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Resarch Article

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Distribution of Vector Sandflies Leishmaniasis from an Endemic Area of Venezuela

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Abstract

Background: Sandflies distribution is associated with different factors linked to climate changes that might cause alterations in their distribution and increase the risk of leishmaniasis transmission.

Present work aims to determine the composition and structure of sandflies from an endemic area of Venezuela and its relationship to environmental variables.

Method: Various locations were sampled, four sandflies capture methods were used and weather variables capture site, altitude, temperature and relative humidity were recorded. Specimens were identified. Abundance, dominance and species richness was estimated and multivariate analysis was performed.

Results: *L. youngi* is the main species associated with sandfly transmission of Leishmania in the highlands (≥ 600 m.asl), while *L. gomezi*, *L. ovallesi* and *L. walkeri* were found in lower altitudes and higher temperatures, prevailed in the lowlands (≤ 600 m. asl). Sandflies in the warmer lower altitudes showed greater species richness, greater biodiversity, and lower dominance than those at higher altitudes.

Conclusion: The sandflies composition and structure changed according to climate factors, showing a species-specific dispersion pattern. Relevant data for sandfly vectors species of Leishmania are provided that should be considered in implementing control and prevention measures.

Keywords: Leishmania; Lutzomyia; Climate; Disease transmission

Introduction

The appearance of leishmaniasis cases has been correlated to the presence of Leishmania-transmitting sandflies. Various epidemiologic leishmaniasis transmission patterns are caused by ecological and environmental conditions that lead to sandflies population dynamics [1-3]. Sandflies life cycles are affected by climate variations due to natural processes, especially weather conditions (e.g., precipitation, temperature, humidity, etc.) [4]. Agricultural development and changes in climate conditions could favour new habitats for vector insects [5-8]. Increases in sandflies dispersion may lead to higher human-vector contact [9-13] and, therefore, higher risk of leishmaniasis transmission [3,14]. Zorilla et al. correlated the presence of cutaneous leishmaniasis to environmental and socioeconomic variables in population of Yaucano Valley in Perú, the adaptability of sandflies to the human environment may lead to an increase in the number of leishmaniasis cases [15]. However, studies in dry climate regions have presented controversial results, such as a lack of correlation between climate variables and sandflies density [16,17]. Temperature increases may also affect sandflies populations [18-20]. Rodríguez et al. suggested that changes in ecological and climate conditions in Mérida, Venezuela from events such as flash flooding along stream beds at Mocotíes Valley may have affected the occurrence of leishmaniasis cases [7]. Thus, it is necessary to characterize the conditions that affect Leishmania-transmitting sandflies populations [4]. The goal of this study was to characterize the composition and structure of sandflies at endemic area of Mocotíes Valley in Mérida, Venezuela, and to determine the relationship between these characteristics with environmental variables.

Materials and Methods

Study area

The selected area for this study was Zea Municipality State Mérida, Venezuela located in the southeastern quadrant of the State (latitude 8°20'20" to 8°33'00", longitude 71°42'10" to 71°49'20"), the transition zone between Mocoties Valley and the southern edge of Lake Maracaibo, located between the mountains and Escalante and Guaruríes rivers. An area of 135 km^2 at 600 m above sea level (ASL) has lower mountain tropical rainforest vegetation. The annual average temperature is 22°C, with annual rainfall of 1390 mm and comprises two Parishes, El Caño Tigre and Zea.

Environmental variables

The altitude of each sampled location was recorded with an altimeter. Measurements were grouped into two altitude ranges: below 600 m ASL, corresponding to Caño El Tigre Parish, and above 600 m ASL, corresponding to Zea Parish. Temperature and relative humidity (RH) were measured with a digital thermo-hygrometer. Four measurements were measured made during a 1-hour period between 19:00 and 20:00. Two average temperature ranges, below 25°C (19-25°C) and above 26°C (26-33°C), and three RH ranges, below 60%, between 60% and 80%, and above 80% were defined.

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Sandflies capture and dissection

To catch sandflies four methods were performed, Shannon trap, light trap attractant (CDC), oiled Trap and capture oral direct aspiration [8]. The specimens under a stereoscopic microscope were dissected. Quick identification on fresh species by comparative morphology of the females was performed [21]. Under optical phase contrast microscopy the extracted digestive systems were observed to determine the presence of Leishmania promastigotes. Parasite development pattern in the intestine for identification of the subgenus was determined [22]. The body segments, head and terminalia of sandflies dissected clarified in Nesbitt and mounted in Berlese for identification [21]. Males captured were processed similarly without removal of the digestive system.

Statistical Analysis

The dominance, abundance and biodiversity Margalef index were determined [23]. Analysis of variance with a confidence level of Sig. = 0.005 was performed. A simple correlation analysis and Pearson's correlation coefficient Sig = 0.005 were calculated, using the IBM SPSS Statistics, available for download free software http://ibm-spss-statistics.softonic.com

Results

In Zea Municipality, 21 species of Lutzomyia from a total of 963 captured sandflies were identified, 82.7% female and 17.3% male. The most abundant species were *L. youngi* (38.42%), *L. gomezi* (28.87%), *L. ovallesi* (18.07%), *L. walkeri* (3.2%), and *L. spinicrassa* (3.1%). Other species were present at levels below 1.9%. The results showed dominance and biodiversity values for Zea Municipality of λ=0.27 and I=3.05, respectively, Table 1. Natural Leishmania infection was present in 17 of the captured sandflies, a natural infection rate of 2%.

In terms of environmental variables, 56.28% of *Lutzomyia* were present at temperatures below 26°C, predominantly *L. youngi* (37.49%), *L. gomezi* (23.36%) and *L. ovallesi* (12.77%) were prevalent above 26°C, Table 2.

Four species were identified below 60% RH, with *L. gomezi* (0.8%) being most abundant. At 60% to 80% RH, 19 species, predominantly *L. gomezi* (17.7%), *L. youngi* (14%), and *L. ovallesi* (14%) were identified. Above 80% RH, 16 species, predominantly *L. youngi* (24.1%) and *L. gomezi* (10.4%) were identified, Table 3.

Altitudes 600m ASL, corresponding to El Caño Tigre Parish, 16 species were identified, predominantly *L. gomezi* (51%) and *L. ovallesi* (31.6%). In Zea Parish, 12 species were identified, with *L. youngi* (87%) being the most abundant. Only five species occurred in both municipalities and altitude levels, Table 4. An ANOVA test comparison between altitudes revealed significant differences (F=1121; GL= 22; Sig=0.000).

Figure 1 show values for biodiversity, dominance, and species richness for sandflies populations in El Caño Tigre Parish (<600 m ASL) and Zea Parish (>600 m ASL). Sandflies in the warmer lower altitudes showed greater species richness, greater biodiversity, and lower dominance than those at higher altitudes.

Linear correlation analysis showed that altitude was directly proportional to *L. youngi* (p= + 0.765) and inversely proportional to *L. gomezi* (p=-0.494); temperature was inversely proportional to *L. spinicrassa* (p= -0.598), Table 5.

The primary sandflies species fell into two population groups: *L. youngi* and *L. spinicrassa* were distributed across higher altitudes and lower temperatures, whereas *L. gomezi*, *L. ovallesi*, and *L. walkeri* were found in lower altitudes and higher temperatures. RH did not show precise distribution patterns, Figure 2.

Species	♀	%	♂	%	Nat Inf.	%	N	%	pi	(λ)
*L. youngi**	370	46	0	0	7	1	370	38.42	0.38	0.15
*L. gomezi**	209	26	69	41	5	1	278	28.87	0.29	0.08
*L. ovallesi**	152	19	22	13	4	0.42	174	18.07	0.18	0.03
*L. walkeri**	9	1	22	13	1	0.1	31	3.22	0.03	0
*L. spinicrassa**	2	0	28	17	0	0	30	3.12	0.03	0
*L. trinidadensi***	9	1	10	6	0	0	19	1.97	0.02	0
*L. panamensis**	11	1	0	0	0	0	11	1.14	0.01	0
*L. hernandezi***	6	1	2	1	0	0	8	0.83	0.01	0
*L. nuneztovari**	7	1	0	0	0	0	7	0.73	0.01	0
L. venezulensis	5	1	3	2	0	0	7	0.73	0.01	0
*L.atroclavata**	1	0	4	2	0	0	5	0.52	0.01	0
*L. migonei**	5	1	0	0	0	0	5	0.52	0.01	0
L. shanoni	2	0	2	1	0	0	4	0.42	0	0
L. puntigeniculata	0	0	2	1	0	0	2	0.21	0	0
L. serrana	1	0	1	1	0	0	2	0.21	0	0
L. dubitans	0	0	2	1	0	0	2	0.21	0	0
*L. lichyi**	2	0	0	0	0	0	2	0.21	0	0
L. cayennensis	1	0	0	0	0	0	1	0.1	0	0
L. olmeca nociva	1	0	0	0	0	0	1	0.1	0	0
L. pilosa	1	0	0	0	0	0	1	0.1	0	0
no_identificada	3	0	0	0	0	0	3	0.31	0	0
Total species: 21	**797**	**100**	**167**	**100**	**17**	**2**	**963**	**100**	**1**	**0.7**

Female (♀) Male (♂) Natural infection (Nat inf.) Relative abundance of species (pi) Simpson index (λ) *Anthropophilic species **Zoophilic species

Table 1: Sandflies species identified at Zea Municipality Merida State.

Species	19-25°C		26-33°C	
	N	%	N	%
L. youngi	361	66.6	9	2.1
L. gomezi	53	9.7	225	53.4
L. ovallesi	51	9.4	123	29.2
L. walkery	8	1.4	23	5.4
L. spinicrassa	30	5.5	-	-
L. trinidadensi	8	1.4	11	2.6
L. panamensis	1	0.1	10	2.3
L. hernandezi	2	0.3	6	1.4
L. nuneztovari	7	1.2	-	-
L. venezuelensis	6	1.1	2	0.4
L.atroclavata	2	0.3	3	0.7
L. migonei	2	0.3	2	0.4
L. shanoni	1	0.1	3	0.7
L. puntigeniculata	1	0.1	1	0.2
L. serrana	2	0.3	-	0
L. dubitans	1	0.1	1	0.2
L. lichyi	2	0.3	-	-
L. cayennensis	-	-	1	0.2
L. olmeca nociva	1	0.1	-	-
L. pilosa	-	-	1	0.24
no_identificada	3	0.5	-	-
Total species: 21	542	100	421	100

Table 2: Distribution of sand fly species identified at Zea Municipality in relation to temperature ranges.

Species	< 60%		60-80%		80 % >	
	N	%	N	%	N	%
L. youngi	-	-	138	26.7	232	53.3
L. gomezi	8	66.7	170	32.9	100	23
L. walkery	0	0	28	5.4	3	0.7
L. ovallesi	1	8.3	137	26.5	36	8.3
L. spinicrassa	-	-	4	0.8	26	6
L. atroclavata	-	-	2	0.4	3	0.7
L. cayennensis	-	-	-	-	1	0.2
L. dubitans	-	-	1	0.2	1	0.2
L. hernandezi	-	-	5	1	3	0.7
L. lichyi	-	-	1	0.2	1	0.2
L. migonei	-	-	2	0.4	3	0.7
L.nuneztovari	-	-	-	-	7	1.6
L. olmeca nociva	-	-	1	0.2	-	-
L. panamensis	-	-	7	1.4	4	0.9
L. pilosa	-	-	1	0.2	-	-
L. puntigeniculata	-	-	2	0.4	-	-
L. serrana	-	-	2	0.4	-	-
L. shanoni	-	-	2	0.4	2	0.5
L. trinidadensi	2	16.7	9	1.7	8	1.8
L. venezuelensis	1	8.3	2	0.4	5	1.1
no_identificada	-	-	3	0.6	-	-
Total species	12	100	514	100	435	100

Table 3: Distribution of sand fly species identified at Zea Municipality in relative humidity ranges.

Discussion

Topographic relief is an important factor in climate differences, especially in intertropical zones where there are different climate plateaus depending on altitude [17, 24]. The results confirmed that sandflies fauna fell into two populations, with differences in composition and structure, between the two Parish that divided geopolitically the Municipality, perhaps motivated to that these two areas are ecologically different. Knowledge of the distribution and other behavioural aspects of the sandfly species that occur in this region are of great importance for the entomology and biodiversity. A diversified and

Species	Zea Parish Altitudinal High (≥ 600 m asl)						Caño El Tigre Parish Flat altitudinal low (≤ 600 m asl)					
	n	%	Nat Inf.	%	pi	(λ)	n	%	Nat Inf.	%	pi	(λ)
L. youngi	367	87.17	7	2	0.87	0.76	3	0.55	-	-	0.01	0
L. spinicrassa	29	6.9	-	-	0.07	0	1	0.18	-	-	0	0
L. nuneztovari	7	1.66	-	-	0.02	0	-	-	-	-	-	-
L. migonei	3	0.71	-	-	0.01	0	-	-	-	-	-	-
L. ovallesi	2	0.48	-	-	0	0	172	31.68	4	0.74	0.32	0.1
L. serrana	2	0.48	-	-	0	0	-	-	-	-	-	-
L. venezulensis	2	0.48	-	-	0	0	6	1.1	-	-	0.01	0
L. dubitans	2	0.48	-	-	0	0	-	-	-	-	-	-
L. lichyi	2	0.48	-	-	0	0	-	-	-	-	-	-
L. atroclavata	1	0.24	-	-	0	0	4	0.74	-	-	0.01	0
L. gomezi	1	0.24	-	-	0	0	277	51.01	5	0.92	0.51	0.26
L. walkery	-	-	-	-	-	-	31	5.71	1	0.18	0.06	0
L. trinidadensi	-	-	-	-	-	-	19	3.5	-	-	0.03	0
L. panamensis	-	-	-	-	-	-	11	2.03	-	-	0.02	0
L. hernandezi	-	-	-	-	-	-	8	1.47	-	-	0.01	0
L. shanoni	-	-	-	-	-	-	4	0.74	-	-	0.01	0
L. migonei	-	-	-	-	-	-	2	0.37	-	-	0	0
L. puntigeniculata	-	-	-	-	-	-	2	0.37	-	-	0	0
L. cayennensis	-	-	-	-	-	-	1	0.18	-	-	0	0
L. olmeca nociva	-	-	-	-	-	-	1	0.18	-	-	0	0
L. pilosa	-	-	-	-	-	-	1	0.18	-	-	0	0
no_identificada	3	0.71	-	-	0.01	0	-	-	-	-	-	-
Total	421	100	7	2	1	0.77	543	100	10	1.84	1	0.37
Natural Infection (Nat Inf.) Relative abundance Specie (pi) Simpson Index (λ)												

Table 4: Composition of sandflies recorded in El Caño Tigre Parish Flat altitudinal low (≤ 600 m asl) and Zea Parish Altitudinal High (≥ 600 m asl).

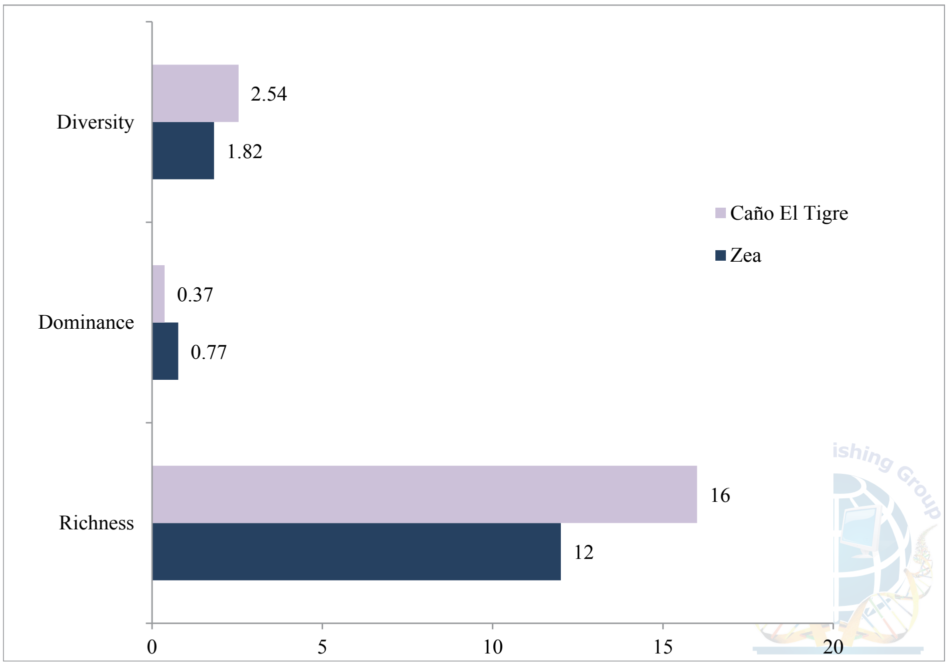

Figure 1: Distribution of main species of sandflies in Zea Municipality in relation to climatic parameters.

Species		Altitude	Temperature Pearson Correlation Sig. (bilateral)	Relative Humidity
L. youngi		0.765	-0.674	0.293
		0	0.038	0.138
L. gomezi		-0.494	0.309	-0.046
		0	0.245	0.821
L. spinicrassa		0.591	-0.598	-0.401
		0	0	0.038

Table 5: Simple linear correlation between key species and environmental variables studied.

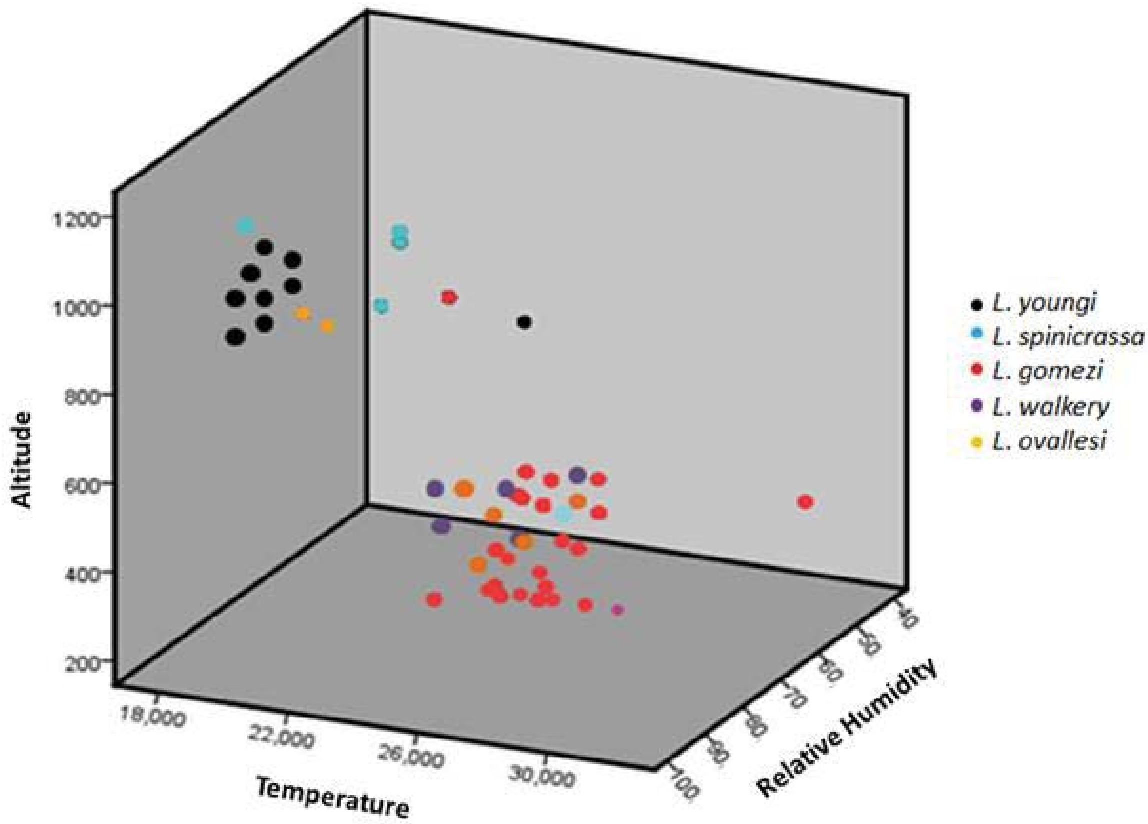

Figure 2: Composition and community structure of sandflies identified in Zea and El Caño Tigre Parish.

widely distributed sandfly fauna represents a significant transmission risk of leishmaniasis.

In Zea Parish (high zone, ≥ 600 m ASL), *L. youngi, L. spinicrassa*, and *L. nuñeztovari* were most abundant, whereas *L. gomezi, L. ovallesi, L. walkeri*, and *L. trinidadensis* predominated in El Caño Tigre Parish (low zone, ≤600 m ASL). Only five species were found at both altitude levels and parish. Our analysis showed correlation directly proportional of *L. youngi* and *L. spinicrassa* with high altitude and low temperature, whereas *L. gomezi* one correlation inversely proportional to low altitude and warm temperature.

L. youngi is considered the primary *Leishmania* transmission species in Mérida State [25], although *L. gomezi* predominates at 300m ASL [26]. Feliciangeli [27] measured low abundance of *L. gomezi* in the State of Carabobo at 85 m ASL. In Trujillo and Táchira States, *L. gomezi* is the most prevalent species at high altitudes (>1,000 m ASL) [28, 29]. Cazorla found this species as the main *Leishmania* vector at higher altitudes in Falcón State [30]. *L. gomezi* can distribute itself across a large altitude range [21]. In Venezuela, *L. gomezi* is naturally infected with *Le. braziliensis* promastigotes [31]. It is considered an alternate vector for tegumentary leishmaniasis in the north-central and other regions of Venezuela [6]. This study, *L. gomezi* with natural Leishmania infection below 600 m ASL were found; thus, we must consider *L. gomezi* an important species in Leishmania transmission in the low zones of Mérida State.

L. walkeri, the third species in order of abundance in the low zones, has a great capacity for adaptation to biotopes found at low altitudes

[27,32].The presence of this species in Venezuela has not been well-documented, and its role as a vector for Leishmania is still under discussion [6]. However, in our study we identified a specimen naturally infected with promastigotes that exhibited biological and morphological characteristics similar to Leishmania. The Zipayare region at Zulia State, a high density of *L. walkeri* was reported as the dominant species over *L. gomezi*. *L. walkeri*, is characterized as being able to exploit new biotic resources, with a fluctuating population dynamic at RH levels of 70%-90%, temperatures of 28-32°C, and altitude of 150 m ASL [33].

L. ovallesi was detected at both high and low altitudes, being more abundant at high altitudes; similar results were reported by a number of researchers [6,28,34]. *L. ovallesi* is considered to be an important vector of *Le. braziliensis*, *Le. mexicana*, and *Le. guyanensis* in Venezuela [6,31,35] and *Le. braziliensis* in Guatemala [36]. These findings suggest that *L. ovallesi* transmits Leishmania to humans in endemic zones at low and medium altitudes in Mérida State.

Our results demonstrate that *L. youngi, L. gomezi, L. ovallesi*, and *L. walkeri* are the most abundant Leishmania-transmitting species at Zea Municipality. The distribution and structure of sandflies fauna changed with climate factors, showing a species-specific dispersion pattern in the area. These factors must be considered when implementing prevention and control measures in at cutaneous leishmaniasis.

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