

Abundance of *Lutzomyia longipalpis* (Diptera: Psychodidae: Phlebotominae) and urban transmission of visceral leishmaniasis in Campo Grande, state of Mato Grosso do Sul, Brazil

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The outspread and urbanization of visceral leishmaniasis (VL) in Campo Grande, state of Mato Grosso do Sul, lead us to undertake the present study over diversity and abundance of sand flies in the urban area to compare with previous search carried out during 1999/2000, before the identification of the disease in the human population. The captures were carried out with automatic light traps, weekly, from February 2004 to February 2005 on three sites including a forested area (Zé Pereira), two peridomicilies (shelters of domestic animals and cultivation areas), and intradomicilie. In the present study 110 collections were obtained during 13 months for 1320 h of collections, resulting in 5004 specimens, 3649 males and 1355 females belonging to the 20 following species: Brumptomyia avellari, Brumptomyia sp., Bichromomyia flaviscutellata, Evandromyia lenti, E. termitophila, E. cortezzii, E. borrouli, Lutzomyia sp., L. longipalpis, Micropygomyia quinquefer, N. antunesi, N. whitmani, Pintomyia christenseni, Pi. damascenoi, Psathyromyia aragai, Ps. campograndensis, Ps. hermanlenti, Ps. shannoni, Pychodopygus clautrei, and Sciopemyia sordellii. L. longipalpis was the most abundant species in the anthropic environment with 92.22% of the captures. This shows an increase of sixty times in the density of L. longipalpis compared to the last sand fly evaluation in 1999/2000. The high density of L. longipalpis in Campo Grande is the main factor of risk in transmission of the disease to human in the urban area. The capture of N. antunesi, typical specie from Amazonian region, in Mato Grosso do Sul is reported for the first time.

Key words: Phlebotominae - *Lutzomyia longipalpis* - visceral leishmaniasis - Mato Grosso do Sul - Brazil

Leishmaniasis are currently one of the biggest problems of public health in the state of Mato Grosso do Sul. Cutaneous leishmaniasis (CL) has been reported in practically all the cities of the state and visceral leishmaniasis (VL), since the decade of 1980 has shown to be highly endemic in the cities of Corumbá and Ladário (northwest of the state). The disease is now expanding for new areas of the state, also reaching the capital, Campo Grande, that registered the first canine autochthonous case in 2000 (Silva et al. 2000) and the first human case in 2002. In this year, 19 cases were diagnosed in the city, and during the years of 2003 and 2004, 100 and 126 cases had been notified, respectively, confirming the endemicity and expansion of the disease (Campo Grande 2006). The presence of the *Lutzomyia longipalpis*, main vector of VL, in the city was evidenced by Oliveira et al. (2000) and from this finding, studies on the sand fly fauna were developed in the urban area

and 28 species of sand fly were identified (Oliveira et al. 2003).

With the objective to compare the current abundance of *L. longipalpis* with the previous period of the beginning of the outbreak of VL in the urban area of Campo Grande, a longitudinal study of its sand fly fauna was carried out from February 2004 to February 2005.

MATERIALS AND METHODS

Study area

General characteristics - The city of Campo Grande, state of Mato Grosso Sul, Brazil, with 8.118,4 km², is located geographically in the central of state, occupying 2.27% of its total area and, according to the Instituto Brasileiro de Geografia e Estatística with an urban population of 734,164 inhabitants in 2004 (Campo Grande 2005). The headquarters of the city are defined by the geographic coordinates 20°26'34" South latitude and 54°38'47" longitude West. The predominant climate of Campo Grande, according to the classification of Köppen, is of the rainy tropical type of Savannah, subtype AW, characterized for bad annual distribution of rains, with occurrence defined of a dry period during the cold months of the year and a rainy period during the summer months. The annual average temperature is around 23°C, December is the hottest month with approximately 25°C, and the coldest is June, with 18°C.

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Studied areas - The captures had been carried out in three points selected from previous work, where we observed a great density and diversity of sand flies (Oliveira et al. 2003): Mata do Zé Pereira, situated in the west region, an outskirt district created by a social program. It consist a part of a remaining forest rich in local fauna and flora; Chácara das Palmeiras, south region of the city, an area of 1 ha, with two houses, orchard with some species of trees fruit, kennel, goat pen, and a chicken house that make border with a residual forest of the neighboring land and Região Central (Taveirópolis district), a house with a chicken house in its yard.

Captures - In each selected fixed point, CDC-like light traps were set simultaneously in the interior of the bush (at ground level and tree canopy), in the edge of the bush (ground level) and man modified environment as: culture areas, chicken house, pigsty, kennel, goat pen, inside house, and balcony. The traps of the canopies had been installed approximately 5 m of height and the traps of the ground 1 m. The collections had been weekly, from 6.00 pm to 6.00 am, without obeying the summer schedules, in the period of February of 2004 to February of 2005. The captured specimens had been carried to the Laboratory of Parasitology of the Federal University of Mato Grosso of Sul for identification. The nomenclature of the adopted species was of Galati (2003).

The comparative analysis in relation to the density of *L. longipalpis* in the two studied years was gotten through the qui-square test (χ^2). For the evaluation of the most abundant species of sand flies, as well as its spacial distribution, the Index of Species Abundance (ISA) converted into a scale between zero and 1 was used, through the Standardised Index of Species Abundance (SISA). In this index, value 1 corresponds to the species most abundant (Roberts & Hsi 1979). For the analysis of diversity and regularity of the species in the respective ecotopes were used the Shannon's Index (H), Pielou's (J), Buzas and Gibson's (E) (Hayek & Buzas 1997).

RESULTS

In 110 captures carried through, during 1320 h, a total of 5004 sand flies, 3649 males and 1355 females were collected. The sand fly species belong to four sub tribes, ten genera and 20 species: BRUMPTOMYIINA *Brumptomyia avellari* (Costa Lima, 1932), *Brumptomyia* sp, LUTZOMYIINA *Evandromyia lenti* (Mangabeira, 1938), *Evandromyia termitophila* (Martins, Falcão & Silva, 1964), *Evandromyia cortelezzi* (Brèthes, 1923), *Lutzomyia longipalpis* (Lutz & Neiva, 1912), *Pintomyia*

christenseni (Young & Duncan, 1994), *Sciopemyia sordellii* (Shannon & Del Ponte, 1927), PSYCHODOPYGINA *Nyssomyia antunesi* (Coutinho, 1939), *Nyssomyia whitmani* (Antunes & Coutinho, 1939), *Psathyromyia aragaoi* (Costa Lima, 1932), *Psathyromyia campograndensis* (Oliveira, Andrade Filho, Falcão & Brazil, 2001) *Psathyromyia hermanlenti* (Martins, Silva & Falcão, 1970), *Psathyromyia shannoni* (Dyar, 1929), *Psychodopygus clautrei* (Abonnenc, Léger & Fauran, 1979) and SERGENTOMYIINA *Micropygomyia quinquefer* (Dyar, 1929).

Between February of 2004 and February of 2005, *L. longipalpis* was the species most frequent in the area studied, with 92.22 % of captured specimens. This fact contrasts with the data obtained in the period of February of 1999 the February of 2000 where the species only represented 8.97% of the captured sand flies (Table V).

The difference in the captures of *L. longipalpis* in the three environments studied during the two investigated periods was statistically significant ($p < 0.001$) as demonstrated in Table I.

It is possible to observe also, in Table II, that the *L. longipalpis* passed from the 5th position in 1999/2000 to the first, becoming the most abundant species in the urban area of the city in the period of 2004/2005.

The total of sand flies captured weekly in the urban area of Campo Grande, the two studied periods (99/00 and 04/05) and the diversity Index (H, E, J) is represented in Tables III and IV.

The diversity Index had been higher in the forested area than in the central area in both periods. The index of regularity (J) in the central region (0,0027) was significantly lesser in the second period in relation to the first one in the same area (0,5156), demonstrating one strong dominance of one species, in the case of *L. longipalpis*.

DISCUSSION

The weekly collection with light traps in the same ecotopes of the city of Campo Grande, in the period of February of 2004 to February of 2005, confirmed the varieties of species in the urban environment as observed in the period of 1999/2000 (Oliveira et al. 2003).

In comparison of the two studied periods, considering the same months and ecotopes, it was observed that some previously species reported by Oliveira et al. (2003) such as *Br. brumpti*, *Br. pinto*, *L. cruzi*, *E. corumbaensis*, *E. teratodes*, *Ex. cerradincola*, *Mi. migonei*, and *Ps. punctigeniculata* was not captured

TABLE I

Total number of *Lutzomyia longipalpis* captured in three environments of Campo Grande, state of Mato Grosso do Sul

| | Mata do Zé Pereira | Região Central | Chácara das Palmeiras | Total |
|--------------------------------|--------------------|----------------|-----------------------|-------|
| Feb/1999-Feb/2000 | 0 | 54 | 17 | 71 |
| Feb/2004-Feb/2005 ^a | 112 | 4385 | 118 | 4615 |
| Total | 112 | 4439 | 135 | 4686 |

χ^2 : 115,75; $p < 0.001$; ^a: with a total of 1320 h of captures.

TABLE II
Standardised Index of Species Abundance (SISA) of sand flies captured in Campo Grande, state of Mato Grosso do Sul

| Species | SISA | Position | SISA | Position |
|-------------------------------------|-------------|------------|-------------|------------------|
| <i>Bumptomys avellari</i> | 0,41 | 5th | 0,27 | 15th |
| <i>Psathyromyia hermanlenti</i> | 0,29 | 8th | 0,35 | 11th |
| <i>Psychodopygus clausi</i> | 0,31 | 7th | 0,32 | 12th |
| <i>Lutzomyia longipalpis</i> | 1 | 1st | 0,62 | 5th |
| <i>Ps. aragaoi</i> | 0,27 | 9th | 0,54 | 7th |
| <i>Micropygomyia quinquefer</i> | 0,22 | 10th | 0,22 | 17th |
| <i>Pintomyia christenseni</i> | 0,02 | 14th | 0,39 | 10th |
| <i>Nyssomyia antunesi</i> | 0,22 | 9th | 0 | |
| <i>N. whitmani</i> | 0,49 | 3rd | 0,13 | 19th |
| <i>Ps. shannoni</i> | 0,45 | 4th | 0,51 | 8th |
| <i>Ps. campograndensis</i> | 0,16 | 11th | 0,28 | 14th |
| <i>Brumptomys sp.</i> | 0,11 | 12th | 0,4 | 9th |
| <i>Evandromyia lenti</i> | 0,4 | 6th | 0,76 | 2nd |
| <i>E. cortelezzii</i> | 0,16 | 10th | 0,63 | 4th |
| <i>Sciopemyia sordellii</i> | 0,27 | 8th | 0,65 | 3rd |
| <i>P. damasceni</i> | 0,02 | 14th | 0,4 | 9th |
| <i>Lutzomyia sp.</i> | 0,02 | 14th | 0 | |
| <i>E. bourrouli</i> | 0,02 | 14th | 0,16 | 18th |
| <i>E. termitophila</i> | 0,58 | 2nd | 0,61 | 6th |
| <i>Bichromyia flaviscutellata</i> | 0,08 | 13th | 0,22 | 17th |
| <i>Mcropygomyia longipennis</i> | 0 | | 0,95 | 1st |
| <i>E. teratodes</i> | 0 | | 0,31 | 13th |
| <i>B. pinto</i> | 0 | | 0,04 | 22th |
| <i>E. corumbaensis</i> | 0 | | 0,1 | 20th |
| <i>Migonemyia migonei</i> | 0 | | 0,05 | 21st |
| <i>Expapillata cerradincola</i> | 0 | | 0,05 | 21sh |
| <i>Ps. punctigeniculata</i> | 0 | | 0,23 | 16th |
| <i>B. brumpti</i> | 0 | | 0,05 | 21 st |
| <i>L. cruzi</i> | 0 | | 0,05 | 21 st |

TABLE III
Number of sand flies captured in the urban areas of Campo Grande, state of Mato Grosso do Sul, in the period of Feb/2004 to Feb/2005

| Species | Mata do Zé Pereira | Região central | Chácara das Palmeiras | Total |
|-----------------------------------|--------------------|----------------|-----------------------|--------|
| <i>Brumptomys avellari</i> | 8 | 0 | 1 | 9 |
| <i>Psathyromyia hermanlenti</i> | 55 | 0 | 0 | 55 |
| <i>Psychodopygus clausi</i> | 89 | 0 | 0 | 89 |
| <i>Lutzomyia longipalpis</i> | 112 | 4385 | 118 | 4615 |
| <i>Ps. aragaoi</i> | 36 | 0 | 0 | 36 |
| <i>Micropygomyia quinquefer</i> | 10 | 0 | 0 | 10 |
| <i>Piintomyia christenseni</i> | 1 | 0 | 0 | 1 |
| <i>Nyssomyia antunesi</i> | 10 | 0 | 0 | 10 |
| <i>N. whitmani</i> | 11 | 0 | 2 | 13 |
| <i>Ps. shannoni</i> | 4 | 0 | 68 | 72 |
| <i>Ps. campograndensis</i> | 6 | 0 | 0 | 6 |
| <i>Brumptomys sp.</i> | 3 | 0 | 0 | 3 |
| <i>Evandromyia lenti</i> | 3 | 0 | 36 | 39 |
| <i>E. cortelezzii</i> | 6 | 0 | 0 | 6 |
| <i>Sciopemyia sordellii</i> | 1 | 0 | 3 | 4 |
| <i>P. damasceni</i> | 1 | 0 | 0 | 1 |
| <i>Lutzomyia sp.</i> | 1 | 0 | 0 | 1 |
| <i>E. bourrouli</i> | 1 | 0 | 0 | 1 |
| <i>E. termitophila</i> | 0 | 1 | 30 | 31 |
| <i>Bichromyia flaviscutellata</i> | 2 | 0 | 0 | 2 |
| Total | 360 | 4386 | 258 | 5004 |
| H: Shannon's index | 1,9572 | 0,0019 | 1,2512 | 1 |
| E: Buza & Gibson's index | 0,3226 | 0,5009 | 0,4992 | 0,1359 |
| J: Pielou's index | 0,6647 | 0,0027 | 0,6429 | 0,3338 |

TABLE IV
Number of sand flies captured in the urban areas of Campo Grande, state of Mato Grosso do Sul, in the period of Feb/1999 to Feb/2000

| Species | Mata do Zé Pereira | Região Central | Chácara das Palmeiras | Total |
|-----------------------------------|--------------------|----------------|-----------------------|--------|
| <i>Brumptomyia avellari</i> | 4 | 0 | 2 | 6 |
| <i>B. brumpti</i> | 0 | 0 | 1 | 1 |
| <i>B. pintoii</i> | 2 | 0 | 0 | 2 |
| <i>Brumptomyia</i> sp. | 13 | 0 | 2 | 15 |
| <i>Psathyromyia hermanlenti</i> | 44 | 0 | 1 | 45 |
| <i>Psychodopygus clautrei</i> | 16 | 0 | 1 | 17 |
| <i>Lutzomyia longipalpis</i> | 0 | 54 | 17 | 71 |
| <i>L. cruzi</i> | 0 | 0 | 1 | 1 |
| <i>Ps. aragaoi</i> | 51 | 0 | 3 | 54 |
| <i>Micropygomyia quinquefer</i> | 9 | 0 | 0 | 9 |
| <i>M. longipennis</i> | 103 | 0 | 1 | 104 |
| <i>Migonemyia migonei</i> | 0 | 0 | 1 | 1 |
| <i>Pintomyia christenseni</i> | 5 | 0 | 3 | 8 |
| <i>Nyssomyia whitmani</i> | 3 | 0 | 1 | 4 |
| <i>Ps. shannoni</i> | 8 | 0 | 52 | 60 |
| <i>Ps. campograndensis</i> | 19 | 0 | 0 | 19 |
| <i>Expapillata cerradincola</i> | 0 | 0 | 1 | 1 |
| <i>Evandromyia lenti</i> | 5 | 1 | 102 | 108 |
| <i>E. cortelezzii</i> | 4 | 3 | 3 | 10 |
| <i>E. corumbaensis</i> | 2 | 0 | 1 | 3 |
| <i>E. teratodes</i> | 2 | 0 | 5 | 7 |
| <i>E. bourrouli</i> | 5 | 0 | 0 | 5 |
| <i>E. termitophila</i> | 0 | 1 | 172 | 173 |
| <i>Sciopemyia sordellii</i> | 4 | 35 | 3 | 42 |
| <i>P. damascenoi</i> | 13 | 0 | 2 | 15 |
| <i>Ps. punctigeniculata</i> | 1 | 0 | 3 | 4 |
| <i>Bichromyia flaviscutellata</i> | 5 | 0 | 1 | 6 |
| Total | 318 | 94 | 379 | 791 |
| H: Shannon's index | 2,2851 | 0,8921 | 1,607 | 2,5279 |
| E: Buza & Gibson's index | 0,4679 | 0,488 | 0,2168 | 0,4639 |
| J: Pielou's index | 0,7505 | 0,5543 | 0,5156 | 0,7669 |

in the present study, although the number of sand flies was approximately six times bigger. As related by Oliveira et al. (2003) the year of 1999 was atypical in Campo Grande with a period of drought, what certainly affected the density of sand flies at the time of captures.

N. whitmani appears as one of the most abundant species (SISA = 0,49), occupying the third position and was present, mainly, in areas next to forest (Table II). In other areas of the state, this species is also one of the most abundant (Galati et al. 1996, 2003, 2006) and was incriminated vector of the CL in several regions of the country (Pessoa & Coutinho 1941, Mayrink et al. 1979, Queiroz et al. 1994, Galati et al. 1996, Luz et al. 2000, Dias-Lima et al. 2003). Even so there is no report of autochthonous cases in the city of Campo Grande, the presence of *N. whitmani* is preoccupying, considering that CL is widely distributed in Mato Grosso Sul and also in neighboring municipalities of Campo Grande. This species is well adapted to the anthropic environment (Souza et al. 2001, Rangel & Lainson 2003, Teodoro et al. 2003) being responsible for the periurban epidemiological pattern of the parasite transmission in several regions of Brazil.

In regard to *Psy. clautrei*, it was observed an increasing in the density when compared the two periods of study, however it was remained restricted to the forest. This species is typically from forest areas having been found naturally infected with parasites of the genus *Leishmania* (Ryan et al. 1987) in the region North of the country, but it is not considered, up to the moment, as vector of *Leishmania*.

N. antunesi, captured only in the forested areas of the Zé Pereira, is reported for the first time in Mato Grosso do Sul. This species is distributed widely through the Amazon region, having been found naturally infected by promastigotes in a endemic area of visceral leishmaniasis in the Island of Marajó, Pará (Ryan et al. 1984) and it was pointed also as a probable vector of *Leishmania (Viannia) lindenbergi* in Belém, Pará (Silveira et al. 2002). The presence of this species in this remaining path of forest in large distance from the Amazon region suggests the continue existence of vegetation in the past, probably destroyed by man action and further studies is necessary to correlates its presence related to the flora and fauna on the region. It is necessary to evaluate the potential of this species as *Leishmania* vector in the area,

TABLE V

Comparative number of sand flies captured in the urban areas of Campo Grande, state of Mato Grosso do Sul in two periods (Feb/1999 to Feb/2000 and Feb/2004 to Feb/2005)

| Species | Feb/2004 to Feb/2005 | | Feb/1999 to Feb/2000 | |
|-----------------------------------|----------------------|-------|----------------------|-------|
| | N | % | n | % |
| <i>Brumptomyia avellari</i> | 9 | 0.18 | 6 | 0.75 |
| <i>Psathyromyia hermanlenti</i> | 55 | 1.1 | 45 | 5.7 |
| <i>Psychodopygus clautrei</i> | 89 | 1.77 | 17 | 2.15 |
| <i>Lutzomyia longipalpis</i> | 4615 | 92.22 | 71 | 8.97 |
| <i>Ps. aragaoi</i> | 36 | 0.72 | 54 | 6.82 |
| <i>Micropygomyia quinquefer</i> | 10 | 0.2 | 9 | 1.13 |
| <i>Pintomyia christenseni</i> | 1 | 0.02 | 8 | 1.01 |
| <i>Nyssomyia antunesi</i> | 10 | 0.2 | 0 | 0 |
| <i>N.whitmani</i> | 13 | 0.26 | 4 | 0.5 |
| <i>Ps.shannoni</i> | 72 | 1.43 | 60 | 7.58 |
| <i>Ps.campograndensis</i> | 6 | 0.12 | 19 | 2.4 |
| <i>Brumptomyia</i> sp. | 3 | 0.06 | 15 | 1.9 |
| <i>Evandromyia lenti</i> | 39 | 0.8 | 108 | 13.65 |
| <i>E.cortelezzii</i> | 6 | 0.12 | 10 | 1.26 |
| <i>Sciopemyia sordellii</i> | 4 | 0.08 | 42 | 5.31 |
| <i>P. damascenoi</i> | 1 | 0.02 | 15 | 1.9 |
| <i>Lutzomyia</i> sp. | 1 | 0.02 | 0 | 0 |
| <i>E.bourrouli</i> | 1 | 0.02 | 5 | 0.63 |
| <i>E.termitophila</i> | 31 | 0.62 | 173 | 21.87 |
| <i>Bichromyia flaviscutellata</i> | 2 | 0.04 | 6 | 0.8 |
| <i>B. brumpti</i> | 0 | 0 | 1 | 0.12 |
| <i>B. pinto</i> | 0 | 0 | 2 | 0.25 |
| <i>L. cruzi</i> | 0 | 0 | 1 | 0.12 |
| <i>M.longipennis</i> | 0 | 0 | 104 | 13.14 |
| <i>Migonemyia migonei</i> | 0 | 0 | 1 | 0.12 |
| <i>Expapillata cerradincola</i> | 0 | 0 | 1 | 0.12 |
| <i>E. corumbaensis</i> | 0 | 0 | 3 | 0.4 |
| <i>E. teratodes</i> | 0 | 0 | 7 | 0.9 |
| <i>Ps.punctigeniculata</i> | 0 | 0 | 4 | 0.5 |
| Total | 5004 | 100 | 791 | 100 |

in view of the proximity of the houses near the forested area.

Bi. flaviscutellata again was captured in one areas (Mata do Zé Pereira) and, even so light trap is not the better method to capture this species, its presence in Campo Grande strengthens its dispersion in Mato Grosso Sul (Dorval et al. 2005, Galati et al. 2006). This species assumes importance in view of its involvement in the transmission of *Le. (Le.) amazonensis*, responsible for the diffuse form of CL in several region of Brazil, and more recently in Bela Vista, Mato Grosso do Sul (Dorval et al. 2006).

The presence of *L. longipalpis* in the city of Campo Grande six years ago (Oliveira et al. 2000) had taken the authors to point out an alert to the Health Authorities in order to prevent the establishment of visceral leishmaniasis in the human population, as observed in other regions of Brazil (Arias et al. 1996). Since our publication in 2000, *L. longipalpis* increased significantly ($p < 0.001$) in its density, practically being captured with exclusiveness in the central areas of the city, what seems of epidemiological importance. In the subsequent years and until the present date more than 246 VL human cases

was reported and around 20% of dogs are seropositive for the disease with an alarming numbers of notifications, reaching children and adults, with early clinical manifestations and lethality index around 11% (Campo Grande 2006) and the data presented in this paper shows the importance of the exposition of the human population to *L. longipalpis*, first factor of risk for the occurrence of the disease.

We believe that the Fundação Nacional de Saúde has the necessity of a reevaluation of all measures used on vector control, as the eco-epidemiology of visceral leishmaniasis in Brazil is more complex than we thought (Lainson & Rangel 2005).

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