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Phlebotomine sand flies (Diptera: Psychodidae) associated with a cutaneous leishmaniasis case in a Crohn's disease patient in a residential area of the Levante region (eastern Spain)

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ABSTRACT

Introduction: Leishmaniasis represents a major public health concern in the Mediterranean basin, where both cutaneous and visceral forms are endemic. Although canine leishmaniasis is well recognized, its impact extends far beyond veterinary health, affecting human populations.

Objective: To characterize the sand fly population in a residential area of Valencia province (eastern Spain) associated with a case of cutaneous leishmaniasis diagnosed in early 2025 in an immunosuppressed teenager with Crohn's disease.

Method: An entomological survey was conducted from July to September 2024 in the garden of a residence in the municipality of L'Eliana, several months before the leishmaniasis case was diagnosed. One BG-Sentinel 2 trap baited with BG-Lure was deployed once per week for 24 hours to monitor *Aedes albopictus* activity. As other hematophagous Diptera had been preserved, sand flies were later analyzed.

Results: The presence of *Phlebotomus perniciosus*, a major vector of *Leishmania infantum* in southern Europe, was confirmed, although in low numbers.

Conclusions: Considering the parasite incubation period, and the absence of recent travel history, the infection was most likely acquired within the residential environment. While leishmaniasis has traditionally been regarded as a rural disease, the present case report sheds light on the importance of peri-urban and residential settings in the transmission of this parasitic disease.

KEYWORDS: *Phlebotomus perniciosus* · *Sergentomyia minuta* · *Leishmania infantum* · Valencia · Spain

INTRODUCTION

Leishmaniasis caused by *Leishmania infantum* (syn. *L. chagasi*) remains a significant neglected and reemerging disease in southern Europe¹. In Spain, both cutaneous and visceral leishmaniasis (CL and VL, respectively) have been reported. While CL typically

curses as self-limiting single lesions, VL can be life-threatening, particularly in infants and immunocompromised individuals². In the Old World, *L. infantum* is transmitted by sand flies of the genus *Phlebotomus*, with *Phlebotomus perniciosus* recognized as the primary vector in Spain, Portugal, and Italy, while *Phlebotomus ariasi* is generally regarded as a secondary vector, although it may replace *P. perniciosus* as the main vector in certain epidemiological contexts, such as in areas of southern France³. Here, we report the sand fly fauna and epidemiological context associated with a case of CL in an immunocompromised patient residing in a residential area of the Levante region (eastern Spain).

METHOD

Within the framework of a study on the domestic bioecology of *Aedes albopictus* (Diptera: Culicidae) (data not shown), entomological surveys were conducted from July to September 2024 in the garden of a residence in L'Elia, Valencia province. For this purpose, BG-Sentinel 2 traps baited with BG-Lure Mosquito Attractant were operated once per week for 24 hours. The entomological collections incidentally included other hematophagous Diptera which were also preserved.

In January 2025, a 15-year-old female adolescent with Crohn's disease treated with immunosuppressants, residing in the neighboring house where the study was conducted, presented four cutaneous ulcers. The lesions appeared as well-demarcated ulcers with raised indurated borders and central necrotic areas, each measuring approximately 5 mm in diameter. They were non-tender, non-exudative, and located on exposed areas ranging from the gluteal region to the ankle (Fig. 1). The clinical presentation was consistent with localized CL, which was subsequently confirmed as being caused by *L. infantum* at the patient's public health centre. According to the family of the patient, no travel history was reported during 2024. The adolescent completed a course of intravenous amphotericin B and achieved complete recovery, with no clinical manifestations other than the documented skin ulcers and no relapse having been reported up to October 2025.

The residence of the patient was situated in a peri-urban residential area composed of detached single-family houses with private gardens. Pet dogs (*Canis lupus familiaris*) and cats (*Felis catus*) were common in neighboring households. Public peri-domestic areas included vacant plots with shrubs and low vegetation, such as carob trees (*Ceratonia siliqua*), pines (*Pinus halepensis*), olive trees (*Olea europaea*), and mastic shrubs (*Pistacia lentiscus*), along with other less frequent Mediterranean species. A feral cat colony was established in the vicinity of the household, and leporids were abundant in the vacant plots. Synanthropic rodents, mainly black rat (*Rattus rattus*) and Norway rat (*R. norvegicus*), were also present within the residential environment of the study area (Fig. 1).

Following the diagnosis of CL, the preserved hematophagous specimens were examined for sand flies, whose identification was carried out using the taxonomic key of Gállego-Berenguer et al.⁴



Figure 1. Evaluation of the epidemiological context of a CL case of a minor suffering from Crohn's disease. **A.** Study area. Location of L'Eliana municipality near the urban area of Valencia. **B.** BG-Sentinel 2 trapping site in the residence adjacent to the residence. **C.** General overview of the epidemiological context of CL transmission in residential areas. Figure prepared in BioRender software. **D.** Characterization of domestic environment of the patient. **E.** Characterization of peridomestic environment of the patient. **F.** Skin lesions (CL) caused by *L. infantum* in a Crohn's disease adolescent patient.

RESULTS

The presence of *P. perniciosus*, the primary vector of leishmaniosis in the Mediterranean basin, was confirmed, albeit at very low density, with only four males and one female having been caught between August and September 2024. *Sergentomyia minuta* was also

detected, occurring at slightly higher densities (three males and ten females) throughout the sampling period (Fig. 2).

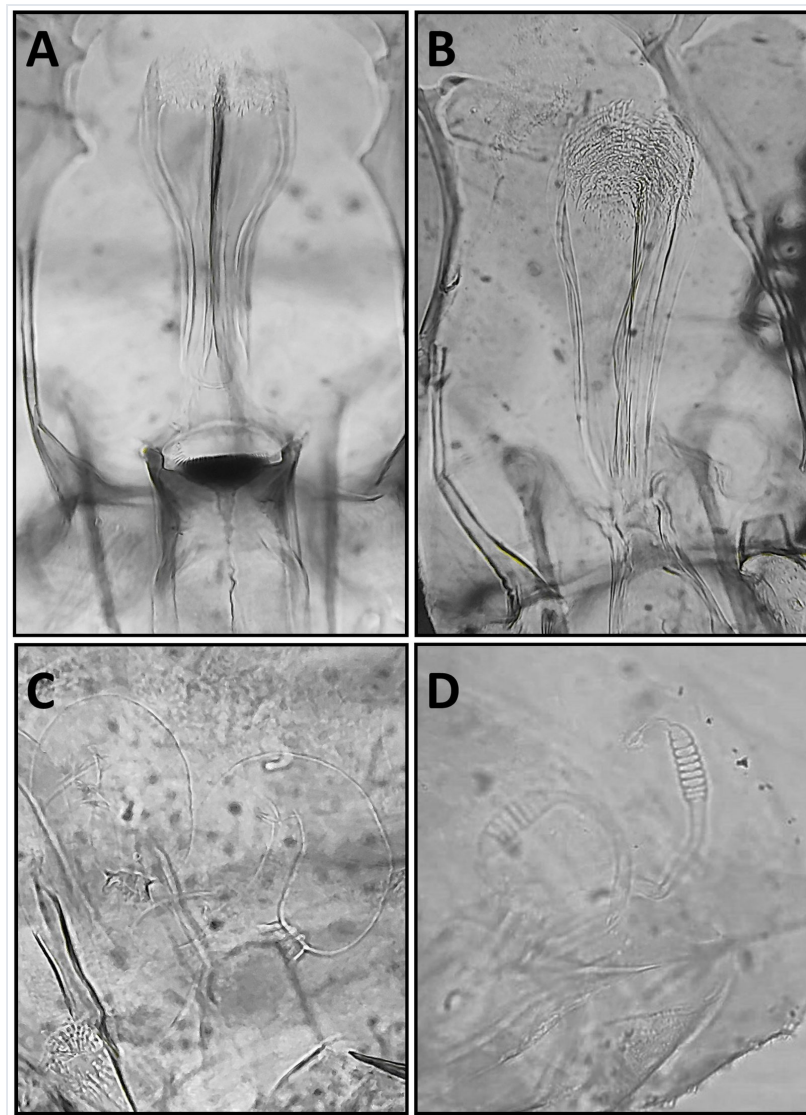


Figure 2. Identified phlebotomine sand flies. **A–C**, *Sergentomyia minuta* pharyngeal armor and spermatheca. **B–D**, *Phlebotomus perniciosus* pharyngeal armor and spermatheca. Images taken on an Olympus CO11 microscope at $\times 40$ magnification.

DISCUSSION

Based on the latest available data, 291 human leishmaniosis cases were reported in the country in 2022, of which 35% occurred in the Valencian Community⁵, making it the region with the highest infection rate in mainland Spain. Despite this epidemiological relevance, information on sand fly populations in this region remains scarce. Furthermore, limited information is available regarding phlebotomine sand flies in urban and peri-urban areas of Spain, even though they represent one of the most common sites of potential contact between humans and zoonotic parasites such as *L. infantum*. Dogs have traditionally been seen as the main reservoir of *L. infantum*⁶, although other sylvatic reservoirs may play major roles in certain leishmaniosis outbreaks in southern Europe⁷. In this sense, during the largest human leishmaniosis outbreak recorded in Europe, which occurred in an urban area of Madrid in 2009 (central Spain), lagomorphs

such as the Iberian hare (*Lepus granatensis*) and the European rabbit (*Oryctolagus cuniculus*), were seen as the main reservoir of *L. infantum*⁸, while dogs played a negligible role⁹.

The residence of the patient was located in an urbanized residential area, characterized by the presence of both urban reservoirs (dogs and cats) and sylvatic reservoirs (leporids). Additionally, urban rodents, whose role as potential reservoirs of *L. infantum* has been suggested¹⁰, were present in the study area. Considering a median time of 3 (1–17) months from exposure to leishmaniosis symptoms of patients returning to non-endemic countries from Spain¹¹, the presence of both potential reservoirs and *P. perniciosus*, and the absence of reported travel history of the patient during 2024, it could be argued that the infection was acquired within the residential area of the adolescent.

The present research was conducted targeting *A. albopictus* mosquitoes, another major vector present in residential areas of Valencia¹². As the captures were incidental and obtained using non-specific mosquito traps, the results cannot be directly compared with previous studies employing specific sampling methods. In this context, implementing targeted sampling methods such as light or sticky traps could yield a more comprehensive and accurate representation of the sand fly fauna^{13,14}. Nevertheless, these findings confirm the presence of *P. perniciosus* in a peri-urban residential area of Valencia.

Previous cases of human leishmaniosis in Crohn's disease patients with immunosuppressive treatments in Spain have been reported, being CL the most common initial manifestation, which may then lead to VL¹⁵. As VL can be a severe and life-threatening condition in immunocompromised individuals, physicians managing patients with inflammatory bowel disorders, such as Crohn's disease, living in leishmaniosis-endemic areas should be aware of CL and specially VL. Accordingly, proper epidemiological surveys, and the characterization of potential risk factors for the presence of sand flies in residential areas, are critical.

While the province of Valencia has reported the highest number of human leishmaniosis cases in the most recent national data⁵ — while 481 reported cases were recorded between 2019 and 2021 (41.04% of the total reported national cases)² —, the study of sand flies in this region is scarce to none. At a national level, available phenological and ecological studies of phlebotomine sand flies have mostly been conducted in central and southeastern mainland Spain¹⁶. Nevertheless, the obtained data may not be representative of sand fly communities in the Levante region, which is characterized by a distinct thermo-Mediterranean bioclimate with general low precipitations and marked torrential storms, resulting in a significant water deficit, especially during the summer season. Furthermore, ongoing global change is expected to alter the phenology and spatial distribution of *L. infantum* vectors in the region, as has recently been documented for numerous other arthropod vectors.

CONCLUSIONS

Future efforts for the characterization of the ecology and phenology of phlebotomine sand fly communities of Valencia and other sites of the Levante region in eastern Spain are necessary for understanding the epidemiological context of VL and CL. The present research highlights the importance of residential areas as potential key sites of *L. infantum*-human infections. Physicians with immunocompromised patients living in risk areas of *L. infantum* transmission should be aware of VL and CL.

AUTHOR CONTRIBUTIONS

P.M.A.E.: Conceptualization, Methodology, Investigation, Resources, Writing – Original Draft, Writing – Review and Editing, Supervision, Project Administration; E.P.M.: Investigation, Writing – Original Draft, Writing – Review and Editing, Visualization; M.L.F.E.: Investigation, Writing – Original Draft, Writing – Review and Editing, Visualization; M.G.T.: Writing – Review and Editing, Supervision; A.E.A.B.: Writing – Review and Editing; S.A.F.: Writing – Review and Editing; M.A.R.S.: Methodology, Investigation, Resources, Writing – Original Draft, Writing – Review and Editing. All authors read and approved the final version of the manuscript.

CONFLICT OF INTEREST

None declared.

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