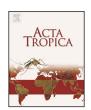
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Intradomiciliary and peridomiciliary captures of sand flies (Diptera: Psychodidae) in the leishmaniasis endemic area of Chapare province, tropic of Cochabamba, Bolivia



C. Ballart^{a,*,1}, G. Vidal^{b,1}, A. Picado^a, M.R. Cortez^b, F. Torrico^{b,c}, M.C. Torrico^{b,c}, R.E. Godoy^d, D. Lozano^{b,c,1}, M. Gállego^{a,e,1}

- ^a Instituto de Salud Global de Barcelona (ISGlobal), Hospital Clínic Universitat de Barcelona, Barcelona, Spain
- ^b CEADES Salud y Medio Ambiente, Cochabamba, Bolivia
- ^c Facultad de Medicina, Universidad Mayor de San Simón (U.M.S.S.), Cochabamba, Bolivia
- d Laboratório de Transmissores de Leishmanioses, Laboratório de Referência em Vigilância Entomológica: Taxonomia e Ecologia de Vetores das Leishmanioses, Instituto Oswaldo Cruz, Fundação Oswaldo Cruz, Rio de Janeiro, Brazil
- ^e Laboratori de Parasitologia, Facultat de Farmàcia, Universitat de Barcelona, Barcelona, Spain

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ABSTRACT

In South America, cutaneous leishmaniasis is the most frequent clinical form of leishmaniasis. Bolivia is one of the countries with higher incidence, with 33 cases per 100,000 individuals, and the disease is endemic in 70% of the territory. In the last decade, the number of cases has increased, the age range has expanded, affecting children under 5 years old, and a similar frequency between men and women is found. An entomological study with CDC light traps was conducted in three localities (Chipiriri, Santa Elena and Pedro Domingo Murillo) of the municipality of Villa Tunari, one of the main towns in the Chapare province (Department of Cochabamba, Bolivia). A total of 16 specimens belonging to 6 species of the genus *Lutzomyia* were captured: *Lu. aragaoi, Lu. andersoni, Lu. antunesi, Lu. shawi, Lu. yuilli yuilli* and *Lu. auraensis*. Our results showed the presence of two incriminated vectors of leishmaniasis in an urbanized area and in the intradomicile. More entomological studies are required in the Chapare province to confirm the role of vector sand flies, the intradomiciliary transmission of the disease and the presence of autochthonous cases of cutaneous leishmaniasis.

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1. Introduction

Leishmaniasis is a parasitic disease affecting man and other mammals. After malaria, leishmaniasis is the most important vector-borne disease in terms of the number of people affected. Globally, 350 million people are at risk of infection in 98 countries around the world (WHO, 2010). In South America, cutaneous leishmaniasis (CL) is the most frequent clinical form, with less presence of cases of mucosal leishmaniasis (ML) and in a minor extent of visceral leishmaniasis (VL) (Rojas et al., 2009). Bolivia is one of the countries with higher incidence of CL in the area, with 33 cases per 100,000 individuals (García et al., 2009). In the last decade, the number of cases has increased up to 2000 new cases per year in the

country (Alvar et al., 2012), where the disease is endemic in 70% of the territory, mainly in the provinces of La Paz, Beni and Pando (Herrera et al., 2013). Leishmania (Viannia) braziliensis is the most prevalent species causing CL in Bolivia (85% of the cases). The other species involved are L. (Leishmania) amazonensis, L. (V.) lainsoni and L. (V.) guyanensis (García et al., 2009).

Transmission to humans and other mammalian species is through the bite of phlebotomine sand flies (Diptera: Psychodidae) (Lainson, 1988). The distribution of leishmaniasis is closely related to the distribution of vector species. From 86 sand fly species present in Bolivia, only few are incriminated as vectors of leishmaniasis: Lutzomyia carrerai, Lu. llanosmartinsi, Lu. yucumensis, Lu. nuneztovari, Lu. shawi and Lu. longipalpis (Bustamante et al., 2012; García et al., 2009).

The epidemiology and clinical features of the disease are highly variable due to the interaction of many factors dependent on the parasite, vector, vertebrate host and the environment (Bailey and Lockwood., 2007). In Cochabamba Department of Bolivia, leishmaniasis was a typically sylvatic disease affecting rural communities,

^{*} Corresponding author at: Fundación Instituto de Salut Global (ISGlobal), Calle Rosselló 132, 4° , 08036, Barcelona, Spain.

E-mail address: cristina.ballart@isglobal.org (C. Ballart).

¹ Authors contributed equally.

basically males in working age (21-30 years old) that enter to the forest to work (traditional pattern) (García et al., 2011). Changes on transmission pattern were noticed in the Chapare province of Bolivia (García et al., 2007), where 65.3% of the 1400 cases of leishmaniasis detected between 2002 and 2010 in the Department of Cochabamba were reported (García et al., 2011). In the last decade, a new and uncommon event occurred in the area: the age range has expanded affecting children and a similar frequency between men and women is found (García et al., 2007). Current data available in the municipality of Villa Tunari, in the period 2009–2013, are: 330 cases in men and 162 cases in women, from these 21 cases in children <5 years (13 women, 8 men) (Vidal, Lozano and Torrico, data not published). In the area, entomological studies were conducted in the Isiboro-Secure national park of Villa Tunari municipality (Bustamante et al., 2012; García et al., 2007), where the risk of transmission is the highest of the Cochabamba Department tropical region. The objective of the present work was to obtain data on the sand flies present outside the national park and in the surroundings of more urbanized areas of this region.

2. Materials and methods

2.1. Area of study and data collection

The study was conducted in the municipality of Villa Tunari, one of the main towns in the Chapare province (Department of Cochabamba, Bolivia). The population of Villa Tunari municipality is about of 71,000 inhabitants (88% from rural areas) (Instituto Nacional de Estadística (INE, 2014, data of 2012; http:// censosbolivia.ine.gob.bo/). The climate is tropical humid with an average annual temperature of 24°C and an average annual relative humidity of 81%. The town of Villa Tunari and its environs consist of valley rain forests between 200 and 400 m above the sea level (a.s.l.) that surrounds the area's main waterway, the Chapare River. Specifically, the study was performed in three localities of an area with a high incidence outside the Isiboro-Secure national park (730 cases/100.000 inhabitants) (Vidal, Lozano and Torrico, data not published): Chipiriri, Santa Elena (Chipiriri district) and Pedro Domingo Murillo (Villa 14 de Septiembre district), near to Villa Tunari town, and with similar environmental conditions (Fig. 1).

Householders of every house were informed in advance and received and signed an informed consent to participate in the study. It also included consent for making photographs of sampling sites and surrounding areas.

The GPS Test mobile application was used to record the geographical coordinates. Data were entered into an Access database (Microsoft).

2.2. Study of sand flies

The entomological survey was carried out in urbanized areas (villages or hamlets with a higher concentration of housing in sylvatic or forested regions). CDC light traps were placed in 36 inhabited houses from September to December 2014 at altitudes ranging from 196 to 309 m a.s.l. The houses were selected following the next criteria: (i) A case of human leishmaniasis occurred in the past in the house or in the neighbouring houses and (ii) The inhabitants slept in the house regularly. Two CDC light traps were placed once at each house: one in peridomiciliary and another in intradomiciliary environments. Peridomicile: outdoors, less than 10m away from the house, where people often perform their activities. Intradomicile: indoors, preferably in the bedroom. The traps were set at sunset and left in operation all night (from 6 p.m. to 6 a.m.).

The bags containing the sand flies were placed in a freezer for 10 min minimum in order to reduce their activity. The sand flies were recovered, collected in vials correctly labelled containing 70% alcohol and stored until their morphological identification. Specimens were mounted on Hoyer's medium and identified following the keys of Young and Duncan (1994).

3. Results

Sand flies were captured in 9 houses (Table 1). A total of 16 specimens belonging to 6 species of the genus *Lutzomyia* were captured (11 females and 5 males): *Lu. aragaoi, Lu. andersoni, Lu. antunesi, Lu. shawi, Lu. yuilli yuilli* and *Lu. auraensis*. All species were captured isolated with the exception of *Lu. andersoni* and *Lu. yuilli yuilli*. *Lu. andersoni, Lu. aragaoi, Lu. antunesi* and *Lu. auraensis* were captured in only one house, whilst *Lu. shawi* and *Lu. yuilli yuilli* in 2 and 5 houses, respectively. In one of the houses two traps were placed at different periods (resulting in the capture of one different species at each time, *Lu. yuilli yuilli* and *Lu. auraensis*. A total of eight specimens were captured in the intradomicile, with the same number in the peridomicile. At least one specimen was found in the intradomicile for each species, with the exception of *Lu. aragaoi* and *Lu. auraensis*.

4. Discussion

The period of capture corresponds with the one with highest captures recorded in previous studies in the area (Bustamante et al., 2012; García et al., 2007), but the present study includes more urbanized areas instead of forested ones. Three localities were chosen taking into account the presence of the disease and the acceptance and access to the communities: Chipiriri, Santa Elena and Pedro Domingo Murillo. Chipiriri was one of the locations with a higher incidence of leishmaniasis in the area between 2009 and 2013 (Vidal, Lozano and Torrico, data not published). All the households selected for the study had presented one case of leishmaniasis in the past, either in the house itself or in the neighbourhood, and shared very similar ecological characteristics but differences in the collected specimens are observed.

All the six species captured were previously found in Bolivia among the 86 recorded in the country (Young and Duncan, 1994), although *Lu. andersoni* and *Lu. auraensis* are new findings in the Chapare region as they were not previously captured in the Isiboro-Secure national park despite the highest number of sand flies captured (4463 and 945 specimens in 2000 and 2007, respectively) (Bustamante et al., 2012; García et al., 2007). In the case of *Lu. auraensis*, this species was previously captured in the nearest province of Carrasco (Bermúdez et al., 1993).

In the present study, the number of captured sand flies is low (16 specimens), as it was in an aforementioned study carried out also with CDC traps in the Isiboro-Secure park of the Chapare province (Bustamante et al., 2012). CDC traps were used in this preliminary study, as they are more useful for capturing inside houses, but additional studies, including a greater number of sites of capture, trapping in different periods of the year as well as using Shannon traps (De Souza et al., 2004) are required to define the characteristics of the phlebotomine fauna in urbanized and sylvatic areas of the region.

None of the females presented blood in their abdomen neither eggs. Even if people did activities in the intra- and peridomicile during sunlight, when sand flies are expected to be more active, this result is not surprising because the few specimens caught as well as the traps were placed only for one night in every house. Three out of the four species captured inside houses are considered antropophilic (*Lu. yuilli yuilli, Lu. shawi* and *Lu. antunesi*) (Le Pont et al., 1990), as it is suspected for one of the species found only

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