Acta Tropica 146 (2015) 53–59

Contents lists available at ScienceDirect

Acta Tropica

journal homepage: www.elsevier.com/locate/actatropica

St. Louis Encephalitis virus mosquito vectors dynamics in three different environments in relation to remotely sensed environmental conditions

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ARTICLE INFO

Article history:
Received 11 October 2014
Received in revised form 5 March 2015
Accepted 7 March 2015
Available online 16 March 2015

Keywords:
SLEV vectors
Culex quinquefasciatus
Culex interfor
Mosquito population dynamic
Remote sensing

ABSTRACT

In Argentina the St. Louis Encephalitis virus (SLEV) is an endemic and widely distributed pathogen transmitted by the cosmopolitan mosquito *Culex quinquefasciatus*. During two outbreaks in Córdoba city, in 2005 and 2010, *Culex interfor* was also found infected, but its role as vector of SLEV is poorly known. This mosquito species is distributed from central Argentina to southern Brazil. The primary aim of this study was to analyze the population dynamic of *Cx. interfor* and *Cx. quinquefasciatus* in three different environments (urban, suburban and non-urban) in relation to remotely sensed environmental data for vegetation (NDVI and NDWI) and temperature (brightness temperature). *Cx. quinquefasciatus* and *Cx. interfor* were found at the three sampled sites, being both the most abundant *Culex* species, with peaks in early and midsummer. Temporal distribution patterns of both mosquito species were highly correlated in a non-urban area of high SLEV risk transmission. *Cx. quinquefasciatus* and *Cx. interfor* were associated with the most urbanized site and the non-urban environment, respectively; high significant correlations were detected between vegetation indices and abundance of both mosquito species confirming these associations. These data provide a foundation for building density maps of these two SLEV mosquito vectors using remotely sensed data to help inform vector control programs.

1. Introduction

Argentina is affected by the emergence and re-emergence of diseases caused by flaviviruses, such as dengue (Estallo et al., 2014), yellow fever (Morales et al., 2011) and human encephalitis by St. Louis Encephalitis virus (SLEV) (Spinsanti et al., 2003) and West Nile virus (Diaz et al., 2011). The SLEV is maintained in nature through biological transmission between birds and *Culex* mosquitoes. Humans are considered dead-end hosts that can suffer from febrile headache to meningoencephalitis and even death (Reisen, 2003). Isolated human cases by SLEV were reported in 1964, 1968, 1971 and 1985 in Argentina (Sabattini et al., 1985). Later, after 17 years of absence of human cases, SLEV reemerged in the central region of the country (i.e., Córdoba province) in 2002 (Spinsanti et al., 2003). In Córdoba, two outbreaks by SLEV were reported in 2005 and 2010 with 47 and 11 individuals infected with this virus, respectively, mostly with neurologic involvement (Spinsanti et al., 2008; Vergara Cid et al., 2011). The isolation of SLEV from *Culex quinquefasciatus* collected in nature at the provinces of Córdoba and Santa Fe (Mitchell et al., 1985; Diaz et al., 2006) and experimental vector competence studies (Diaz, 2009; Flores et al., 2010) indicate that this species is involved as the urban vector in Argentina. *Cx. quinquefasciatus* is widely distributed in America, from United States of America to the central region of Argentina (WRBU, 2014). On the other hand, during the 2005 and 2010 outbreaks, *Culex interfor* was also found infected in Córdoba city with SLEV (Diaz et al., 2006). *Cx. interfor* is present in several provinces

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http://dx.doi.org/10.1016/j.actatropica.2015.03.009
0001-706X/

in northern and central Argentina, probably extending into Bolivia and southern Brazil (Dibo et al., 2011; Diez et al., 2011; Mureb Sallum et al., 1996; Rossi et al., 2006; Visintin et al., 2009). However, its role as vector and several aspects of its biology remains largely unknown. Therefore, further investigations regarding vector population dynamics are needed.

Since there is no treatment or vaccine against SLEV, entomological surveillance and mosquito control remain the main public health strategies to prevent human infection. A deeper knowledge about variables affecting vector distribution and abundance leads to the improvement of disease control measures. Environmental alteration by human activities often influences over vector populations dynamics. Deforestation and urbanization increase the proliferation of artificial containers for developing larvae urban mosquito species (Norris, 2004). Certain species are more abundant in urban areas according to landscape characteristics, as *Culex maxi* and *Culex apicinus* that showed opposite patterns depending on the proximity to waterways in Córdoba city (Argentina); *Cx. maxi* abundance was higher near to waterways as the percentages of un-built lots increased (Gleiser and Zalazar, 2009).

The remotely sensed data provides a moderate-cost alternative approach to mapping vector species distribution (Kalluri et al., 2007; Wilschut et al., 2013). Satellite imagery may also improve the efficiency of control programs for mosquitoes, by indicating areas or periods of the year under higher risk of mosquito proliferation (Gleiser and Gorla, 2007). Several authors have observed that the spatial and temporal patterns of mosquito population dynamics are controlled by environmental factors that can be remotely observed (Brown et al., 2008; Chuang et al., 2012; Estallo et al., 2008, 2012). In epidemiology, the use of remote sensing data involves the study of environmental variables that characterize the vector ecosystem such as land use, temperature, vegetation cover and precipitation (Johnson et al., 2008; Tran et al., 2002). The normalized difference vegetation index (NDVI) and the normalized difference water index (NDWI) are two vegetation indices derived from satellite images that can be used as indicative of potential mosquito habitat existence (Brown et al., 2008). Thus, the use of remotely sensed data could aid in detecting foci of virus activity by identifying environmental conditions suitable for SLEV vectors in both urban and nonurban areas. In Argentina, proximity to vegetated areas with NDVI > 0.3 and the presence of low density of urban construction were the main landscape elements that contributed to human infections in the last SLEV outbreak in 2010 in Cordoba city (Vergara Cid et al., 2013). Therefore, this study evaluated the temporal distribution of the vector of SLEV *Cx. quinquefasciatus* and the potential vector *Cx. interfor* in three different environments in relation to remotely sensed environmental data for vegetation and temperature in Córdoba city, Argentina.

2. Materials and methods

2.1. Study area and mosquito collections

Córdoba city (Córdoba Province) has a population of approximately 1,330,000 inhabitants (INDEC, 2010) and is located in the central region of Argentina (64°11′W–31°24′S), at 450 m above sea level. Climate is temperate with mean annual temperature ranging between 16 °C and 17 °C, with a warm and wet season between September and May, and mean annual rainfall of 800 mm (Jarsún et al., 2003).

Adult mosquitoes were collected using CDC CO_2 light-baited traps (three per site) in three areas of Córdoba city: San Vicente (SV), Camino 60 Cuadras (CC) and Bajo Grande (BG). These sampling sites (each separated by >8 km) were selected to represent different environments: SV is urban (SV), CC is suburban and BG is non-urban. The three areas where sampling sites were located were completely different as seen in Fig. 1. At SV, traps were placed at a nursing home located in the south east of the city; vegetation was dominated by *Melia azedarach* (commonly called "paraíso") and *Pinus* spp.; also ornamental plants and an orchard were present; the nursing home is surrounded by high density of housing and other building constructions. The sampling site is located in an urbanized area characterized by residential, commercial and industrial uses. The suburban site (CC) is located in the south of the city and traps were placed at a family dwelling; vegetation was represented by a mix of autochthonous (*Acacia* spp.) and introduced species (*M. azedarach*, *Pinus* spp., *Ligustrum lucidum*, *Platanus* spp.), surrounding a swimming pool, the house and a fruit tree area. This site is surrounded by some crop areas and is 1 km away from the nearest urbanized settlement. The non-urban site (BG) is located in the eastern of Córdoba, on the outskirts of the city, 2 km away from city urban area and it is characterized by many open areas intended for vegetable and fruit culture. Traps were placed at the sewage treatment plant of the City Hall, which has sewage pool containers and some building constructions, surrounding by ponds with aquatic vegetation, few isolated houses and many open areas. Vegetation is dominated by *M. azedarach*, *Morus alba*, *L. lucidum*, *Pinus* spp., *Ricinus* spp. and grassland.

Traps were placed simultaneously at each sampling site every 2 weeks from 6:00 p.m. to 10:00 a.m. A total of 45 nights of trapping from January 2008 to December 2009 were performed. Captured mosquitoes were frozen in the field and transported to the laboratory. Mosquitoes were then counted and identified to genus and species using the key by Darsie (1985).

2.2. Environmental characterization

Satellite-derived variables were used to characterize and identify suitable environmental conditions for the development of mosquito vectors.

Widely used vegetation indices (NDVI and NDWI) and temperature were obtained from a set of 17 Landsat 5 (L5TM) and 7 (L7 ETM +) path/row 229/82 satellite images (16 days temporal resolution) from January2008 to December 2009. Satellite images were obtained from the Argentine Space Agency (CONAE) catalog by academic cooperation. ENVI (Environment for Visualizing Images, Research Systems) 4.2 software (2004) was used for image processing. The images were geo-referenced using an image from the GLCF (Global Land Cover Facility). Subsequently, a subset of 240 km^2 (800 × 800 pixels) area that included Córdoba city was generated. The images were calibrated to convert Landsat 5 TM or Landsat 7 ETM+ digital numbers to exoatmospheric reflectance (reflectance above the atmosphere) using their respective coefficients (USGS Landsat, 2010).

In order to measure the vegetation coverage and the vegetation water content, we calculated the NDVI and the NDWI, both indices applied for mosquito studies as indicative of potentials mosquito habitat existence (Brown et al., 2008; Estallo et al., 2012, 2008). Values of the earth's surface temperature were estimated through the brightness temperature (BT) (Landsat images band 6), which gives an approximation of the environmental temperature (Estallo et al., 2008, 2012; Kalluri et al., 2007). A buffer zone of 5 × 5 pixels (150 m) was delimitated around each sampling site in order to calculate the values of NDVI, NDWI and BT of each pixel. From each buffer zone the average values were calculated for each environmental variable.

Since there were only 17 satellite images available for the study (January, February, April, June, July, August, October, November 2008; January, March, April, May, June, July, October, November, December 2009), we used linear interpolation to obtain the missing indices values (March, May, September, December 2008 and