Open Peer Review on Qeios

Sandflies Phlebotominae (Insecta: Diptera Psychodidae).

Carlos Henrique Marchiori[1]

1 Instituto Federal Goiano

Potential competing interests: No potential competing interests to declare.

Co-authors: Marco Vinícios de Oliveira Santana[2] and Klebert de Paula Malheiros[3].

[2-3]Instituto Marco Santana, Goiânia, Goiás, Brazil.

Sandflies are small insects whose lengths vary between 1 and 3 mm and rarely reach 5 mm. They have long legs and a densely hairy body and are generally brown, hence the nickname “straw mosquito”. In the winged phase, sandflies show sexual dimorphism expressed not only in differences in body shape but also in feeding behavior, which is expressed in the exclusive hematophagy of females. In males, the proboscis's mouthparts are shorter and atrophied, unlike females, which have a longer proboscis, adapted to piercing the skin of vertebrates and sucking blood. [1-3].

Locomotion is characterized by hopping flight and, unlike many. Diptera keep their wings erect when at rest. Sandflies have a pantropical distribution, with some species found in the regions of the Americas, they are distributed from the extreme south of Canada to the north of Argentina. Some species have a restricted, regional, or local distribution, while others have a broad continental distribution, resulting in wide overlapping ranges. Knowledge of the sand fly fauna proved to be of great importance due to the ability of these insects to transmit pathogens. In the New World, the genus Lutzomyia França, 1924, is the most important, with some species involved in the transmission of the causative agents leishmaniasis, bartonellosis, and arbovirus (Figure 1) [3-5].

Figure 1. *Phlebotomus papatasi* (Scopoli, 1786). Source: https://pt.wikipedia.org/wiki/Phlebotominae.

The eggs have an ellipsoid shape and are deposited in terrestrial microhabitats, rich in organic content. and soon after laying they appear whitish or yellowish, turning dark brown within a few hours. The larval forms have a vermiform appearance, whose body is divided into a head, three thoracic segments, and nine abdominal segments, with the last segments having pseudopods for locomotion on the substrate. Soon after hatching, they feed on eggshells, the bodies of dead sandflies, and other organic matter available in the breeding area. The larval phase goes through four larvae stages 1, 2, 3, and 4, and can last from 15 to 70 days. The pupa has a cylindrical body divided into a cephalothorax, head thorax, and abdomen, the latter with nine segments. Pupae typically remain attached to a hardened substrate until hatching (Figure 2) [5-8].

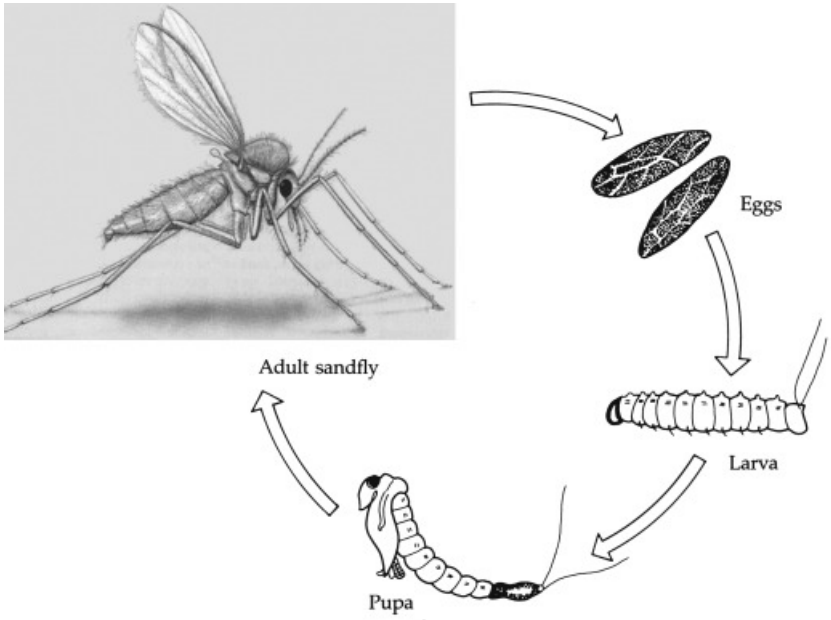

Figure 2. Stages in the life cycle of the sandfly*Phlebotomus* spp.
Source: tps://www.sciencedirect.com/topics/biochemistry-genetics-and-molecular-biology/phlebotominae.

The life cycle goes through four phases: egg, larva, pupa, and adult with wings. The duration of the evolutionary cycle can vary, on average, between 30 and 45 days, depending on the species and the influence of temperature, humidity, and food availability conditions. Females can lay 40 to 100 eggs, varying depending on the species. In general, they carry out only one oviposition throughout their lives: Studies indicate that, under experimental conditions, adult insects can live between 20 and 30 days. Both sexes need carbohydrates as a source of energy, feeding on nectar from flowers, fruits, plant sap, and substances excreted by aphids. Adult females feed mainly during the twilight and nocturnal periods and are attracted to houses by positive phototropism. Some species have a high degree of anthropophilia and endophilia, while others are eclectic and exophilic [8-11].

Regarding behavior, adults are concentrated in places close to breeding sites and peridomestic outbuildings, mainly in shelters for domestic animals, such as dogs, and farm animals, such as chickens and pigs. They have reduced flight

capacity, with an average dispersion of around 400 meters. During the day, sandflies remain sheltered, showing greater activity from early evening until dawn. During this period, females go in search of food, preferably the blood of animals, including humans. Brazil is among the countries that concentrate the largest number of sandfly species in the world [11-14].

The main species of sandflies associated with the transmission of thermotropic leishmaniasis in humans are:*Lutzomyia intermedia* (Lutz & Neiva, 1912); *Lutzomyia neiva*i (Pinto, 1926); *Lutzomyia whitmani* (Antunes & Coutinho, 1939); *Lutzomyia umbratilis* Ward & Fraiha, 1977; *Lutzomyia flaviscutellata* (Mangabeira, 1942); *Lutzomyia antunesi* (Coutinho, 1939); *Lutzomyia migonei* (France, 1920); *Lutzomyia fischeri* (Pinto, 1926); *Lutzomyia personali* (Coutinho & Barretto, 1940); *Lutzomyia wellcomei*; *Lutzomyia complexa* (Mangabeira, 1941*); Lutzomyia zai* (Barretto & Coutinho, 1940); *Lutzomyia paraensis* (Barretto & Coutinho, 1940);; *Lutzomyia amazonensis* (Root, 1934); *Lutzomyia hirsuta* (Mangabeira, 1942); *Lutzomyia ubiquitalis* (Mangabeira, 1942); *Lutzomyia gomezi*; *Lutzomyia tuberculata* (Mangabeira, 1941) (Figure 3) 14-16].

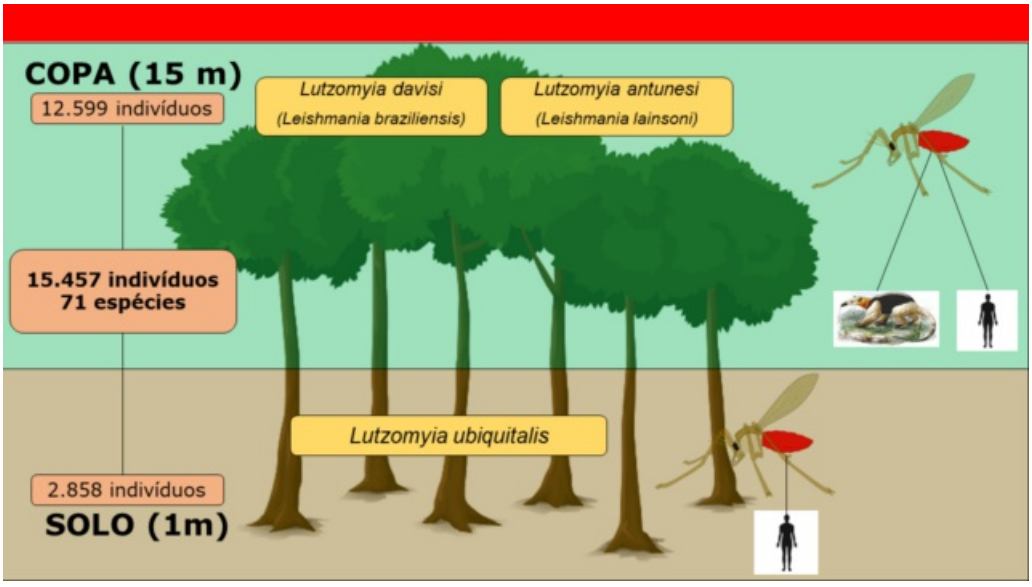

Figure 3. Results showed greater capture of insects at a height of 15 meters:*Lutzomyia davis* (Davis 1993) and *Lutzomyia antunesi* (Coutinho, 1939). Sources: Image: Fapero reproduction and https://confap.org.br/news/estudo-da-fiocruz-em-rondonia-mostra-avancos-no-conhecimento-dos-transmissores-de-leishmaniose/.

American cutaneous leishmaniasis (ATL) is a diseaseinfectious-parasitic disease that affects humans and is caused by several species of protozoa of the genus Leishmania (Ross, 1903). All species in the genus are transmitted by bite from infected females of dipterans of the Phlebotominae subfamily, belonging to the genera *Lutzomyia* França, 1924, in the New World and *Phlebotomus* Rondani & Berté 1840, in the Old World (Figure 4A) (14-16).

Figure 4A. Preference of *Lutzomyia longipalpis* (the sand fly that transmits leishmaniasis) for *Cannabis sativa* L., 1753 (Rosales: Cannabaceae), over other plants. During the study, researchers found *Cannabis Sativa* DNA in a significant percentage of sandflies at five out of six sites in different parts of the world*. Cannabis* DNA was found in sand flies in five out of six locations in different parts of the world. This occurred in two species that transmit cutaneous leishmaniasis in the Middle East and Central Asia and two species that transmit visceral Leishmaniasis in Africa and South America. Source:nhttps://sbmt.org.br/leishmaniose-pesquisa-revela-preferencia-da-lutzomyia-longipalpis-pela-cannabis/.

In Brazil, the literature scientifically incriminates*Lutzomyia longipalpis* (Lutz & Neiva, 1912), as the main vector of visceral leishmania as it was often found naturally infected and was collected from sucking dogs and foxes that are reservoirs of Lutzomyia infantum, having a high degree of anthropophilia has been incriminated as a transmitter in some municipalities in the Central-West region. Some researchers, however, disagree with this anthropophilic term and consider *L. longipalpis* an eclectic species. The geographic distribution of sandfly species is influenced by several factors, such as physical barriers, precipitation, vegetation, light, and abundance of vertebrate hosts. The presence of animals in the home plays a fundamental role in the epidemiology of leishmaniasis, as it allows the concentration of many sandflies in this environment. This abundance of domestic animals, mainly dogs, and birds, in the home, would promote the attraction of sandflies, attracted by the kairomones odor or the release of CO2 (16-19).

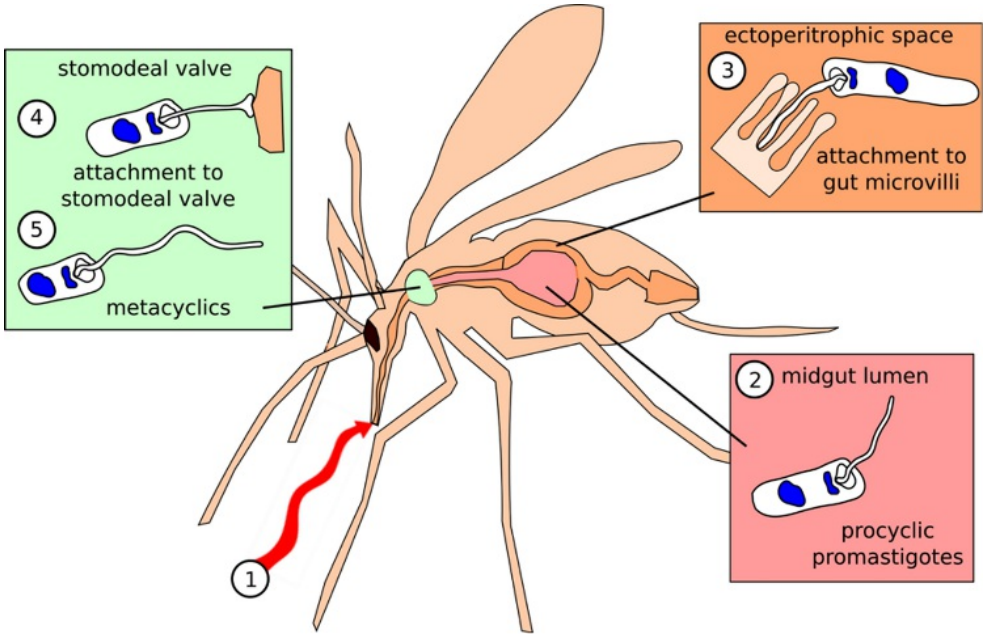

Figure 4B. Schematic of the three major Leishmania stages in sand flies. Shortly after ingestion (1, red arrow) of the blood meal, promastigotes are localized in the midgut lumen, in the blood-meal bolus surrounded by peritrophic matrix (2; 1 day post blood meal [PBM]). Leishmania waits until the peritrophic matrix is broken down, and then at the end of the digestive process, it enters the endoperitrophic space and attaches to the epithelial wall (3; > 4 days PBM). Finally, where parasites have migrated anteriorly to the thoracic midgut, the stomodeal valve of the fly and the human-infective metacyclic forms differentiate from the earlier stages (4, 5; > 9 days PBM). Source: DOI: https://doi.org/10.1186/s13071-020-04498-0 and https://parasitesandvectors.biomedcentral.com/articles/10.1186/s13071-020-04498-0#citeas

Leishmania Ross, 1903 (Kinetoplastida: Trypanosomatidae) transmitted to humans by female hematophagous sandflies of the subfamily Phlebotominae (Diptera: Psychodidae), an obligatory intracellular parasite of cells of the mononuclear phagocytic system, with two main forms: a flagellate or promastigote, found in the digestive tract of the insect vector, and another aflagellate or amastigote observed in vertebrate hosts. In Brazil, eight species of Leishmania are known and can be transmitted to mammals, including humans, through the bite of several species of sandfly females in search of blood. Among them, the most important are *Leishmania chagasi* Cunha & Chagas, 1937 (causing VL), *Leishmania braziliensis*, Vianna, 1911, *Leishmania guyanensis* Floch, 1954, *Leishmania (leishmania) amazonensis* Lainson, and Shaw, 1972, and Personal protection: use a mosquito net with fine mesh screen doors and windows with fine mesh, use repellents do not expose yourself during vector activity times dusk and night (Figure 5) [19-22].

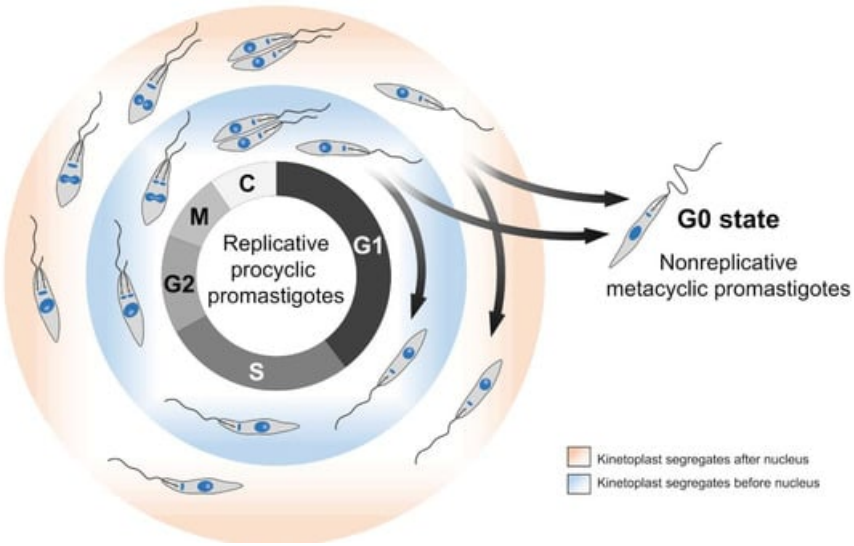

Figure 5. Scheme showing the two distinct patterns of kinetoplast segregation relative to the nucleus in promastigotes of Leishmania spp. throughout the cell cycle. Most *Leishmania* Ross, 1903, species present one of the two kinetoplast segregation patterns: kinetoplast segregates after the nucleus (light red) or kinetoplast segregates before the nucleus (light blue). For instance, *Leishmania mexicana* Garnham, 1962, segregates its kinetoplast predominantly after the nucleus [37], while *Leishmania major* Vasili and Schokhor, 1914 and *Leishmania tarentolae* Wenyon, 1921, do the opposite [38,39]. However, *Leishmania donovani* Laveran and Mesnil, 1903,and *Leishmania amazonensis* Lainson and Shaw, 1972, exhibit these two patterns distributed in the population [40,41]. Source: Assis LHC, et al. Cell Cycle, Telomeres, and Telomerase in Leishmania spp.: What Do We Know So Far? Cells. 2021: 10(11): 3195 and https://www.mdpi.com/2073-4409/10/11/3195.

The identification of the blood food sources of sandflies is of great epidemiological importance, allowing us to understand their relationship with reservoir hosts and the transmission dynamics of the disease. Molecular techniques, such as the Polymerase Chain Reaction (PCR), have been used to identify the blood food sources of these insects. Furthermore, molecular biology techniques have also been applied to the detection and identification of infections by *Leishmania* spp. in sandflies. PCR-RFLP is one of the most used techniques for this purpose, allowing the identification of the parasite's DNA in the insect vector. The faunal diversity of sandflies varies according to the region and human interference in the environment[19-25].

In cutaneous leishmaniasis, the wild animals that act as reservoirs are wild rodents, anteaters, and sloths. In visceral leishmaniasis, the main source of infection is the field fox. Although some canids, foxes, dogs, rodents, edentulous animals, anteaters, sloths, and equids can be a reservoir of the protozoan and a source of vector infection, in urban centers transmission becomes potentially dangerous due to the large number of dogs, which acquire the infection. and develop a clinical picture like that of humans (Figure 6) [24-25].

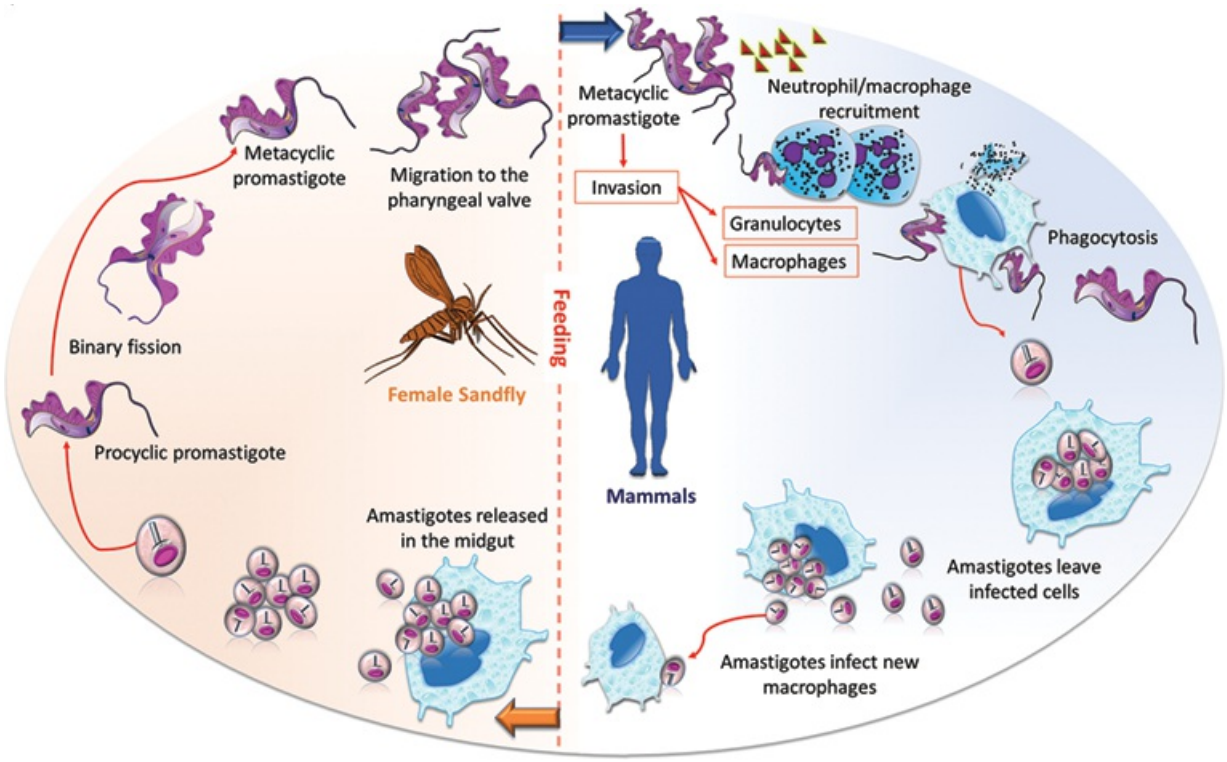

Figure 6. Life cycle of *Leishmania* spp. Leishmania parasites are transmitted by the bites of infected female sandflies during their blood meals. The vector injects the metacyclic promastigote forms, which are engulfed by phagocytic cells at the bite site. Inside the cells, promastigotes transform into amastigotes, the tissue stage of the parasite, which will then reproduce by binary fission and progress to infect other mononuclear phagocytic cells. Interactions between parasite, host, and other factors will determine whether the infection progresses to cutaneous or visceral leishmaniasis. Sandflies become infected by ingesting infected cells during blood meals. In the digestive tract of the vector, amastigotes differentiate into promastigotes and migrate to the proboscis, from where they are injected into the hosts during the bite. Sources: DOI: 10.5772/65787 and https://www.intechopen.com/chapters/53752.

The disease is not contagious or transmitted directly from one person to another, from one animal to another, or from animals to people. Transmission of the parasite occurs only through the bite of an infected female mosquito. The incubation period is usually 2 to 4 months, but it can range from 10 days to 24 months [25-27].

The disease mainly affects dogs, but also wild and urban animals such as cats, rats, and humans. It is estimated that there are an average of 200 infected dogs for each case in humans. If the person is not treated, visceral disease can kill in 90% of cases. Canine visceral leishmaniasis is considered more important than human disease, since, in addition to being more prevalent, there is a huge number of dogs infected with the skin parasite, which end up serving as a source of contamination for mosquito vectors. Therefore, the domestic dog is the main reservoir of the parasite [27-29].

In the Americas, visceral leishmaniasis is the most serious form of the disease and can be fatal if not treated properly. It is endemic in several countries and is called American visceral leishmaniasis. The etiological agent *is L. chagasi*. In Brazil, transmission has been attributed mainly to *L. longipalpis*, but other species of sandflies are considered potential

transmitters of the disease [28-30].

In Pará, *Lutzomyia antunesi* (Coutinho, 1939) may be an alternative vector of LVA. In Mato Grosso do Sul, in the region of Corumbá and Ladário, *Lutzomyia cruzi* (Mangabeira, 1938) is the predominant species and sympatric with *Lutzomyia forattinii* (Galati, Rego, Nunes & Teruya, 1985); both have been considered to transmit the agent of visceral leishmaniasis, *Lutzomyia almerioi* (Galati & Nunes, 1999), presented natural infection by *L. chagasi* and *Leishmania* sp., high density and marked anthropophilic, suggestive of the possibility of involvement in the transmission of the disease in this region (Figure 7) [28-30].

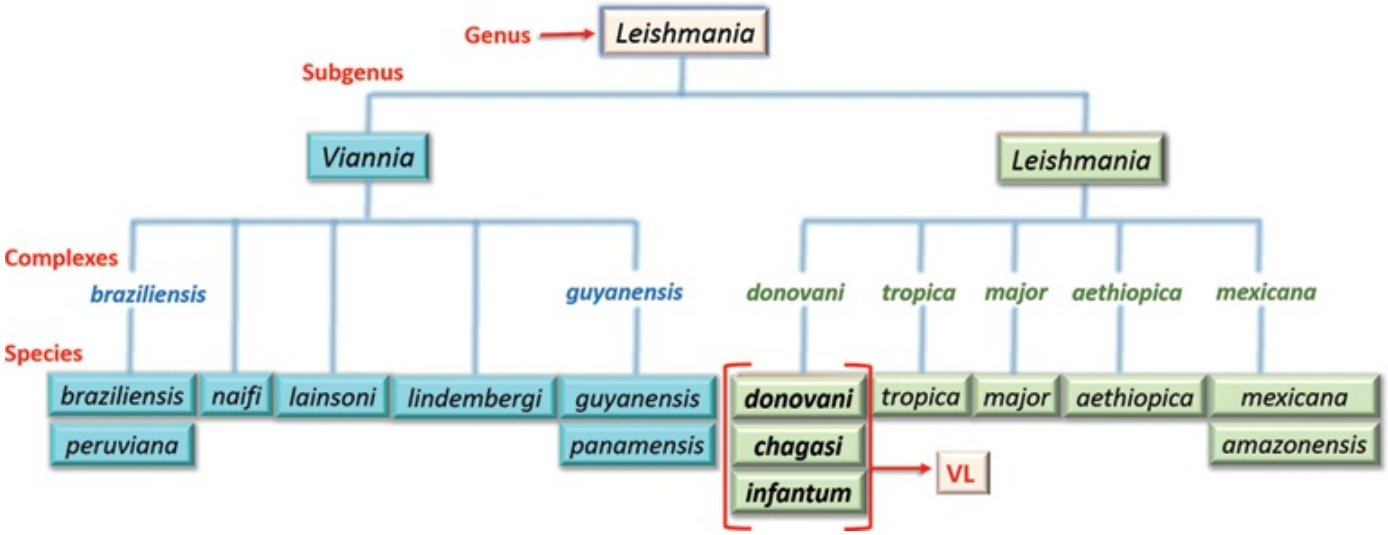

Figure 7. Main species of the *Leishmania* Ross, 1903. Those causing visceral leishmaniasis (VL): *Leishmania (Leishmania) donovani* (Laveran & Mesnil, 1903), in Asia, *Leishmania chagasi* Cunha & Chagas, 1937 in the Americas, and *Leishmania infantum* (Nicole, 1908), in Asia, Europe, and Africa. Note: *L. infantum* nomenclature was proposed by Marcili et al. using phylogenetic analysis of Leishmania species occurring in Latin America. Sources: DOI: 10.5772/65787 and https://www.intechopen.com/chapters/53752.

Symptoms: Visceral leishmaniasis (LVA): Irregular and prolonged fever; anemia; indisposition; paleness of the skin and/or mucous membranes; lack of appetite; weight loss; swelling of the abdomen due to enlargement of the liver and spleen. Cutaneous leishmaniasis: Two to three weeks after the sandfly bite, a small, raised papule appears on the reddish skin that increases in size until it forms a wound covered by a crust or purulent secretion. The disease can also manifest as inflammatory lesions on the mucous membranes of the nose or mouth [28-32].

In the context of public health, American cutaneous Leishmania (LTA) has significantly increased its importance due to its growth. urbanization process. Initially, the disease was predominantly rural, and, more recently, it has been expanding to medium and large urban areas. Rapidly expanding in geographic location in Brazil, LTA is considered one of the most important dermatological treatments, not only in terms of frequency but mainly due to the therapeutic difficulties, deformities, and consequences that can cause socioeconomic pressures that lead to the expansion of areas of endemic diseases and the emergence of new outbreaks. In this way, sandfly species, which somehow resist adverse conditions,

can explore new environments, moving closer and closer to homes, facilitating the transmission of diseases [28-32].

Environmental management to control the vector: clean backyards, lands, and public squares by collecting leaves and branches, eliminate organic solid waste, and provide appropriate disposal for this waste avoid excessive shading of the patio, and eliminate sources of moisture. The sources of infection for leishmaniasis are mainly wild animals and sandflies that harbor the parasite in their digestive tract, however, the host can also be domestic dogs and horses [28-32].

References

[1] Missawa NA, Maciel GBML. List of species in the genus*Lutzomyia*, França, 1924 (Psychodidae, Phlebotominae) from the State of Mato Grosso. Journal of the Brazilian Society of Tropical Medicine. 2007; 40(1): 11-14.

[2] Sants DR. Training Course: Collection and Identification of Sand flies. 1st ed. Campo Grande: Juris Abientes Consultores. 2014.

[3] Assis LHC, et al. Cell cycle, Telomeres, and telomerase in*Leishmania* spp.: What Do We Know So Far? Cells. 2021: 10(11): 3195 and https://www.mdpi.com/2073-4409/10/11/3195.

[4] Claborn DM. The Biology and Control of Leishmaniasis Vectors. Journal of Global Infectious Diseases.2010; 2 2): 127–134.

[5] Killick-Kendrick R, et al. Guide to the identification and geographic distribution of*Lutzomyia* sand flies in Mexico, the West Indies, Central and South America (Diptera: Psychodidae). Transactions of The Royal Society of Tropical Medicine and Hygiene.1994; 8(10): 125.

[6] Rio Grande do Sul Health Department [Internet]. Porto Alegre: Government of the State of Rio Grande do Sul; @2024 [cited 2024 Mar 28]. Available from https://saude.rs.gov.br/leishmanione-visceral.

[7] Lainson R, Rangel EF. *Lutzomyia longipalpis* and the eco-epidemiology of American visceral leishmaniasis, with particular reference to Brazil: a review. Memorias do Instituto Oswaldo Cruz. 2005; 100(8): 811-827.

[8] Rocha L. Leishmaniasis: learn about the insects that transmit it and learn how to prevent it [Internet]. Rio de Janeiro: Instituto Oswaldo Cruz; @2019 [cited 2024 Mar 28]. Available from https://portal.fiocruz.br/noticia/leishmanioses-conheca-os-insetos-transmissores-e-saiba-como-se-prevenir.

[9] Castro R. Increase in cases of leishmaniasis in dogs raises awareness about the disease in humans [Internet]. Rio de Janeiro: Instituto Oswaldo Cruz; @2023 [cited 2024 Mar 28]. Available from https://portal.fiocruz.br/noticia/aumento-de-casos-de-leishmaniose-em-caes-acende-alerta-para-doenca-em-humanos.

[10] Galati EAB. Morphology and terminology of phlebotominae (Diptera: Psychodidae). Classification and identification of taxa from the Americas. Postgraduate Program in Public Health. 1st ed. São Paulo: Faculty of Public Health of the University of São Paulo. 2018.

[11] Lidani KCF, Andrade FA, Tizzot MRPA, Costa-Ribeiro MCV, Beltrame MH, Messias-Reason IJ. Visceral leishmaniasis and natural infection rates of *Leishmania* in *Lutzomyia longipalpis* in Latin America [Internet]. The Epidemiology and Ecology of Leishmaniasis. InTech; 2017. Available from: http://dx.doi.org/10.5772/65787.

[12] Dorval MEC. Modification of Disney trap for capture of sand flies (Diptera: Psychodidae: Phlebotominae). Memoirs of the Institute Oswaldo Cruz. 2007; 102: 877–878.

[13] Silva JGD, Werneck GL Cruz MSP, Costa CHN, Mendonça IL. Natural infection of*Lutzomyia longipalpis* by *Leishmania* sp. in Teresina, Piauí, Brazil. Public Health Notebooks. 2007; 23(7):1715-1720,

[14] Mohammad A, et al. A historical overview of the classification, evolution, and dispersion of*Leishmania* parasites and sandflies. PLoS Neglected Tropical Diseases. 2016; 10(60): e0004770.

[15] Killick-Kendrick R. Guide to the Identification and geographic distribution of*Lutzomyia* sand flies in Mexico, the West Indies, Central and South America (Diptera: Psychodidae). Transactions of The Royal Society of Tropical Medicine and Hygiene.1994; 8(10): 125.

[16] Costa GS, Pereira Júnior AM, Pessoa FAC, Shimabukuro PHF, Medeiros JF. New records of phlebotomine sand flies (Diptera: Psychodidae) from the Western Brazilian Amazon and the description of the female of *Pintomyia fiocruzi*. Journal Medical Entomology. 2020, 57(4):1328-1333.

[17] Almeida PS, et al. Species of sandflies (Diptera, Psychodidae) collected in an urban environment in municipalities with transmission of visceral Leishmaniasis in the State of Mato Grosso do Sul, Brazil. Brazilian Journal of Entomology. 2010; 54(2): 304–310.

[18] Oliveira MR, Soares MRA. Technological innovations applied to the study of vector insects: a technological prospection on different traps for sandflies (Diptera: Psychodidae). Prospecting Notebooks. 2023; 16(6): 2003–2016. 2023; 16(6): 2003–2016.

[19] Days ES. Sandflies (Diptera: Psychodidae) in an outbreak of cutaneous leishmaniasis in the State of Minas Gerais. Journal of the Brazilian Society of Tropical Medicine. 2007; 40(1): 49-52.

[20] Oliveira GMG, et al. Survey of phlebotomine sand flies (Diptera: Psychodidae: Phlebotominae) in Três Lagoas Municipality, Mato Grosso do Sul State, Brazil, an area of intense transmission of American visceral leishmaniasis. Pan-Amazonian Health Magazine. 2010; 1(3): 83-94.

[21] Costa SM, Cordeiro JLP, Rangel EF. Environmental suitability for*Lutzomyia (Nyssomyia) whitmani* (Diptera: Psychodidae: Phlebotominae) and the occurrence of american cutaneous leishmaniasis in Brazil. Parasites & Vectors. 2018; 11: 155.

[22] Alves VR, Freitas RA, Santos FL, Oliveira AFJ, Barrett TV, Shimabukuro PHF. Sand flies (Diptera, Psychodidae, Phlebotominae) from Central Amazonia and four new records for the Amazonas state, Brazil. Revista Brasileira de

Entomology; 2012; 56(2): 220-227.

[23] Vilela ML, Pita-Pereira D, Azevedo CG, Godoy RE, Britto C, Rangel EF. The phlebotomine fauna (Diptera: Psychodidae) of Guaraí, state of Tocantins, with an emphasis on the putative vectors of American cutaneous leishmaniasis in rural settlement and periurban areas. Memories of the Oswaldo Cruz Institute. 2013; 108(5): 578-585.

[24] Tonelli GB, Binder C, Nogueira VLC, Prado MH, Theobald GG, Fields AM, et al. The sand fly (Diptera: Psychodidae) fauna of Lassance, Northeast Minas Gerais, Brazil. PLoS One. 2020; 16(10): e0257043.

[25] Pirajá GV, Lucheis SB. Epidemiological surveillance of phlebotomine as planning and control actions in leishmaniasis. Veterinary and Animal Science Magazine. 2024; 21(4): 503-155.

[26] Ndrade-Filho JD, Galati EAB, Falcão AL. *Nyssomyia intermedia* (Lutz & Neiva, 1912) and *Nyssomyia neivai* (Pinto, 1926) (Diptera: Psychodidae: Phlebotominae) geographical distribution and epidemiological importance. Memories of the Oswaldo Cruz Institute. 2007; 102(4): 481-487.

[27] Shimabukuro P, Tolezano J, Galati E. Illustrated identification key for the Phlebotominae (Diptera, Psychodidae) from the state of São Paulo, Brazil. Zoology Loose Papers. 2010; 51(27): 399-441.

[28] Galardo AKR, Galardo C, Santana AA, Mende JCC. First occurrence of *Lutzomyia (Lutzomyia) longipalpis* Lutz & Neiva, 1912 (Diptera: Psychodidae: Phlebotominae) in the State of Amapá, Brazil. Amazona Biota. 2013; 3(2): 179-183

[29] Abbasi I, Cunio R, Warbug A. Identification of blood meals imbibed by Phlebotominae sand flies using Cytochrome B PCR and reverse line blotting. Vector-borne and zoonotic diseases. 2009; 9(1): 79-86.

[30] Almeida PS, Nascimento JC, Ferreira AD, Minzão LD, Portes F, Miranda AM, Faccenda O, Andrade Filho JD. Species of sandflies (Diptera, Psychodidae) collected in an urban environment in municipalities with transmission of visceral leishmaniasis in the State of Mato Grosso do Sul, Brazil. Magazine Brazilian Entomology; 2010; 54: 304-310.

[31] Assis TSM, Caligiorne RB, Romero GAS, Rabello A. Detection of Leishmania DNA in human serum samples for the diagnosis of visceral leishmaniasis. Transactions of The Royal Society of Tropical Medicine and Hygiene. 2009; 103(12): 1269-1272.

[32]. Barata RA, França-Silva JC, Mayrink W, Silva JC, Prata A, Lorosa ES, Fiúza JA, Gonçalves CM, Paula KM, Dias ES Aspects of the ecology and behavior of sandflies in an endemic area for visceral leishmaniasis, Minas Gerais. Journal of the Brazilian Society of Tropical Medicine. 2005; 38(5): 421-425.

[33] Sloan MA., Sadlova J, Lestinova T., et al. The *Phlebotomus papatasi* systemic transcriptional response to trypanosomatid-contaminated blood does not differ from the non-infected blood meal. Parasites Vectors. 2021; 14, 15.