

Light trapping of the vector *Lutzomyia longipalpis* (Diptera: Psychodidae) using the SS trap in peridomestic urban areas endemic for visceral leishmaniasis in northeast Brazil

Captura do vetor *Lutzomyia longipalpis* (Diptera: Psychodidae) com armadilhas luminosas SS em áreas peridomiciliares urbanas endêmicas para a leishmaniose visceral na região nordeste do Brasil

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Abstract

Objective: to use the SS trap in sampling *Lutzomyia longipalpis* (Lutz & Neiva 1912) in urban peridomestic settings endemic for visceral leishmaniasis in Maranhão state, northeast Brazil. **Method:** two SS traps, one equipped with a 520 nm green LED and another with a 470 nm blue LED, and one CDC-type light trap, were used to sample sand flies. **Result:** a total of 1,143 sand flies were captured, and *L. longipalpis* was the most frequent species (83.2%). The second and third most frequent species were *Nyssomyia whitmani* (Antunes & Coutinho, 1939) (8.5%) and *Evandromyia sallesi* (Galvão & Coutinho, 1939) (2.1%), respectively. The two former species are proven vectors of visceral and cutaneous leishmaniasis, respectively, and have been frequently found in peridomestic areas with endemic disease. The third one was found for the first time in the northeastern part of the state and has already been found naturally infected with *L. (L.) infantum* in southeast Brazil. The SS trap equipped with 520-nm green LEDs attracted the most phlebotomine sand flies (14.94±2.5), followed by CDC (11.06±2.5) and SS blue light traps (9.71±1.3). **Conclusion:** without being statistically significant, these differences demonstrate that the SS trap is an efficient tool for collecting and monitoring *L. longipalpis* and other sand flies in urban peridomestic sites.

Keywords: light trapping; leishmaniasis; Silva suction trap; vector control.

Resumo

Objetivo: utilizar a armadilha Silva de sucção (SS) nas coletas do vetor *Lutzomyia longipalpis* (Lutz & Neiva 1912) em áreas peridomiciliares urbanas endêmicas para a leishmaniose visceral no estado do Maranhão, região nordeste do Brasil. **Método:** duas armadilhas SS, uma com LED de 520 nm (verde) e outra com LED de 470 nm (azul), e uma armadilha tipo CDC foram usadas nas coletas dos flebotomíneos. **Resultado:** um total de 1.143 flebotomíneos foi coletado, sendo *L. longipalpis* a espécie mais frequente (83,2%), seguida de *Nyssomyia whitmani* (Antunes & Coutinho, 1939) (8,5%) e *Evandromyia sallesi* (Galvão & Coutinho, 1939) (2,1%). As duas primeiras espécies são comprovadamente vetores da leishmaniose visceral e leishmaniose tegumentar, respectivamente, e são encontradas com frequência em ambientes peridomiciliares onde a doença é endêmica. A terceira espécie é registrada pela primeira vez nesta parte do estado e já foi encontrada naturalmente infectada pela *L. (L.) infantum* no sul do Brasil. A armadilha SS com o LED verde foi atrativa para a maioria dos flebotomíneos (14,94±2,5), seguida pela CDC (11,06±2,5) e SS com o LED azul (9,71±1,3). **Conclusão:** mesmo sem significância estatística, os resultados demonstram que a armadilha SS é uma ferramenta eficiente para coletar e monitoramento de *L. longipalpis* e de outras espécies de flebotomíneos em áreas peridomiciliares urbanas.

Palavras chave: captura com armadilha luminosa; leishmaniose; armadilha Silva de sucção; controle de vetor.

INTRODUCTION

Visceral leishmaniasis (VL), also known as kala-azar, is a neglected parasitic disease that occurs in several countries worldwide¹. The most important causative agents of VL are *Leishmania (Leishmania) donovani* and *L. (L.) infantum* (*syn. L. chagasi*). The former is responsible for severe forms in Southeast Asia (e.g., India, Bangladesh) and Eastern Africa (e.g., Ethiopia, Kenya), and the latter causes VL in the Mediterranean area, the Middle East, Afghanistan, Iran, Pakistan, and the

Americas². In the Americas, 90% of VL cases occur in Brazil, mainly in the North and Northeast regions, where the highest risk of VL transmission continues to occur³.

In Brazil, the primary vector of VL is the phlebotomine sand fly *Lutzomyia longipalpis* (Lutz & Neiva 1912), a species widely distributed in several Brazilian urban areas, especially in visceral leishmaniasis endemic foci⁴. This anthropophilic sand fly species

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is commonly found in peridomestic settings, where conditions such as irregular occupation, presence of domestic animals in high numbers near households, lack of basic sanitation, and poverty are met^{5,6}. In Maranhão state, northeastern Brazil, VL is endemic in several municipalities⁷, including the municipality of Chapadinha, where this study was conducted. According to data available on the Brazilian Notifiable Disease Information System (SINAN)⁸, from 2019 to 2022, 70 autochthonous human cases of VL were registered in rural and urban zones of Chapadinha.

The spread of *L. longipalpis* and the urbanization of VL in Brazil have been ongoing for a long time⁹, and, in an epidemiological context, monitoring of the insect vector has become paramount. The monitoring of sand flies has been achieved mainly with light traps, which provide relevant information on sand fly density and distribution, efficiently contributing to the detection of areas of higher risk and high priority for directing VL control programs. The most widely used light-trapping device for sand fly work has been the Centers for Disease Control and Prevention (CDC) light trap¹⁰, which uses light for attracting sand flies. Additionally, olfactory cues such as CO₂ and octenol and visual cues such as Light-Emitting Diode (LED)-based attractants have been applied to improve the efficiency of CDC light traps^{11,12}.

With the advantages of using LEDs as visual attractants to insects¹¹, and the continuous need for insect trapping improvement, besides some drawbacks of using CDC light traps¹³, a LED-baited suction trap was recently developed for collecting small night-flying insects¹⁴. This new light trap (named SS trap) has already been field validated for sampling phlebotomine sand flies in a very rural landscape, where it was proven to be a good alternative to the conventional CDC light trap¹⁴. Despite the proven effectiveness of the SS trap in sampling phlebotomine sand flies, its use in urban endemic areas for VL has not yet been evaluated. Therefore, the aim of this study was to investigate the effectiveness of the SS trap in the light trapping of *L. longipalpis* in endemic urban peridomestic areas for LV in Maranhão state, northeastern Brazil.

METHODS

Study site

This study was conducted in two peridomestic settings (Vila Isamara and Novo Castelo districts) of the municipality of Chapadinha, Maranhão state, Northeastern Brazil (Figure 1). This municipality is located 230 km away from the state capital (São Luis) and has a population of 81,386 inhabitants and an area of 3,247,385 km². The Brazilian savanna (Cerrado) is the predominant vegetation type in the region. Vila Isamara is a densely populated urbanized district with confirmed human and canine cases of VL. In the Vila Isamara district peridomestic area (area 1) (3.7559 S, 43.3626 W), large pigs were observed to be raised for sale in the backyard, and bad hygienic conditions were detected. Some other domestic animals, like dogs, chickens, and cats, were present over the study period. Fruit plants such as orange trees, mango trees, and banana trees were also present at the collecting site. The Novo Castelo

district peridomestic area (area 2) (3.729808 S, 43.3372 W) is a more wooded habitat and larger than area 1. Several fruit trees (lemon trees, jack trees, cherry trees, coconut trees, cashew trees, among others) and domestic animal pens (dogs, pigs, chickens) were present in the surroundings of the sampling site. Additionally, synanthropic animals (opossums) were also sporadically seen in area 2 during the study period.

Figure 1. Map of the study area.



Source: Joany da Costa Viana and Jefferson Mesquita Brito

According to the Köppen climate classification, the predominant climate type in the state is tropical with dry winter (Aw). The mean air temperature is between 27.1°C and 27.5°C, and the annual rainfall is between 1700 mm and 2000 mm. There are two well-defined periods: a dry period between July and December and a rainy period between January and June¹⁵. This research was performed in the dry period, between July and September.

Study design and statistical analysis

In this study, two Silva Suction (SS) traps and one CDC-type light trap were used to capture sand flies. The SS trap is a patent-pending tool developed for sampling small night-flying insects¹⁴. In the SS traps, two different wavelengths were used as light attractants, where one SS was equipped with a 520 nm green LED (15,000 millicandela) and the other SS trap was equipped with a 470 nm blue LED (15,000 millicandela). The 33-night study was undertaken between July and September 2024. At each location, all traps were deployed approximately 20 m from each other, at 1 m above the soil, from sunset to sunrise, and distributed following a 3x3 Latin square design with a total of 11 replications (5 replications for area 1 and 6 replications for area 2). In the laboratory, the captured specimens were processed according to standard procedures for taxonomic identification, following¹⁶. Data were analyzed using GraphPad Prism version 8.0.2 (GraphPad Software). Prior analysis, data were examined for normality using the Smirnov-Kolmogorov test. Non-Gaussian data were analyzed using Kruskal-Wallis or the Mann-Whitney U-test as appropriate. Some distributions that failed tests of normality were log₁₀-transformed into parametric analyses to standardize variances and meet the normality assumption.

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Statistical significance was considered when $p < 0.05$.

RESULTS

A total of 1,143 specimens belonging to 11 species were

collected. The *L. longipalpis* sand fly was the most frequent species (83.2%) in both areas, followed by *Nyssomyia whitmani* (8.5%) and *Evandromyia sallesi* (2.1%), which together accounted for 93.8% of all the specimens collected (Table 1).

Table 1. Species of sand flies captured with a new light trap in two endemic areas for visceral leishmaniasis in northeast Brazil.

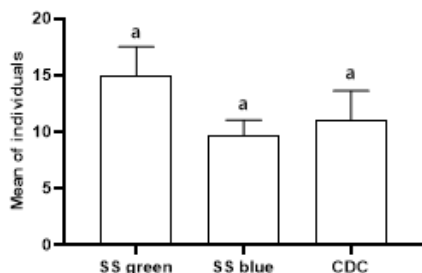
Species	Area				Light traps						Total
	1		2		SS green		SS blue		CDC		
	N	F:M	N	F:M	N	F:M	N	F:M	N	F:M	
<i>Lu. longipalpis</i> (Lutz & Neiva, 1912)	378	1:2.9	574	1:3.1	415	1:5.2	249	1:5.3	288	1:3.7	952
<i>Ny. whitmani</i> (Antunes & Coutinho, 1939)	7	1:1.3	91	1:12	22	1:22	46	1:6.6	30	1:9	98
<i>Ev. sallesi</i> (Galvão & Coutinho, 1939)	18	0:18	6	1:2	20	1:19	-	-	4	1:3	24
<i>Ev. evandroi</i> (Costa Lima & Antunes, 1936)	2	2:0	16	1:1.6	4	1:1	5	1:1.5	9	1:1.2	18
<i>Ev. lenti</i> (Mangabeira, 1928)	-	-	14	0:14	4	0:4	3	0:3	7	0:7	14
<i>Bi. flaviscutellata</i> (Mangabeira, 1942)	7	0:7	1	0:1	-	-	4	0:4	4	0:4	8
<i>Sc. sordellii</i> (Shannon & Del Ponte, 1927)	-	-	7	1:6	5	1:4	1	0:1	1	0:1	7
<i>Ps. wellcomei</i> (Fraiha, Shaw & Lainson, 1971)	7	1.3:1	-	-	3	1:2	2	1:1	2	1:1	7
<i>Mi. quinquefer</i> (Dyar, 1929)	3	3:0	4	4:0	-	-	-	-	7	7:0	7
<i>Mi. vilhelai</i> (Martins, Falcão & Silva, 1962)	1	0:1	5	1:1.5	3	0:3	2	2:0	1	0:1	6
<i>Ev. termitophila</i> (Martins, Falcão & Silva, 1964)	-	-	2	1:1	2	1:1	-	-	-	-	2
Total number of specimens	423	1:2.9	720	1:3.4	478	1:5.5	312	1:5.2	353	1:3.5	1,143
Total number of species	8		10		9		8		10		

Lu: *Lutzomyia*; Ny: *Nyssomyia*; Ev: *Evandromyia*; Bi: *Bichromomyia*; Sc: *Sciopemyia*; Ps: *Psychodopygus*; Mi: *Micropygomyia*; Area 1: Vila Isamara district; Area 2: Novo Castelo district; N: number of individuals per trap; F:M: sex ratio female:male; SS green: SS trap baited with green LEDs; SS blue: SS trap baited with blue LE

Although not statistically significant ($t = 0.22$, $p = 0.82$), *L. longipalpis* was found more frequently in area 2, the most wooded habitat (Table 1). Regarding the sex ratio, males of *L. longipalpis* (82.4%) predominated over females (female-male sex ratio of 1:4.7), and these differences were statistically significant (Mann-Whitney U = 138.5, $p < 0.0001$).

When considering the light trap attractiveness, the SS trap equipped with 520-green LEDs attracted the most phlebotomine sand flies (14.94 ± 2.5), followed by CDC (11.06 ± 2.5) and SS blue light traps (9.71 ± 1.3) (Table 1). However, statistically, no significant difference was found among the three light traps (Kruskal-Wallis = 3.545, $p = 0.1699$) (Figure 2).

Figure 2. Mean catches (\pm SEM) of phlebotomine sand flies captured by silica suction (SS) equipped with green and blue LEDs and Centers for Disease Control and Prevention (CDC) light traps in northeastern Brazil.



Source: The same letters indicate no significant difference (Kruskal-Wallis, $P < 0.05$).

DISCUSSION

Lutzomyia longipalpis and *Nyssomyia whitmani* have been frequently found in peridomestic settings in Maranhão state^{17,12}, where conditions such as the presence of domestic animals, fruit trees, and organic matter accumulation are environmentally suitable for their occurrence. In the Chapadinha municipality, where VL endemicity is high, human cases of the cutaneous leishmaniasis (CL) form of the disease have also been reported (SINAN)⁸. In fact, in Brazil, *L. longipalpis* and *N. whitmani* are proven vectors of visceral and cutaneous leishmaniasis, respectively¹⁸, and a concomitant circulation of the causative agents of both leishmanial forms should be occurring in the study region, similarly to what is observed in the northern¹⁹, and eastern parts of the state.

The presence of *E. sallesi* in this study represents the first record of this species for northeastern Maranhão, being scarcely found in other climatically similar regions in this state²⁰. In Brazil, *E. sallesi* has a restricted distribution²¹ and is generally found in VL transmission areas in which *L. longipalpis* is the predominant insect vector^{22,23}. The present finding is quite relevant since this species has already been found naturally infected with *L. (L.) infantum* in the Brazilian state of Minas Gerais²⁴ and harboring *L. braziliensis* complex DNA in a cave environment²⁵, suggesting a possible involvement of this sand fly species in leishmaniasis transmission cycles, but further investigation is needed.

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Lutzomyia longipalpis has successively been established in urban areas²⁶, where environmental features such as low socio-economic level, bad sanitation conditions, abundant vegetation, organic matter, and domestic animals give support to the occurrence of high densities of this species⁵. All the above-mentioned environmental factors were observed in both areas of the present study.

The occurrence of *L. longipalpis* in the most wooded habitat is possibly because abundant vegetation, as well as a high number of animal pens (pigs, chickens) found in that area, provide more suitable feeding and breeding conditions for sand flies, especially in hot and dry months, as in the period in which this study was conducted. This result was the same for *N. whitmani*, a sand fly species more associated with chicken sheds and peridomestic settings, mostly in proximity to forested habitats²⁷.

A high proportion of males of *L. longipalpis* has also been found in other endemic Brazilian areas^{5,28}, and this may be attributed to a variety of interconnected factors, including light trapping, high host availability, lek-like behavior, and nearby breeding sites. Sand fly males search actively for hosts for mating. Near or on them, males aggregate and release sex pheromones to attract females as well as other males²⁹. As females are generally seeking blood meals and rest after feeding, males exhibit a more active flight behavior. It may explain the high capture frequency of sand fly males in light traps, mainly in areas of high host availability, creating a male-biased sex ratio.

The results concerning LED-baited light trap corroborate previous findings that the 520-green LED light is an efficient light attractant for phlebotomine sand flies, although without

statistical significance when compared with other light sources^{11,30,14}. In the current study, despite the non-significant results, the SS traps have proven to be operationally and economically efficient in improving catches of phlebotomine sand flies in endemic foci of leishmaniasis. The use of LEDs, small and rechargeable batteries, and a low-speed fan (i.e., low-power consumption), as well as a lightweight and non-assembled configuration, are some of the advantageous qualities that SS traps present over the standard CDC light trap¹⁴.

CONCLUSION

In conclusion, this research highlights the need for continuous light-trapping monitoring of sand flies in urban peridomestic areas. This approach provides insights into VL epidemiology and helps to detect areas with high densities of leishmaniasis vectors and reservoirs, which pose a threat to human health and the environment. Furthermore, regular monitoring in urban areas is important to find new potential leishmaniasis vector species and identify priority areas for public health intervention targets. In this context, the SS trap has been proven to be an efficient and selective (i.e., small flying insects) tool for collecting and monitoring *L. longipalpis* and other sand flies in urban peridomestic sites. This novel-monitoring light trap is expected to be available commercially soon.

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