

SHORT COMMUNICATION

Recent Observations on the Sand Fly (Diptera: Psychodidae) Fauna of the State of Rondônia, Western Amazônia, Brazil: the Importance of *Psychodopygus davisii* as a Vector of Zoonotic Cutaneous Leishmaniasis

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Sand flies were collected in the central region of the state of Rondônia (W 64°30' to 63°00' and S 10°00' to 11°00') using Shannon and CDC light traps from October 1997 to August 2000. A total of 85,850 specimens representing 78 named species were captured. Of these 14 were new records for Rondônia. The proportion of males/females was 1/1.131. Trypanosomatids, that are presently being identified, were detected in 11 species. Leishmania (Viannia) naiffi was recorded from Psychodopygus davisii and P. hirsutus. In the present study the dominant species was P. davisii (39.6%) followed by Lutzomyia whitmani (13.1%), P. carrerai (11.6%), and P. hirsutus (10.2%). The importance of P. davisii as a vector of zoonotic cutaneous leishmaniasis is discussed.

Key words: sand fly - leishmaniasis - Psychodidae - flagellates - trypanosomatids - *Leishmania (Viannia) naiffi* - *Psychodopygus davisii* - Amazon - Rondônia - Brazil

There are published records of 78 sand fly species in Rondônia (Martins et al. 1965, 1978, Biancardi et al. 1982) and 88 species were listed as being present in the state by the Computer-aided Identification of Phlebotomine Sandflies of America (CIPA) group (<http://cipa.snv.jussieu.fr>) in 1999. The origin of the specimens referred to in these publications was from captures made during short periods at different times of the year. There are no details of the location within the state of the 10 species added to this list by the CIPA group. In order to obtain a more complete picture of the sand fly fauna of the central region of Rondônia, captures were made monthly over an 18-month period. The principal observations were made in the region of Cacaúlândia and occasionally in Monte Negro, Buritis, and Campo Novo. All are located in the central region of the state (W 64°30' and 63°00' and S 10°00' to 11°00'). During the eighties and nineties this region experienced an enormous influx of colonists that resulted in a drastic reduction of the original forest.

Around 30% of this forest was destroyed being replaced by small farms and townships. In spite of the human degradation of the environment patches of forest survived. The principal land usages are cattle ranching and coffee planting, although lumbering still continues to degrade the natural forest it is not as intense as it was.

Cutaneous leishmaniasis is one of the major public health problems of the state. Brazilian Ministry of Health data shows that the region is highly endemic with an annual prevalence in the order of 115 cases per 100,000 inhabitants which represents 5.5% of the total cases reported in 2000 in Brazil (Basano et al., pers. commun.). It is unknown how many *Leishmania* species are responsible for the human disease. In the study region it appears that at least 3 species, *Leishmania (Viannia) braziliensis*, *L. (V.) lainsoni*, and *L. (V.) guyanensis*, infect man (Shaw et al., unpublished results).

The region's climate is characterized by well marked dry and wet seasons. The mean average annual temperature is 28.6°C, with variations that range from 12°C to 37°C during the dry period. The rains occur principally between the months of November and April (total rainfall 1440 mm). During the rest of the year the climate is much drier (557 mm rainfall). The driest months are between May and August with an average monthly rain fall of 47 mm whereas in the wet season it is 263 mm.

Sand flies were captured during 21 visits to Cacaúlândia (W 62°17'53.81" and 10°7'96") and 1 each to Monte Negro (W 63°17'49.2" and S 10°15'43.4"), Buritis (W 63°41'99.7" and S 10°9'22.8"), and Campo Novo (W 63°30'62.5" and S 10°38'54.9") between October 1997 and August 2000.

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On the average captures were made for 5-6 days during each visit. Four CDC light traps were set between 18 h and 6 h: 2 were suspended at 1 m and 2 others at 6 to 7 m. The sand flies were separated from the other insects and put into vials containing 70% ethanol. In addition, sand flies were also caught in a Shannon trap between 18 h and 20 h. A 300 candle power spotlight was placed at the base of the central panel of the trap and any sand fly that settled on either the individuals in the trap or the surface of the trap were captured with an aspirator, killed and stored in 70% ethanol. They were prepared for identification according to the methods described by Young and Duncan (1994).

Eighty five thousand, eight hundred and fifty sand flies (45,564 females and 40,286 males) belonging to 79 different species were collected (Table I): *Brumptomyia* (6 species), *Lutzomyia* (61 species), and *Psychodopygus* (12 species); 14 species (759 males and females) were reported for the first time in Rondônia (Table I). Amongst the new records was *L. migonei*, associated with cutaneous leishmaniasis transmission in other regions of Brazil (Azevedo & Rangel 1991, Camargo-Neves et al. 2002) and *L. longipalpis*, the visceral leishmaniasis vector (Shaw 2002).

The following 15 species have been previously recorded from Rondônia but were not captured in the present study: *L. acanthopharynx*, *L. avellari*, *L. calcarata*, *L. caligata*, *L. cayennensis cayennensis*, *L. cerqueirai*, *L. dasymera*, *L. flabellata*, *L. flochi*, *L. inflata*, *L. loretonensis*, *L. marinkellei*, *L. octavioi*, *L. tuberculata*, and *P. chagasi*. All except *L. flochi*, *L. loretonensis*, *L. octavioi*, *L. tuberculata*, and *P. chagasi* were considered as being rare. Since our captures were made in one area over a long period their absence may be due to their

TABLE I

Sand flies captured in the state of Rondônia from October 1997 to August 2000 with Shannon and CDC light traps

Genus/Species	Female	Male	Total	First Report
<i>B. avellari</i>	0	39	39	No ^a
<i>B. cunhai</i>	0	2	2	Yes
<i>B. pentacantha</i>	0	1	1	No
<i>B. pinto</i>	1	6	7	No
<i>Brumptomyia</i> sp.	3	0	3	No
<i>B. travassosi</i>	40	81	121	Yes
<i>L. abunaensis</i>	0	1	1	No
<i>L. abonnenci</i>	0	7	7	No
<i>L. anduzei</i>	16	0	16	No
<i>L. antunesi</i>	666	540	1206	No
<i>L. aragaio</i>	20	10	30	No
<i>L. auraensis</i>	167	885	1052	No
<i>L. bacula</i>	103	31	134	No
<i>L. barretto</i>	3	29	32	Yes
<i>L. begoniae</i>	1	0	1	No
<i>L. brachipyga</i>	32	11	43	Yes
<i>L. brasiliensis</i>	0	7	7	No
<i>L. campbelli</i>	72	55	127	No
<i>L. carvalhoi</i>	15	0	15	Yes
<i>L. damascenoi</i>	18	1	19	No

<i>L. dasypodogeton</i>	1	0	1	No
<i>L. dendrophila</i>	254	336	590	No
<i>L. dreisbachi</i>	1	0	1	No
<i>L. evandroi</i>	2	0	2	Yes
<i>L. evangelistai</i>	92	65	157	No
<i>L. flaviscutellata</i>	280	186	466	No
<i>L. furcata</i>	83	89	172	No
<i>L. goiana</i>	17	0	17	No ^a
<i>L. gomezi</i>	317	101	418	No
<i>L. inpai</i>	1	1	2	Yes
<i>L. infraspinoso</i>	115	14	129	No
<i>L. longipalpis</i>	19	22	41	Yes
<i>L. longipennis</i>	11	0	11	Yes
<i>L. longispina</i>	1	2	3	No
<i>L. lutziana</i>	44	57	101	No
<i>L. micropyga</i>	23	21	44	No
<i>L. migonei</i>	151	95	246	Yes
<i>L. monstrosa</i>	3	0	3	No
<i>L. nevesi</i>	2604	792	3396	No
<i>L. oswaldoi</i>	0	1	1	Yes
<i>L. pacae</i>	1	0	1	No ^a
<i>L. peresi</i>	1	0	1	Yes
<i>L. pilosa</i>	5	1	6	No
<i>L. punctigeniculata</i>	1	1	2	No ^a
<i>L. richardwardi</i>	1	0	1	No
<i>L. rorotaensis</i>	33	1	34	No
<i>L. saulensis</i>	21	1	22	No
<i>L. scaffii</i>	0	0	0	No
<i>L. sericea</i>	2	0	2	No
<i>L. serrana</i>	99	88	187	No
<i>L. servulolimai</i>	16	40	56	No
<i>L. shannoni</i>	31	42	73	No
<i>L. shawi</i>	2438	674	3112	No ^a
<i>L. sordellii</i>	213	126	339	No
<i>L. spinosa</i>	0	1	1	No
<i>L. termitophila</i>	18	10	28	No
<i>L. triacantha</i>	0	1	1	No
<i>L. trichopyga</i>	1	0	1	No ^a
<i>L. trinidadensis</i>	551	401	952	No
<i>L. trispinosa</i>	29	47	76	No ^a
<i>L. ubiquitousalis</i>	541	617	1158	No
<i>L. umbratilis</i>	2	2	4	No
<i>L. waltoni</i>	8	0	8	No
<i>L. whitmani</i>	5579	5672	11,251	No
<i>L. williamsi</i>	1	0	1	Yes
<i>L. wilsoni</i>	5	5	10	No
<i>L. yuilli</i>	2	0	2	No
<i>P. amazonensis</i>	97	130	227	No
<i>P. ayrozai</i>	7	0	7	No
<i>P. bispinosus</i>	184	57	241	Yes
<i>P. carrerai</i>	4855	5070	9925	No
<i>P. clautrei</i>	755	457	1212	No
<i>P. complexus</i>	332	0	332	No
<i>P. corossoniensis</i>	459	2	461	No
<i>P. davis</i>	16,815	17,210	34,025	No
<i>P. geniculatus</i>	2069	1306	3375	No ^a
<i>P. hirsutus</i>	4677	4037	8714	No
<i>P. lainsoni</i>	473	127	600	No
<i>P. paraensis</i>	5	0	5	No
<i>P. complexus/wellcomei</i>	0	662	662	No
Not identified	62	10	72	b
	45,564	40,286	85,850	

B.: *Brumptomyia*; L.: *Lutzomyia*; P.: *Psychodopygus*; a: CIPA record - no locality available; b: includes 36 female and 4 males belonging to a *L. (Trichopygomyia)* sp.

specific ecological requirements rather than seasonal variation. Because there are no clear morphological distinctions between the females of *P. complexus* and *P. wellcomei* they are included together in Table I. However, since all the males were *P. complexus* we consider that it is very unlikely that any of the females were *P. wellcomei*. The greatest number of *P. complexus* males were taken between January and April which coincides with the wettest months. Except for *L. migonei* none of the species recorded by us for the first time in Rondônia were common, including *L. longipalpis*. This species was in fact rare and only captured between the months of August and November which is when the rains begin.

Four species: *P. davisii* (39.6%), *L. whitmani* (13.1%), *P. carrerai* (11.6%), and *P. hirsutus* (10.1%) accounted for 74.4% of the sand flies. All 4 species had previously been reported in Rondônia. There are published records of *L. (V.) braziliensis* in *P. davisii* and *P. carrerai* captured in Rondônia (Grimaldi et al. 1991) as well as in *L. whitmani* from the states of Bahia, Paraná, and Ceará (Ryan et al. 1990, Azevedo et al. 1990, Queiroz et al. 1994, Luz et al. 2000). Although only 12 of the 79 species belonged to the genus *Psychodopygus* they represented 69.34% of the total phlebotomines captured. This is typical of many regions of Amazônia, where this genus predominates (Ward et al. 1973). For instance in one area of primary forest in the state of Mato Grosso 71.3% of the sand flies belonged to the genus *Psychodopygus* (Azevedo et al. 2002). In an area of "terra firme" near Manaus, state of Amazonas, they represented only 20% of the phlebotomine fauna (Dias-Lima et al. 2002). In areas of secondary forest, "capoeira", on the borders of Amazônia, in the state of Maranhão, there are no *Psychodopygus* (Barros et al. 2000), but they do occur in small numbers in the denser forest further east in Ceará (Ready et al. 1983). The genus *Psychodopygus* seems to have diversified in Amazônia more than in any other region of the Americas. Some species do, however, occur in other Brazilian forests, such as the *arthuri* series which is limited to the Atlantic rainforest habitat, where it may occur in large numbers. *P. hirsutus*, *P. ayrozai*, and *P. geniculatus* also occur outside the Amazon region and have been found infected with *Leishmania* (Rangel et al. 1985). However, it is only in Amazônia that *Psychodopygus* species are proving to be important vectors of cutaneous leishmaniasis in both enzootic and zoonotic cycles.

The preference of the predominant 4 species according to the capture method (Table II) is as follows: the Shannon

trap was more efficient than CDC light traps ($\chi^2 = 40.7$ $p > .0000001$); *L. whitmani* showed a preference for CDC traps ($\chi^2 = 18.69$, $p = 0.000087$); *P. davisii* and *P. carrerai* showed a preference for the Shannon trap ($\chi^2 = 55.06$, $p = 0.0000001$ and $\chi^2 = 17.03$, $p = 0.0002$); *P. hirsutus* showed the same preference for ground level CDC light and Shannon traps. The greater numbers of *P. davisii* and *P. carrerai* caught in Shannon traps is possibly a reflection of their anthrophilic habits.

We might consider that *L. whitmani* is the most important species since it is already known to be an important vector of cutaneous leishmaniasis in other Brazilian states in both peridomestic and forest environments (Teodoro et al. 1998, Ryan et al. 1990, Campbell-Lendrum et al. 2000, Luz et al. 2000). However, in our study it was less common in Shannon traps than *P. davisii* and *P. carrerai* and appeared to have more arboreal than terrestrial habits. Such arboreal preferences have been reported for *L. whitmani* in Amazonas (Dias-Lima et al. 2002). In Mato Grosso it was more common at ground level at one capture site and in the canopy at another (Azevedo et al. 2002). This suggests that different ecological pressures may affect vertical distribution.

We are presently identifying some of the isolates found in 11 species: *L. rorotaensis*, *L. shannoni*, *L. dendrophila*, *L. nevesi*, *L. trinidadensis*, *L. sordelli*, *L. gomezi*, *L. whitmani*, *L. infraspinoso*, *P. davisii*, and *P. hirsutus*. These results are the subject of a future publication. In 1972, flagellates that were considered to be stages in the life cycle of a trypanosome, were recorded in specimens of *P. davisii* caught in the Utinga forest, Belém, state of Pará (Shaw & Lainson 1972). Three isolates from *P. davisii* collected in Rondônia were identified as *L. (V.) braziliensis* (Grimaldi et al. 1991). An undescribed *Leishmania* of the subgenus *L. (Viannia)* was found in a specimen of *P. hirsutus* captured in Além Paraíba, state of Minas Gerais (Rangel et al. 1985). A preliminary analysis, with a panel of monoclonal antibodies, indicates that the promastigotes isolated from *P. hirsutus* and *L. davisii* in the present study are *L. (V.) naiffi*. We conclude that present and previous results on the biology and infections of *P. davisii* strongly suggest that this species should be considered as a potential vector of zoonotic cutaneous leishmaniasis in the central region of Rondônia. Even more, it appears to be highly anthrophilic, outnumbering *L. whitmani* by 58.1 to 1 in Shannon traps. In Mato Grosso and Amazonas *P. davisii* was less often recorded and was more arboreal than terrestrial (Azevedo et al. 2002, Dias-Lima et al. 2002),

TABLE II
The relative abundance of the four prevalent sand fly species by capture method

Trap type/Species	Shannon Flies/hour	CDC Floor Flies/hour	CDC Canopy Flies/hour	Total Flies/hour
<i>L. whitmani</i>	1.0	3.8	16.2	7.2
<i>P. carrerai</i>	17.1	5.9	1.0	12.3
<i>P. davisii</i>	58.2	20.3	4.5	21.5
<i>P. hirsutus</i>	7.1	8.0	1.6	5.9
Total	83.4	38.0	23.4	25.4

L.: *Lutzomyia*; *P.*: *Psychodopygus*

which contrasts with the present findings. This species appears to be commoner in the more humid regions of Amazônia. For instance, in Mato Grosso it represented only 2.2% of the sand fly fauna (Azevedo et al. 2002) while in our catches it was 39.6% of the total. However, one must be careful when drawing conclusions on the relative abundance of different species. For instance in two capture sites that were only 22 km apart sand flies of the subgenera *L. (Nyssomyia)* represented 94.2% of the species captured and in the other 71.3% were *Psychodopygus* (Azevedo et al. 2002).

A total of 77.6% of the specimens were collected during the rainy season. The difference between this figure and that obtained in the dry season is highly significant ($\chi^2 = 13.94$, $p < 0.00001$), suggesting an increased risk of transmission to man during the wet season as in other regions (Condino et al. 1998, Barros et al. 2000, Marcondes et al. 2001, Salomon et al. 2002).

With the 14 species found in the present study, and those recorded by the CIPA group the total number of species known to occur in Rondônia now totalizes 102. Of these, 18 have been associated with *Leishmania* infections.

Finding *L. longipalpis* suggests that there is a risk, under favorable epidemiological conditions, that visceral leishmaniasis could become endemic in Rondônia. A combination of factors must determine whether or not this will occur. Perhaps the most important is the existence in the gene pool of individuals that are capable of adapting to the peridomestic and domestic habitats. Preventive measures, such as capturing stray dogs in the 43 new towns of Rondônia associated with epidemiological surveillance are desirable to monitor this possibility.

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