OXFORD

# Vegetation Cover and Microspatial Distribution of Sand Flies (Diptera: Psychodidae) in an Endemic Locality for Cutaneous Leishmaniasis in Northern Argentina

Mariana del M. Chanampa,<sup>1</sup> Raquel M. Gleiser,<sup>2,3</sup> Carlos L. Hoyos,<sup>4</sup> Griselda N. Copa,<sup>4</sup> Carolina Mangudo,<sup>1,4</sup> Julio R. Nasser,<sup>4,5</sup> and José F. Gil<sup>1,4,5,6,●</sup>

<sup>1</sup>Instituto de Investigaciones en Energía no Convencional (INENCO-CONICET), Universidad Nacional de Salta, Sector Salud y Ambiente, Av. Bolivia 5150, 4400 Salta, Argentina, <sup>2</sup>Universidad Nacional de Córdoba-CONICET, Instituto Multidisciplinario de Biología Vegetal (IMBIV)-Centro de Relevamiento y Evaluación de Recursos Agrícolas y Naturales (CREAN-IMBIV), 5016 Córdoba, Argentina, <sup>3</sup>Universidad Nacional de Córdoba, Facultad de Ciencias Exactas, Físicas y Naturales, Departamento de Diversidad Biológica y Ecología, 5000 Córdoba, Argentina, <sup>4</sup>Instituto de Investigaciones de Enfermedades Tropicales, Sede Regional Orán, Universidad Nacional de Salta, 4530 San Ramón de la Nueva Orán, Argentina, <sup>5</sup>Cátedra de Química Biológica y Biología Molecular, Facultad de Ciencias Naturales, Universidad Nacional de Salta, 4400 Salta, Argentina, and <sup>6</sup>Corresponding author, e-mail: jgil@ conicet.gov.ar

Subject Editor: David Florin

Received 30 August 2017; Editorial decision 13 July 2018

## Abstract

The sand fly fauna in Hipólito Yrigoyen, Argentina, a locality where cutaneous leishmaniasis cases occur, was surveyed with zones of higher abundance of sand flies correlated to vegetation cover estimated through normalized difference vegetation index (NDVI). Sand flies were collected with 10 CDC traps during six nights, from December 2009 to January 2010. A map was built of expected sand flies abundance in which levels of NDVI were categorized. In total, 1,392 Phlebotominae (Diptera: Psychodidae) specimens were collected, comprised of the following species: *Nyssomyia neivai* (Pinto 1926), *Migonemyia migonei* (França 1920), species of the *cortelezzii* complex (Brèthes 1923), *Evandromyia sallesi* (Galvão & Coutinho 1940), and *Psathyromyia shannoni* (Dyar 1929). Positive correlations were found between the abundance of sand flies and the NDVI (P < 0.05) for buffer areas of <150 m radii from the trap location points, i.e., the sand fly abundance was greater where vegetation cover and density were greater. In this context, plant cover should be taken into account to prioritize surveillance and control areas within the program of sand flies control in northern Argentina.

Key words: phlebotomine, leishmaniasis, normalized difference vegetation index

The Psychodidae family, subfamily Phlebotominae has a global distribution, and includes species of medical interest, which are vectors of different types of leishmaniasis (Salomón 2009). Among the species described for Argentina, eight are found in the province of Salta: Nyssomyia neivai (Pinto, 1926), Migonemyia migonei (França, 1920), Psathyromyia shannoni (Dyar, 1929), Psathyromyia punctigeniculata (Floch & Abonnenc, 1944), Evandromyia cortelezzii (Brèthes, 1923), Evandromyia sallesi (Galvão & Coutinho, 1940), Lutzomyia longipalpis (Lutz & Neiva, 1912), and Micropygomyia quinquefer (Dyar, 1929) (Salomón et al. 2001, 2004, 2008; Barroso et al. 2007; Salomón 2009; Bravo et al. 2013; Krolewiecki et al. 2013; Quintana et al. 2013).

Originally associated with forested areas that had dense vegetation, the transmission cycle of cutaneous leishmaniasis (CL) has changed, to now also being associated with urbanized environments in many countries in Latin America (Walsh et al. 1993, Campbell-Lendrum et al. 2001, Yadon et al. 2003). Yet, sand flies inhabiting urban environments have still been associated with 'green spaces' (Mikery Pacheco et al. 2015) and peridomiciles (Ontivero et al. 2018), where vegetation presumably provides refuge and acts as 'islands' through which these insects can disperse to feed and reproduce. Females of most sand fly species oviposit in cool, moist habitat with decaying debris where their larvae develop, and thus they are more likely found in moist and shadowy places like those offered by trees (e.g., holes, barks, fallen trunks) (Kravchenko et al. 2004, Queiroz et al. 2012).

The spatial or temporal variation in regional or local vegetation cover, estimated through the normalized difference vegetation index (NDVI), has been used as a 'proxy' for environmental factors associated with some vector populations and diseases (Tourre

<sup>©</sup> The Author(s) 2018. Published by Oxford University Press on behalf of Entomological Society of America. All rights reserved. For permissions, please e-mail: journals.permissions@oup.com.

et al. 2008, Cerbino Neto et al. 2009, Gleiser 2016). In a rural area within an Atlantic Forest-dominated locality in Pernambuco, Brazil, *Nyssomyia whitmani* (Antunes & Coutinho, 1939) abundance was positively associated with the NDVI (Donalisio et al. 2012). Plants absorb much of the visible light and reflect almost all the radiation in the infrared spectrum. The reflected radiation is exploited using remote sensing to highlight the differences in absorption and reflectivity in the ground cover. This allows building the NDVI and estimation of the spatial variation of vegetation cover. Since this index only considers differences between the visible and infrared spectrum, it does not allow to differentiate between plant species or vegetation types. Furthermore, the fact that NDVI is an indicator of vegetation cover and density, it indirectly provides information of a series of conditions such as temperature, humidity, refuge, and resources, such as food availability (Gleiser 2016).

Hipólito Yrigoyen is a city of approximately 10,300 inhabitants (Instituto Nacional de Estadísticas y Censos de la República Argentina (INDEC 2010), in the northwest of Argentina. It is located near National Highway Number 50, an important route of commercial transport and communication between northwest Argentina with Bolivia. Every year there are cases of CL along the swath associated with this highway (Hoyos et al. 2016), yet there are no formal studies of the sand fly fauna in Hipólito Yrigoyen. This work describes the sand fly fauna in Hipólito Yrigoyen and its spatial distribution within the urban-wild area. We chose NDVI as a surrogate ('proxy') to vegetation because of the relative ease to obtain indirectly holistic variables that have been shown to be useful to map or describe seasonal fluctuations in several insects of medical relevance (see references in Gleiser 2016).

## **Materials and Methods**

#### Study Area

Hipólito Yrigoyen (Department of Orán, Salta province; 23°14'S -64°16'W; Fig. 1) is 285 m above sea level and comprises an expanse of approximately 206 km<sup>2</sup>. Founded in 1948, it is considered the youngest city of Salta province. The area is included within the limits of the Yungas Man and Biosphere Reserve of UNESCO (Lomáscolo et al. 2010). The Yungas are high biodiversity biomes, subtropical montane moist or rain forests on the eastern slopes of the southern Andes (Brown et al. 2005, Brooks 2018) that are characterized by a strong altitudinal gradient. Hipólito Yrigoyen is located in the Pedemontane rain forest floor, the climate is subtropical, with average maximum and minimum temperatures of 32.4°C (January) and 9.1°C (July), respectively, and mean annual rainfall of 1,000 mm. The locality has been subjected to ecological modifications related to human activities, mainly urbanization, agriculture, and forestry, and has important sugar cane production activities. To the north and east of the city border, it is covered by modified natural vegetation, while to the west and south there is mostly sugar cane farming. The dominant vegetation in the remnant patches of wild forest is represented by the following trees: Phyllostylon rhamnoides (J. Poiss.) Taub., Calycophyllum multiflorum Griseb. (Castelo), Patagonula americana L., Anadenanthera macrocarpa (Benth.) Brenan, Parapiptadenia excelsa (Griseb.) Burkart,

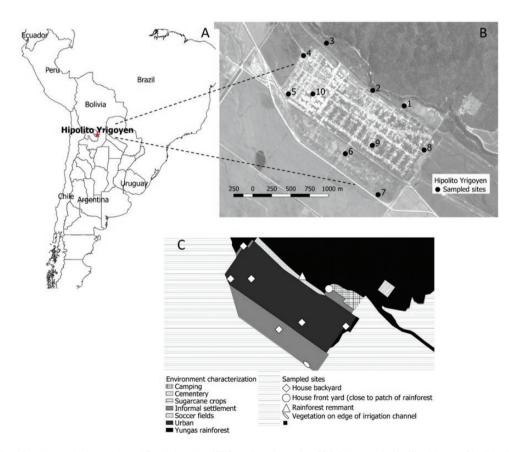


Fig. 1. (A) Location of study area in Argentina and South America. (B) Sampling sites using CDC light traps in Hipólito Yrigoyen Municipality, Salta province, Argentina. (C) Environmental characteristics of the study area.

Tipuana tipu (Benth.) Kuntze, Myroxylon peruiferum L.f., Cedrela angustifolia Sessé & Moc. ex DC., Myracrodruon urundeuva F.F. & M.F. Allemão, Tabebuia avellanedae Lor. ex Griseb., Chlorophora tinctoria (L.) Gaud., Enterolobium contortisiliquum (Vell.) Morong, Acacia aroma Gillies ex Hook. & Arn., Tessaria integrifolia Ruiz & Pav., Salix humboldtiana Willd., Jacaranda mimosifolia D. Don., and Handroanthus impetiginosus (Mart. ex DC.) Standl. There are also numerous herbs, lianas, and epiphytic plants (Cabrera 1971).

The urban landscape is represented by one-story constructions, with (40–100 m<sup>2</sup>) backyards and smaller front yards. Vegetated zones consisting of lawns/grasses and scattered trees characterize the house backyards, scattered vacant lots (usually up to 300 m<sup>2</sup> area), and parks or plazas typically expanding 1 ha. The forested zones in the city differ from the overall surrounding natural areas due to a higher prevalence of exotic species and a more open canopy cover. The dominant vegetation in the urban areas is represented by a combination of gardening plants, lawn grasses, and ornamental (native or exotic) trees such as *C. tinctoria* (L.) Gaud., *T. avellanedae* Lor. ex Griseb., *E. contortisiliquum* (Vell.) Morong, *Anadenanthera peregrina* (L.) Speg., *Citrus limon* (L.) Burm. f., *Citrus sinensis* (L.) Osbeck., *Ligustrum lucidum* W.T. Aiton, *Melia azedarach* L., *Persea americana* var. americana, and *Mangifera indica* L.

## Sampling and Processing Specimen

Phlebotominae sampling was carried out from 10 December to 12 December in 2010 and from 4 January to 6 January in 2011 (a total of six nights), at 10 sites (Fig. 1; Table 1). In total, 10 CDC miniature light traps (www.JohnWHock.com) were used simultaneously, one per sampling site, from 7:00 p.m. to 7:00 a.m. so as to collect maximal numbers of these crepuscular and nocturnally active insects (Fuenzalida et al. 2011). The only attractant used was the trap light (without other lure). There is no evidence of species with diurnal activity in the area. The traps were set outdoors hanging from tree branches, the bottom of the trap approximately 1 m above the ground. Captured specimens were preserved in 70% ethanol and subsequently identified to species based on morphological characteristics of cibarius and spermatheca (females) and genitalia (males) (Young and Duncan 1994, Barroso et al. 2007). Selection of sampling sites was done using a grid of  $10 \times 6$  quadrants ( $200 \times 200$  m each approximately). Of these 60 quadrants, 10 were randomly selected to perform the sampling. Within each of the selected quadrants, the collection site was chosen based on the accessibility and intention of the home owners to participate. Coordinates of sampling sites were taken with GPS (Global Positioning System, Garmin eTrex Vista®).

#### Normalized Difference Vegetation Index

An NDVI image was obtained from Landsat 5 (scale 1:250,000, path: 231 and row: 76; product date: 11–19 December 2010; Google Earth Engine: https://explorer.earthengine.google.com/#index) to

assess whether there was an association between the abundance of sand flies and vegetation density, using IDRISI Selva software (Eastman 2012). The NDVI is calculated taking advantage of the differential characteristics of the red band (Band 3) that is given by the absorbance of chlorophyll pigment and the near-infrared band (Band 4) that is highly reflected by the plant materials:

$$NDVI = \frac{Band 4 - Band 3}{Band 4 + Band 3}$$

This index ranges between -1 and 1; high NDVI values correspond to higher vegetation cover and/or vegetation density or biomass.

Circular buffer areas of 50, 100, 150, and 200 m radius around the sampling sites were generated to explore effects of local scale on vegetation-abundance associations. Radii wider than 200 m were not considered so to avoid overlapping between sampling sites. The average NDVI of each circular buffer area was obtained using the tool 'statistic zone' of QGIS software (QGIS 2.18.0-Las Palmas). The projection used was POSGAR 98/Argentina 4, EPGS: 22174.

#### **Statistical Analysis**

To assess if vegetation density and sand flies abundances were related, correlation analysis was carried out between the number of sand flies collected (sum of the number of sand flies for 6 days) and the average NDVI per site (at each spatial scale) using the Spearman's rank correlations test (R 3.3.2; R Development Core Team 2008). Correlation analyses were conducted using the 10 samples sites for each species and for all sand flies grouped together.

To assess whether sand flies abundance was higher near wild vegetation patches, sites were grouped in two categories: 'near' included sites 1–4 and 8 located >50 m from a natural forest patch, and 'far' grouped sites 5–7 and 9–10 located >200 m from a natural green patch. Sand flies abundances (total or per more abundant species) were compared between near and far sites using generalized linear models (GLMs) (Poisson link function).

#### Results

In total, 1,392 sand flies were collected, of which 695 were female. Species collected were: *Ny. neivai* (71.84%), *Mg. migonei* (11.71%), *Ev. sallesi-cortelezzii* complex (10.27%), *Ev. sallesi* (1.58%), *Pa. shannoni* (0.72%) (Table 2) and 31 specimens (3.88%) that could not be identified to the species level (due to partial destruction of the specimens). *Ny. neivai* and *Ev. sallesi-cortelezzii* complex were found in different abundances at each of the sampling sites. Abundance was higher in sampling sites located near wild vegetation (P < 0.05) (Fig. 2). Also, the proportion of gravid females did not differ between sites on the criterion of being located close or far from a natural forest patch (P > 0.05).

Table 1. Descriptions of sand fly collection sites in Hipólito Yrigoyen, Argentina

Site	Characteristics			
1	Tree path on a house front yard next to park like vegetation (camping area) and in close proximity (<50 m) to native rain forest			
2 3	Pedemontane rain forest with typical trees, numerous herbs, lianas, and epiphytic plants			
4	House backyard covered by trees, shrubbery, and herbs			
5	House backyard in urban neighborhood with low plant cover, next to sugar cane plantations			
6	Backyard in informal settlement with scarce plant cover, mainly scattered and small trees, shrubbery, and herbs			
7	Backyard in informal settlement with scarce plant cover, mainly shrubbery and herbs next to sugar cane plantations			
8	Backyard with large plant cover (trees, shrubs, herbs)			
9	Backyard in urban neighborhood with large ornamental or fruit bearing trees			
10	Backyard in urban neighborhood with low or absent plant cover			

Downloaded from https://academic.oup.com/jme/advance-article-abstract/doi/10.1093/jme/tjy129/5064906 by Western Cape University user on 03 August 2018

Site	Ny. Neivai	Mg. migonei	Ev. cortelezzii complex	Pa. shannoni	Not identified	Total
1	124	44	24	5	6	203
2	307	26	43	1	5	382
3	447	30	25	0	13	515
4	12	3	13	0	0	28
5	7	0	4	0	0	11
6	21	8	23	0	0	52
7	7	4	10	0	0	21
8	40	43	31	2	2	118
9	28	3	12	0	5	48
10	7	2	3	2	0	14
Total	1,000	163	188	10	31	1,392

Table 2. Absolute number of sand flies captured per sampling site in Hipólito Yrigoyen, Salta, Argentina

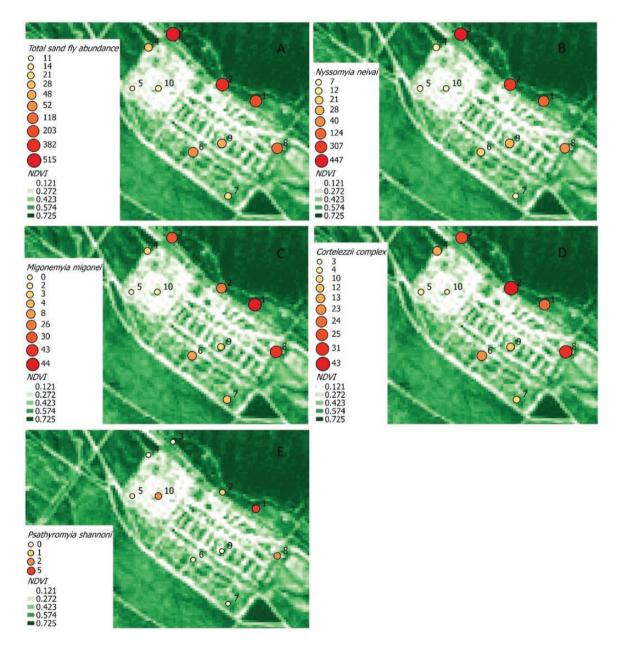


Fig. 2. Sand fly abundance per site: (A) total sand flies, (B) Ny. neivai, (C) Mg. migonei, (D) cortelezzii complex, and (E) Pa. shannoni.

Specimens of *Ny. neivai* were collected at every sampling location. The lower abundance values were recorded in sites 5, 7 (next to sugar cane farming), and site 10 (center neighborhood lacking

vegetation in backyards). The higher abundance was found in the wild vegetation area where sites 1, 2, and 3 were located (Fig. 2B; Tables 1 and 2).

*Migonemya migonei* (Fig. 2C) and the *Ev. cortelezzii* complex (Fig. 2D) showed a similar spatial pattern but with lower abundance variations compared to *Ny. neivai*. The only sites where *Mg. migonei* was not found were sites 5 and 9, where the traps were located in the backyard of houses with little vegetation cover, and site 5 was also next to a sugar cane field (Tables 1 and 2).

Positive significant correlations were found between the NDVI and total sand flies, *Ny. neivai*, and *Mg. migonei* abundances for buffer areas with radii smaller than 150 m (Table 3). As the radii increased, there was lower variation (i.e., smaller min-max range) in NDVI values between sites, which might explain loss of significance.

Based on the distribution of total sand flies abundance according to the NDVI at 100 m buffer area (selected due to higher correlation values), we defined arbitrarily two levels of sand flies presence: low = NDVI  $\leq 0.37$  and high = NDVI > 0.37 (Figs. 3 and 4). The NDVI break value (0.37) was selected due to the relatively sudden change in slope in the graph shown in Fig. 3. The mean sand fly abundance for increased NDVI values was significantly higher than the mean abundance for the lower NDVI values (P = 0.027), using nonparametric test. Higher or lower NDVI cutoff values resulted in two groups of very different sizes in terms of numbers of sites included. A map (Fig. 4) resulting from a reclassification of an NDVI image based on these categories shows that for a large part of the city there is a low chance of coming in contact with sand flies. However, there are patches with high vegetation cover in the south and center of the city that show high levels of NDVI, suggesting a higher probability of contact with a species of sand fly in those areas.

#### Discussion

This work provides the first description of sand fly fauna presence, spatial distribution, and abundance for Hipólito Yrigoyen, a locality of northwest Argentina in the Yungas biome. Similar species composition have been reported for other localities in the province of Salta (Salomón et al. 2001, 2004, 2008; Barroso et al. 2007; Krolewiecki et al. 2013; Quintana et al. 2013).

Although the number of cases of CL in Argentina have increased since 1985 (Salomón et al. 2006, Gil et al. 2010), knowledge of the vector status of suspected sand fly species (as defined in Killick-Kendrick 1990) is still incomplete due to insufficient studies. Laboratory studies on the sand flies incrimination (for the species found in this study) in the transmission of *Leishmania* parasites are scarce (Nieves and Pimenta 2000, Saraiva et al. 2009, Guimarães et al. 2016). Even the CL primary wild reservoirs in northern Argentine are still unknown. All species described in this work show some degree of anthropophily (de Aguiar et al. 1987, Rangel et al. 1990, Hashiguchi et al. 1992, Salomón et al. 1995, Marassá et al. 2013, Paternina et al. 2016) and therefore at least raise the possibility of being vectors. In addition,

the distribution of CL human cases with the presence of these species of sand flies is a basis indicating that at least one of the species is a competent vector in nature (Salomón et al. 2001, 2008; Sosa-Estani et al. 2001; Gil et al. 2010; Krolewiecki et al. 2013; Quintana et al. 2013; Locatelli et al. 2014; Hoyos et al. 2016).

In the province of Chaco, Argentina, the *cortelezzii* complex was proposed as possible vector of CL as it was found naturally infected with *Leishmania (Viannia) braziliensis* (Rosa et al. 2012). In Hipólito Yrigoyen, sand flies from the *cortelezzii* complex were captured in every sampling site.

In this work, *Mg. migonei* was the second most abundant species after *Ny. neivai. Migonemyia migonei* is also suspected of being a vector of CL, considering it was found naturally infected with *L. (V.) braziliensis* in Brazil (de Pita-Pereira et al. 2005). In Puerto Iguazú, Argentina, the species was found for the first time infected by *L. infantum* (Moya et al. 2015), the etiological agent of the visceral leishmaniasis (VL), and in an outbreak of VL in the province of Santiago del Estero in Argentina, the absence of *Lu. longipalpis* led to the proposal of *Mg. migonei* as the putative vector (Salomón et al. 2010). In a similar situation, *Mg. migonei* and *cortelezzii* complex (*Lu. cortelezzii/Lu. sallesi*) were the only species detected together with human and canine infections of *L. infantum* in the department of San Martin, Salta (Barroso et al. 2015).

Nyssomyia neivai was the dominant sand fly species in Hipólito Yrigoyen, amounting to more than 70% of the specimens collected. This species has been found naturally infected with Leishmania spp., L. (V.) braziliensis, and with a consistent spatial distribution with all sites of CL transmission studied in the north of Salta (Salomón et al. 2001, 2004; Córdoba-Lanús et al. 2006; Pita-Pereira et al. 2009). The abundance of Ny. neivai is usually reported as highest in borders of primary vegetation near modified areas (Quintana et al. 2010, Krolewiecki et al. 2013). It has also been found in periurban places and peripheral neighborhoods of urban areas bordering with primary and secondary vegetation (Salomón et al. 2004, Gil et al. 2010). Consistently, in Hipólito Yrigoyen, the abundance of this species was highest in sampling sites close to natural remaining vegetation, as opposed to more central urbanized sites. The fact that Ny. neivai, Mg. migonei, and E. sallesi-cortelezzii complex were found mostly near natural cover remnants, but also at other sites depending on vegetation cover in Hipólito Yrigoyen, highlights a potential for the transmission of leishmaniasis in this locality.

Overall average NDVI values for each sampling site were similar between buffer radii because larger buffer included values from lower buffers. However, variations in NDVI values between sites were more evident at more local scales, because at larger spatial scales the urban landscape may be more homogeneous (Philpott et al. 2014). Consistently, correlation analysis between sand flies abundance and NDVI showed higher correlation coefficients for

Table 3. Correlations between average abundance of sand flies and average NDVI values within four buffer areas around the sampling site

	Buffer area				
Abundances <sup>a</sup>	50 m	100 m	150 m	200 m	
Total sand flies	0.71 (0.03)	0.72 (0.03)	0.61 (0.07)	0.42 (0.21)	
Ny. Neivai	0.64 (0.04)	0.69 (0.02)	0.56 (0.09)	0.4 (0.25)	
Mg. Migonei	0.75 (0.01)	0.71 (0.03)	0.58 (0.08)	0.38 (0.27)	
Ev. cortelezzii-sallesi	0.59 (0.08)	0.56 (0.09)	0.48 (0.15)	0.28 (0.39)	
NDVI mean (range)	0.35 (0.22-0.54)	0.35 (0.23-0.52)	0.36 (0.23-0.50)	0.36 (0.23-0.48)	

Spearman's rank correlation coefficient and *P*-value (in parentheses) are shown. Also shown are mean NDVI and minimum–maximum range per buffer area. *"Psathyromyia shannoni* was excluded from this table since its abundance was too low to attempt meaningful correlations.

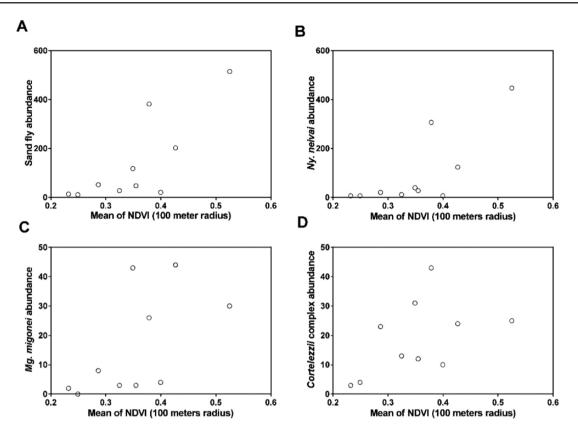


Fig. 3. Sand fly abundance in relation with average NDVI obtained from buffers of 100 m radius around sampling sites, for (A) total sand flies abundance, (B) Ny. neivai, (C) Mg. migonei, and (D) cortelezzii complex.

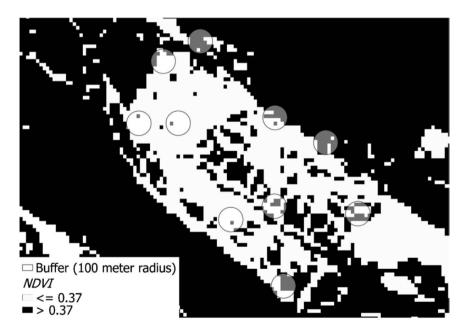


Fig. 4. Map of two levels of NDVI that use categories chosen from Fig. 3 to show levels of low and high probability to find sand flies. The value 0.72 was the maximum NDVI in the chosen Landsat scene.

the 50 or 100 m radius buffer areas compared to wider areas (200 m radius). Yet, sand flies abundances were higher in sites located closer to natural forest patches that could act as source habitat. Site 4, which corresponds to an isolated small patch of wild vegetation (NDVI of 0.4), may have shown a low abundance of sand flies due

to its small size and proximity to the sugar plantations, which receive periodic fumigations (see habitat map in Fig. 1C).

Although we cannot ensure which conditions and resources determined the significant positive correlation between *Ny. neivai*, *Mg. migonei* abundances, and the vegetation cover, we can suggest that the higher humidity, nectar and blood sources, and organic matter (compost) availability on the ground provide refuge and feeding to adults and larval habitat for immature forms of these species. Other work suggests that the abundance of *Ny. neivai* depends to a great extent on the humidity, having found in the autumn high abundance of these sand flies at 22°C and high humidity levels (Fuenzalida et al. 2011). The absence of vegetation cover leads to a high desiccation of the local environment and may provide a lower (or any) biological diversity of feeding sources (Chaves et al. 2008). In addition, a recent study showed that phlebotomine larval habitats are located mainly at the base of the trees (Alencar et al. 2011). Feeding and gravid females seem to prefer areas with trees. We speculate that sites with higher NDVI values (i.e., higher vegetation/tree cover) could function as source population zones and areas with low NDVI may contain sink populations.

The dispersion range for some species of sand flies such as *Ny. neivai* is estimated to be 100–500 m (Galati et al. 2009), with a mean dispersal distance of 55 m (Casanova et al. 2005). In this context, we can consider zones of higher vegetation as islands through which sand flies move from city edges to houses' backyards. Thus, we believe average NDVI from 50 to 100 m radii may be adequate as a proxy variable of the habitat where sand flies adults are more likely to occur.

A study carried out in Teresina, a tropical city in Brazil, indicated that the higher incidence of VL occurred in recently expanded periphery neighborhoods, having the highest vegetation cover (estimated through NDVI) and worst socioeconomic indicators; therefore, the authors suggest vector control intervention in those areas (Cerbino Neto et al. 2009). If urban transmission of CL is conclusively documented in Argentina, the vegetation density of neighborhoods should also be considered when designing control strategies to optimize resources, focusing efforts in periphery areas and those with greater density of vegetation.

Future studies should be conducted in unsampled municipalities and include ground truth validations of estimates used in this study to determine whether the cutoff (0.37 of NDVI), arbitrary selected, is operationally informative. Another consideration in future studies is that the correlation between total sand flies and NDVI, mainly due to the abundances of Ny. neivai and Mg. migonei in this study, is applicable to other species. The NDVI, as well as any other remote sensing derived variables, may be useful to identify or map favorable environmental conditions for a given species, but may not accurately describe the actual species distribution (the realized niche) that may also be influenced, among other factors, by species interactions or other local conditions not detected from the satellite imagery. For example, vector presence or abundance and/or epidemiological risk may be related with reservoir availability (Alexander et al. 2002) and human population density (Ramos et al. 2014). Site 7 had a relatively high average NDVI (=0.40 for the 100 m radius buffer) but a low total sand fly abundance (21 flies), which we speculate may have been a consequence of its proximity to agricultural land (potential exposure to agrochemicals).

The fact that we used only 10 sites and six nights of sampling could generate an underestimation of the degree of correlation between variables. Sand flies that are found in northern Argentina show preferential nocturnal activity and are not observed during the day in open areas with direct sun exposure. Therefore, we believe that an increase in the sample size would not change the positive correlation. However, it cannot be ruled out that other variables exist that were not examined that are at least an important factor for sand flies spatial distribution. On the other hand, a rare species may require a greater capture effort and therefore may simply not have been detected in our study.

We found an association between the average vegetation cover (NDVI) at a local spatial resolution scale and the abundance of these insects. This information may aid in the development of an entomological surveillance system aimed at the efficient design of prevention and control strategies of CL for this locality and other regions with similar conditions.

#### Acknowledgments

We thank Nicanor Sosa and Jorge Gallardo (authorities of the Hipólito Yrigoyen municipality) for their collaboration in the logistics of the field work; Dr. Juan Aparicio for carefully reviewing the manuscript. We thank Gabriela Castillo, Inés Lopez Quiroga, and Fernanda Oreste for their collaboration in the collection and processing of samples. We acknowledge Marcelo Scavuzzo for assistance with the remote sensing products. We thank Diego Marco and Paola Barroso for lending us two CDC traps. This study was supported by the Consejo de Investigación de la Universidad Nacional de Salta (CIUNSa) 2020 and FONCYT (PICT 2013-3213). J.F.G. and R.M.G. are career members of CONICET; M.d.M.C., C.L.H., and G.N.C. hold doctoral scholarships from CONICET, and C.M. is a postdoctorate from the same institution.

#### **References Cited**

- de Aguiar, G. M., M. L. Vilela, and R. B. Lima. 1987. Ecology of the sandflies of Itaguaí, an area of cutaneous leishmaniasis in the State of Rio de Janeiro. Food preferences (Diptera, Psychodidae, Phlebotominae). Mem. Inst. Oswaldo Cruz. 82: 583–584.
- Alencar, R. B., R. G. de Queiroz, and T. V. Barrett. 2011. Breeding sites of phlebotomine sand flies (Diptera: Psychodidae) and efficiency of extraction techniques for immature stages in terra-firme forest in Amazonas State, Brazil. Acta Trop. 118: 204–208.
- Alexander, B., R. L. de Carvalho, H. McCallum, and M. H. Pereira. 2002. Role of the domestic chicken (*Gallus gallus*) in the epidemiology of urban visceral leishmaniasis in Brazil. Emerg. Infect. Dis. 8: 1480–1485.
- Barroso, P. A., J. D. Marco, H. Kato, R. Tarama, P. Rueda, S. P. Cajal, M. A. Basombrío, M. Korenaga, N. J. Taranto, and Y. Hashiguchi. 2007. The identification of sandfly species, from an area of Argentina with endemic leishmaniasis, by the PCR-based analysis of the gene coding for 18S ribosomal RNA. Ann. Trop. Med. Parasitol. 101: 247–253.
- Barroso, P. A., J. D. Marco, F. M. Locatelli, R. M. Cardozo, C. L. Hoyos, M. C. Mora, M. F. García Bustos, I. López-Quiroga, T. Mimori, A. G. Gentile, et al. 2015. Visceral leishmaniasis caused by *Leishmania infantum* in Salta, Argentina: possible reservoirs and vectors. Am. J. Trop. Med. Hyg. 93: 334–339.
- Bravo, A. G., M. G. Quintana, M. Abril, and O. D. Salomón. 2013. The first record of *Lutzomyia longipalpis* in the Argentine northwest. Mem. Inst. Oswaldo Cruz. 108: 1071–1073.
- Brooks, D. 2018. Tropical and subtropical moist broadleaf forests. World Wildlife Fund (WWF), Central South America: Bolivia and Argentina. https://www.worldwildlife.org/ecoregions/nt0165 (accessed 9 March 2018).
- Brown, A. D., S. Pacheco, T. Lomáscolo, and L. Malizia. 2005. Situación Ambiental en los Bosques Andinos Yungueños, pp. 53–61. *In* A. D. Brown, U. Martínez Ortiz, M. Ascerbi, and J. Corcuera (eds.), Situación Ambiental Argentina 2005. Fundación Vida Silvestre, Buenos Aires, Argentina.
- Cabrera, A. L. 1971. Fitogeografía de la república Argentina. Bol. Soc. Argentina Bot. 6: 1-42.
- Campbell-Lendrum, D., J. P. Dujardin, E. Martinez, M. D. Feliciangeli, J. E. Perez, L. N. Silans, and P. Desjeux. 2001. Domestic and peridomestic transmission of American cutaneous leishmaniasis: changing epidemiological patterns present new control opportunities. Mem. Inst. Oswaldo Cruz. 96: 159–162.
- Casanova, C., A. I. Costa, and D. Natal. 2005. Dispersal pattern of the sand fly *Lutzomyia neivai* (Diptera: Psychodidae) in a cutaneous leishmaniasis endemic rural area in Southeastern Brazil. Mem. Inst. Oswaldo Cruz. 100: 719–724.

- Cerbino Neto, J., G. L. Werneck, and C. H. Costa. 2009. Factors associated with the incidence of urban visceral leishmaniasis: an ecological study in Teresina, Piauí State, Brazil. Cad. Saude Publica. 25: 1543–1551.
- Chaves, L. F., J. M. Cohen, M. Pascual, and M. L. Wilson. 2008. Social exclusion modifies climate and deforestation impacts on a vector-borne disease. PLoS Negl. Trop. Dis. 2: e176.
- Córdoba-Lanús, E., M. L. De Grosso, J. E. Piñero, B. Valladares, and O. D. Salomón. 2006. Natural infection of *Lutzomyia neivai* with *Leishmania* spp. in northwestern Argentina. Acta Trop. 98: 1–5.
- Donalisio, M. R., A. T. Peterson, P. L. Costa, F. J. da Silva, H. F. Valença, J. J. Shaw, and S. P. Brandão Filho. 2012. Microspatial distributional patterns of vectors of cutaneous leishmaniasis in Pernambuco, northeastern Brazil. J. Trop. Med. 2012: 642910.

Eastman, J. 2012. IDRISI selva. Clark University, Worcester, MA.

- Fuenzalida, A. D., M. G. Quintana, O. D. Salomón, and M. S. de Grosso. 2011. Hourly activity of *Lutzomyia neivai* in the endemic zone of cutaneous leishmaniasis in Tucumán, Argentina: preliminary results. Mem. Inst. Oswaldo Cruz. 106: 635–638.
- Galati, E. A., M. B. Fonseca, A. M. Marassá, and E. F. Bueno. 2009. Dispersal and survival of *Nyssomyia intermedia* and *Nyssomyia neivai* (Diptera: Psychodidae: Phlebotominae) in a cutaneous leishmaniasis endemic area of the speleological province of the Ribeira Valley, state of São Paulo, Brazil. Mem. Inst. Oswaldo Cruz. 104: 1148–1158.
- Gil, J. F., J. R. Nasser, S. P. Cajal, M. Juarez, N. Acosta, R. O. Cimino, P. Diosque, and A. J. Krolewiecki. 2010. Urban transmission of American cutaneous leishmaniasis in Argentina: spatial analysis study. Am. J. Trop. Med. Hyg. 82: 433–440.
- Gleiser, R. M. 2016. Geoprocessing and expected distribution of diseases (including deforestation, global warming and other changes), pp. 577– 604. *In* C. Brisola Marcondes (ed.), Arthropod borne diseases, vol. 37, 1st ed. Springer, Cham.
- Guimarães, V. C., K. Pruzinova, J. Sadlova, V. Volfova, J. Myskova, S. P. Filho, and P. Volf. 2016. Lutzomyia migonei is a permissive vector competent for Leishmania infantum. Parasit. Vectors. 9: 159.
- Hashiguchi, Y., T. Chiller, A. Inchausti, A. De Arias, M. Kawabata, and J. B. Alexander. 1992. Phlebotomine sandfly species in Paraguay and their infection with *Leishmania*. Ann. Trop. Med. Parasitol. 86: 175–180.
- Hoyos, C. L., S. P. Cajal, M. Juarez, J. D. Marco, A. M. Alberti D'Amato, M. Cayo, I. Torrejón, R. O. Cimino, P. Diosque, A. J. Krolewiecki, et al. 2016. Epidemiology of American tegumentary leishmaniasis and *Trypanosoma cruzi* infection in the Northwestern Argentina. Biomed Res. Int. 2016: 6456031.
- INDEC. 2010. Instituto Nacional de Estadística y Censos de la República Argentina. Censo Nacional de Población, Hogares y Viviendas 2010. https://www.indec.gob.ar (accessed 9 March 2018).
- Killick-Kendrick, R. 1990. Phlebotomine vectors of the leishmaniases: a review. Med. Vet. Entomol. 4: 1–24.
- Kravchenko, V., G. Wasserberg, and A. Warburg. 2004. Bionomics of phlebotomine sandflies in the Galilee focus of cutaneous leishmaniasis in northern Israel. Med. Vet. Entomol. 18: 418–428.
- Krolewiecki, A. J., J. F. Gil, M. Quipildor, S. P. Cajal, C. Pravia, M. Juarez, C. Villalpando, F. M. Locatelli, M. Chanampa, G. Castillo, et al. 2013. Restricted outbreak of American tegumentary leishmaniasis with high microfocal transmission. Am. J. Trop. Med. Hyg. 88: 578–582.
- Locatelli, F. M., S. P. Cajal, P. A. Barroso, J. J. Lauthier, M. C. Mora, M. Juarez, H. Kato, J. R. Nasser, Y. Hashiguchi, M. Korenaga, et al. 2014. The isolation and molecular characterization of *Leishmania* spp. from patients with American tegumentary leishmaniasis in northwest Argentina. Acta Trop. 131: 16–21.
- Lomáscolo, T., A. D. Brown, and L. R. Malizia. 2010. Reserva de Biosfera de las Yungas. Ediciones del Subtrópico, Tucumán.
- Marassá, A. M., E. A. Galati, D. P. Bergamaschi, and C. A. Consales. 2013. Blood feeding patterns of *Nyssomyia intermedia* and *Nyssomyia neivai* (Diptera, Psychodidae) in a cutaneous leishmaniasis endemic area of the Ribeira Valley, State of São Paulo, Brazil. Rev. Soc. Bras. Med. Trop. 46: 547–554.
- Mikery Pacheco, O. F. M., J. C. Rojas León, E. A. Rebollar-Téllez, and A. Castillo Vera. 2015. Sandfly (Diptera: Psychodidae: Phlebotominae)

species diversity in an urban area of the municipality of Tapachula, Chiapas, Mexico. Mem. Inst. Oswaldo Cruz. 110: 142–144.

- Moya, S. L., M. G. Giuliani, M. Manteca Acosta, O. D. Salomón, and D. J. Liotta. 2015. First description of *Migonemyia migonei* (França) and *Nyssomyia whitmani* (Antunes & Coutinho) (Psychodidae: Phlebotominae) natural infected by *Leishmania infantum* in Argentina. Acta Trop. 152: 181–184.
- Nieves, E., and P. F. Pimenta. 2000. Development of Leishmania (Viannia) braziliensis and Leishmania (Leishmania) amazonensis in the sand fly Lutzomyia migonei (Diptera: Psychodidae). J. Med. Entomol. 37: 134–140.
- Ontivero, I. M., M. D. Beranek, J. R. Rosa, F. F. Ludueña-Almeida, and W. R. Almirón. 2018. Seasonal distribution of phlebotomine sandfly in a vulnerable area for tegumentary leishmaniasis transmission in Córdoba, Argentina. Acta Trop. 178: 81–85.
- Paternina, L. E., D. Verbel-Vergara, L. Romero-Ricardo, A. Pérez-Doria, M. Paternina-Gómez, L. Martínez, and E. E. Bejarano. 2016. Evidence for anthropophily in five species of phlebotomine sand flies (Diptera: Psychodidae) from northern Colombia, revealed by molecular identification of bloodmeals. Acta Trop. 153: 86–92.
- Philpott, S. M., J. Cotton, P. Bichier, R. L. Friedrich, L. C. Moorhead, S. Uno, and M. Valdez. 2014. Local and landscape drivers of arthropod abundance, richness, and trophic composition in urban habitats. Urban Ecosyst. 17: 513–532.
- de Pita-Pereira, D., C. R. Alves, M. B. Souza, R. P. Brazil, A. L. Bertho, A. de Figueiredo Barbosa, and C. C. Britto. 2005. Identification of naturally infected *Lutzomyia intermedia* and *Lutzomyia migonei* with *Leishmania (Viannia) braziliensis* in Rio de Janeiro (Brazil) revealed by a PCR multiplex non-isotopic hybridisation assay. Trans. R. Soc. Trop. Med. Hyg. 99: 905–913.
- Pita-Pereira, D., G. D. Souza, A. Zwetsch, C. R. Alves, C. Britto, and E. F. Rangel. 2009. First report of *Lutzomyia (Nyssomyia) neivai* (Diptera: Psychodidae: Phlebotominae) naturally infected by *Leishmania (Viannia) braziliensis* in a periurban area of south Brazil using a multiplex polymerase chain reaction assay. Am. J. Trop. Med. Hyg. 80: 593–595.
- Queiroz, M. F., J. R. Varjão, S. C. Moraes, and G. E. Salcedo. 2012. Analysis of sandflies (Diptera: Psychodidae) in Barra do Garças, State of Mato Grosso, Brazil, and the influence of environmental variables on the vector density of *Lutzomyia longipalpis* (Lutz & Neiva, 1912). Rev. Soc. Bras. Med. Trop. 45: 313–317.
- Quintana, M. G., O. D. Salomón, and M. S. De Grosso. 2010. Distribution of phlebotomine sand flies (Diptera: Psychodidae) in a primary forest-crop interface, Salta, Argentina. J. Med. Entomol. 47: 1003–1010.
- Quintana, M., O. Salomón, R. Guerra, M. L. De Grosso, and A. Fuenzalida. 2013. Phlebotominae of epidemiological importance in cutaneous leishmaniasis in northwestern Argentina: risk maps and ecological niche models. Med. Vet. Entomol. 27: 39–48.
- Ramos, W. R., J. F. Medeiros, G. R. Julião, C. M. Ríos-Velásquez, E. F. Marialva, S. J. Desmouliére, S. L. Luz, and F. A. Pessoa. 2014. Anthropic effects on sand fly (Diptera: Psychodidae) abundance and diversity in an Amazonian rural settlement, Brazil. Acta Trop. 139: 44–52.
- Rangel, E. F., A. C. Azevedo, C. A. Andrade, N. A. Souza, and E. D. Wermelinger. 1990. Studies on sandfly fauna (Diptera: Psychodidae) in a foci of cutaneous leishmaniasis in mesquita, Rio de Janeiro State, Brazil. Mem. Inst. Oswaldo Cruz. 85: 39–45.
- R Development Core Team. 2008. R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. http://www.R-project.org.
- Rosa, J., D. P. Pereira, R. P. Brazil, J. D. Filho, O. Salomón, and E. Szelag. 2012. Natural infection of *cortelezzii* complex (Diptera: Psychodidae: Phlebotominae) with *Leishmania braziliensis* in Chaco, Argentina. Acta Trop. 123: 128–131.
- Salomón, O. D. 2009. [Leishmaniasis vectors in the Americas]. Gaz. Médica da Bahia. 79: 3–15.
- Salomón, O. D., B. L. Travi, and E. L. Segura. 1995. Note on sandflies associated with a tegumentary leishmaniasis focus in Salta, Argentina, 1988. Rev. Inst. Med. Trop. Sao Paulo. 37: 91–92.
- Salomón, O. D., S. Sosa Estani, L. Canini, and E. Cordoba Lanus. 2001. [Tegumentary leishmaniasis in area with epidemic levels of transmission, Salta, Argentina, 1998]. Medicina (B. Aires). 61: 284–290.

- Salomón, O. D., M. L. Wilson, L. E. Munstermann, and B. L. Travi. 2004. Spatial and temporal patterns of phlebotomine sand flies (Diptera: Psychodidae) in a cutaneous leishmaniasis focus in northern Argentina. J. Med. Entomol. 41: 33–39.
- Salomón, O. D., P. W. Orellano, M. G. Quintana, S. Perez, S. Sosa Estani, S. Acardi, and M. Lamfri. 2006. [Transmission of tegumentary leishmaniasis in Argentina]. Medicina (B. Aires). 66: 211–219.
- Salomón, O. D., M. G. Quintana, and M. Zaidenberg. 2008. Urban distribution of Phlebotominae in a cutaneous leishmaniasis focus, Argentina. Mem. Inst. Oswaldo Cruz. 103: 282–287.
- Salomón, O. D., M. G. Quintana, G. Bezzi, M. L. Morán, E. Betbeder, and D. V. Valdéz. 2010. *Lutzomyia migonei* as putative vector of visceral leishmaniasis in La Banda, Argentina. Acta Trop. 113: 84–87.
- Saraiva, L., G. M. Carvalho, C. M. Gontijo, P. F. Quaresma, A. C. Lima, A. L. Falcão, and J. D. Andrade Filho. 2009. Natural infection of *Lutzomyia* neivai and *Lutzomyia sallesi* (Diptera: Psychodidae) by *Leishmania* infantum chagasi in Brazil. J. Med. Entomol. 46: 1159–1163.

- Sosa-Estani, S., E. L. Segura, A. Gomez, O. D. Salomón, M. Peralta, V. Coutada, and L. M. Ruiz. 2001. [Cutaneous leishmaniasis in Northern Argentina: identification of risk factors in a case-cohort study of three municipalities in Salta]. Rev. Soc. Bras. Med. Trop. 34: 511–517.
- Tourre, Y. M., L. Jarlan, J. P. Lacaux, C. H. Rotela, and M. Lafaye. 2008. Spatio-temporal variability of NDVI-precipitation over southernmost South America: possible linkages between climate signals and epidemics. Environ. Res. Lett. 3. 1–9.
- Walsh, J. F., D. H. Molyneux, and M. H. Birley. 1993. Deforestation: effects on vector-borne disease. Parasitology. 106(Suppl): S55–S75.
- Yadon, Z. E., L. C. Rodrigues, C. R. Davies, and M. A. Quigley. 2003. Indoor and peridomestic transmission of American cutaneous leishmaniasis in northwestern Argentina: a retrospective case-control study. Am. J. Trop. Med. Hyg. 68: 519–526.
- Young, D. G., M. A. Duncan. 1994. Guide to the identification and geographic distribution of *Lutzomyia* sand flies in Mexico, the West Indies, Central and South America (Diptera: Psychodidae). Mem. Am. Entomol. Inst. 54: 54–81.