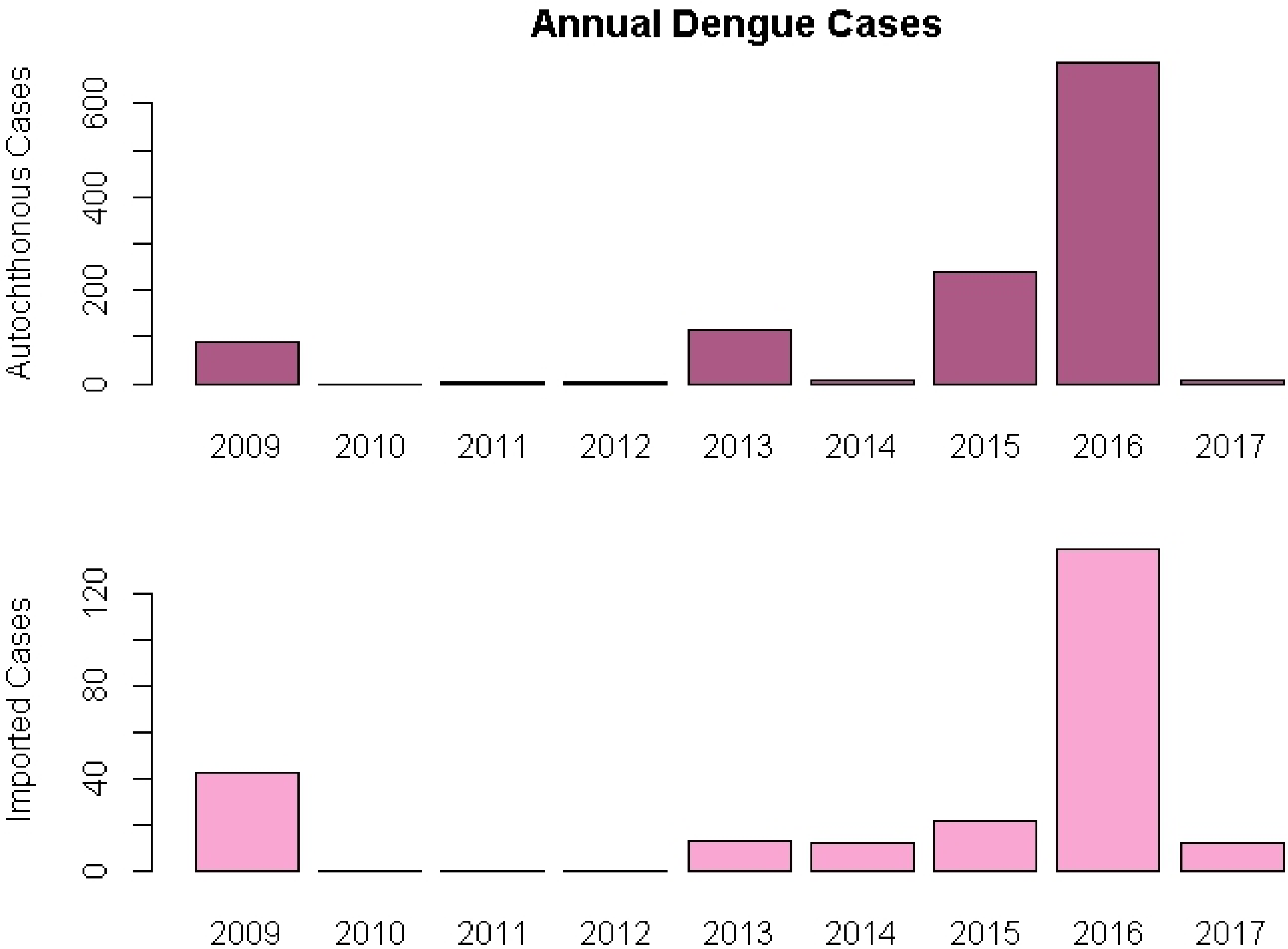

Annual Larval Indices
Percent of Residences
with Positive Containers
0
10
20
30
2009-10
2010-11
2011-12
2012-13
2013-14
2014-15
2015-16
2016-17
Percent of Neighborhoods
with Positive COntainers
20
40
60
80
2009-10
2010-11
2011-12
2012-13
2013-14
2014-15
2015-16
2016-17

Figure



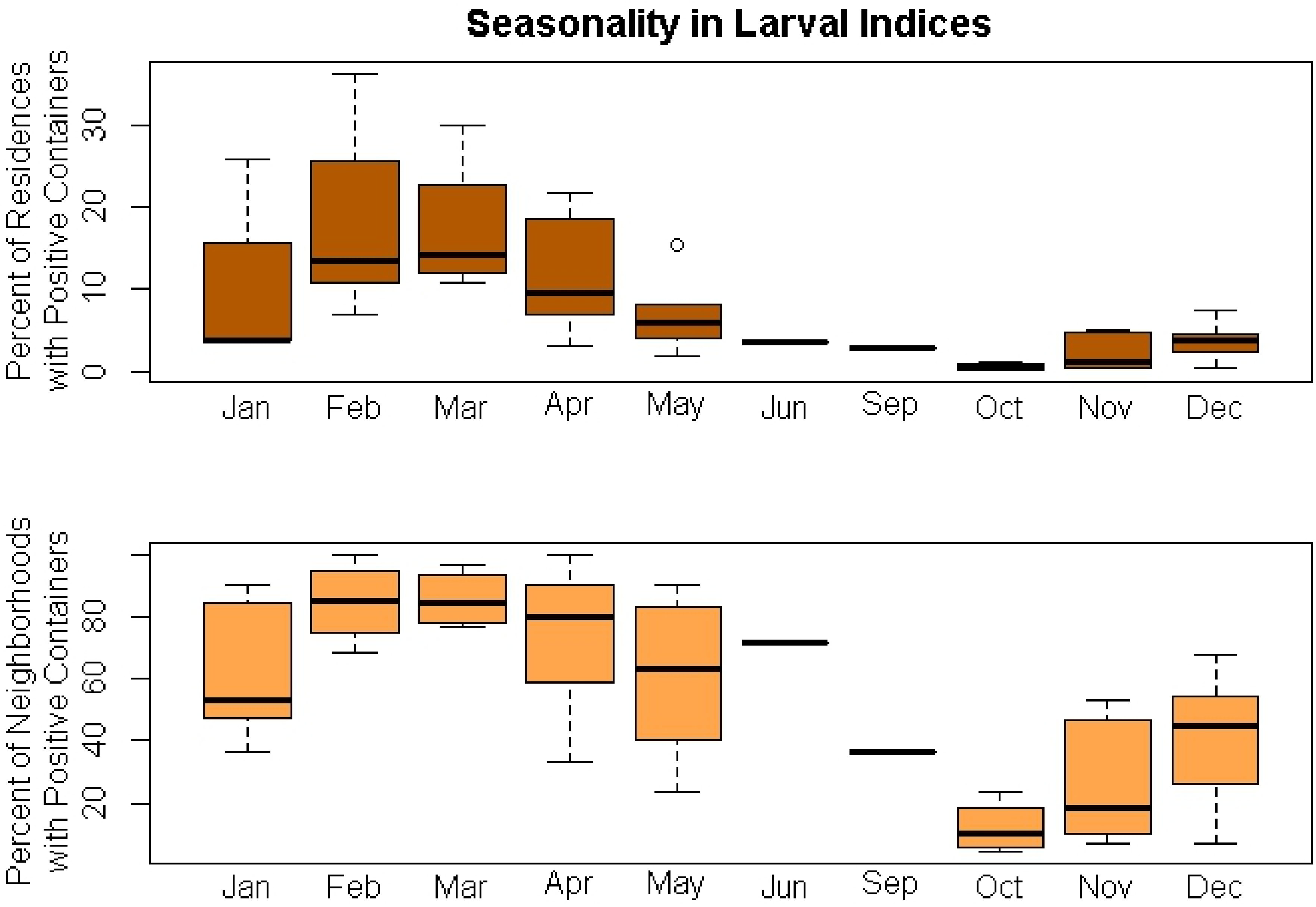
Dengue Cases
Autochthonous Case Count
0
50
150
250
Jan
Feb
Mar
Apr
May
Jun
Jul
Aug
Sep
Oct
Nov
Dec
Imported Case Count
0
10
20
30
40
Jan
Feb
Mar
Apr
May
Jun
Jul
Aug
Sep
Oct
Nov
Dec

Figure



Annual Dengue Cases
Autochthonous Cases
0
200
400
600
2009
2010
2011
2012
2013
2014
2015
2016
2017
Imported Cases
0
40
80
120
2009
2010
2011
2012
2013
2014
2015
2016
2017

Figure



Seasonality in Larval Indices
Percent of Residences with Positive Containers
0
10
20
30
Jan
Feb
Mar
Apr
May
Jun
Sep
Oct
Nov
Dec
Percent of Neighborhoods with Positive Containers
20
40
60
80
Jan
Feb
Mar
Apr
May
Jun
Sep
Oct
Nov
Dec

Figure