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To cite this version:

HAL Id: hal-03349719
https://hal.univ-guyane.fr/hal-03349719
Submitted on 21 Sep 2021

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Spatial variations in Leishmaniasis: A biogeographic approach to mapping the distribution of *Leishmania* species

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ARTICLE INFO

Keywords:
Biogeography
Cutaneous leishmaniasis
Stacked species distribution modelling
Deforestation
Zoonosis

ABSTRACT

Cutaneous Leishmaniasis (CL) is the most prevalent form of Leishmaniasis and is widely endemic in the Americas. Several species of *Leishmania* are responsible for CL, a severely neglected tropical disease and the treatment of CL vary according to the different species of *Leishmania*. We proposed to map the distribution of the *Leishmania* species reported in French Guiana (FG) using a biogeographic approach based on environmental predictors. We also measured species endemism i.e., the uniqueness of species to a defined geographic location. Our results show that the distribution patterns varied between *Leishmania* spp. and were spatially dependent on climatic covariates. The species distribution modelling of the eco-epidemiological spatial patterns of *Leishmania* spp. is the first to measure endemism based on bioclimatic factors in FG. The study also emphasizes the impact of tree cover loss and climate on the increasing distribution of *L. (Viannia) braziliensis* in the most anthropized regions. Detection of high-risk regions for the different between *Leishmania* spp. is essential for monitoring and active surveillance of the vector. As climate plays a major role in the spatial distribution of the vector and reservoir and the survival of the pathogen, climatic covariates should be included in the analysis and mapping of vector-borne diseases. This study underscores the significance of local land management and the urgency of considering the impact of climate change in the development of vector-borne disease management strategies at the global scale.

1. Introduction

With the global attention on novel emerging infectious diseases, research on ‘older’ diseases of high burden in the tropics remains limited. Due to a lack of global priority setting, these tropical diseases are often left neglected. In 2014, the WHO ranked Leishmaniasis as a group 1 emerging and uncontrolled disease Neglected Tropical Disease (NTD) [1]. Cutaneous Leishmaniasis (CL), the most prevalent form of the disease, is widely distributed from the Indian subcontinent, across West Asia and the Middle East, through the Mediterranean and Northern Africa to Central and South America. CL is a dermal infection caused by protozoan parasites belonging to the genus *Leishmania* (family Trypanosomatidae) and is transmitted by infected female sandflies. Worldwide, an estimated 600,000 to 1,200,000 cases of CL occur annually [2].

Cutaneous Leishmaniasis can be etiologically divided into Anthropoctic (old world) CL endemic to Asia and the Mediterranean caused by *L. tropica* and Zoonotic (new world) CL limited to the Americas. It is caused by two subgenera of *Leishmania*: *Leishmania Leishmania* complex and *Leishmania Viannia* complex. Species of the Viannia subgenus may systematically disseminate in the skin and lead to clinical complications such as mucosal leishmaniasis (ML). This is particularly associated with *L. (V.) braziliensis*, where ML can develop in 2–5% of patients with CL, days to years after recovery [3]. ML is characterized by the destruction of the mucous membranes of the nose, mouth and throat and can lead to long-term structural scarring and secondary bacterial infections [4].

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https://doi.org/10.1016/j.onehlt.2021.100307
Received 23 September 2020; Received in revised form 9 August 2021; Accepted 10 August 2021
Available online 12 August 2021
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Systemic anti-leishmanial drugs are used to treat CL caused by *Leishmania Viannia*, to promote healing of the primary lesion and reduce the risk of developing ML [5,6].

CL was described in French Guiana (FG) for the first time in 1954 [7]. With over 95% of the country attributed to primary rainforests, FG hosts a high diversity of mammals that can act as reservoirs for *Leishmania* spp. Five human *Leishmania* species have been described in FG: 86.9% *L. (V.) guyanensis*, 9.7% *L. (V.) braziliensis*, 2.8% *L. (L.) amazonensis*, 1.3% *L. (V.) lainsoni* and sporadic cases *L. (V.) naiffi* [8]. Apart from the species-dependent clinical manifestations, a main challenge for public health care is that the various *Leishmania* spp. differ in terms of treatment sensitivities. In non-endemic regions where experience with management of leishmaniasis is limited, this may result in delay of diagnosis and treatment failure as often observed in people travelling from endemic regions [9,10]. Therefore, characterizing the distribution of the five *Leishmania* spp. across FG could prove to be significant to assess the endemcity of the *Leishmania* species and aid in the management of clinical cases.

CL is a vector-borne zoonotic disease transmitted by sandflies of *Lutzomyia* genus in the New World. The lifecycle of *Leishmania* is dependent on transmission between female sandflies and animal reservoirs; humans being accidental hosts. As with most vector-borne diseases, seasonal variations of the number of cases along with recent trends of the distribution of the cases hint at the role of environmental factors in the transmission WHO lists Leishmaniasis as a climate-sensitive disease as changes in temperature, rainfall and humidity can alter the spatial distribution and survival of vectors and reservoir hosts [11]. Furthermore, changes in temperature may affect the development of *Leishmania* promastigotes in sandflies, expanding the transmission in areas that were not previously endemic. Using a biogeographic approach, would highlight the significance of environmental variables in the distribution of the *Leishmania* spp. in FG where such environmental mapping based on the environmental variables is currently lacking.

Here, we mapped the distribution of the five *Leishmania* spp. in FG using a biogeographic approach with environmental predictors. In addition to mapping *Leishmania* species distribution and richness, we also measured species endemism i.e., the uniqueness of the species to a defined geographic location. To measure the impact of deforestation on the different *Leishmania* spp., we compared the tree cover loss around the occurrence sites with pseudo-absence sites.

2. Methods

2.1. Ethics statement

All patients were informed using written documents that case records and biological data might be further used in research and that they had the right to refuse. The monocentric audit of retrospective anonymized case record data was approved by the National Commission for Informatics and Liberties (CNIL; number 1805118v0), France and the by the Ethical Committee at Andree-Rosemann General Hospital, Cayenne, FG.

2.2. Study area

FG is a French Overseas territory located at 3.9339° N, 53.1258° W in South America (Fig. 1). This tropical territory is split into the littoral region, that consists of a thin coastal strip at the north of the territory and is more urbanized, and the primary rainforests, where there are sparse settlements and sporadic illegal gold mining camps. Over 95% of the total land area of FG is classified as primary rainforest contributing to the highly biodiverse Guiana shield, which is in danger of anthropization to meet the demand of the increasing population. The land cover also includes distinct areas of savannahs, wetlands and coastal
mangroves.

2.3. Patient data

We used for our analysis, the records of patients in consultation for a suspicion of leishmaniasis at the Cayenne General Hospital and associated administratively health centers between January 1994 and January 2015. New cases were defined as cases without a history of leishmaniasis in the previous 12 months. Diagnosis was confirmed by the presence of *Leishmania* parasites on microscopic examination of May-Grünwald Giemsa-colored skin smears, and/or by a positive culture on RPMI culture medium, and/or by the detection of *Leishmania* DNA by PCR. Species identification was done by PCR-RFLP retrospectively for the cases after 2006 [8]. For the identification of *Leishmania* before 2006, the samples were sent to the national reference center for leishmaniosis in Montpellier and the identification was made according to the official method by isoenzymes. Not all cases could be identified due to insufficient *Leishmania* DNA quantity.

2.4. Location of exposure

The patient database of 470 cases contained the details on the location of potential exposure including the distance from the road or river in cases of forest chalets or gold mining camps. The sites of exposure were georeferenced using IGN (Institut National de l’Information Géographique et Forestière/ National Institute of Geography and Forestry) maps and projected at WGS84 and this projection was maintained throughout the study. The georeferenced exposures were subject to thinning to remove sampling bias.

2.5. Environmental variables

We extracted the climatic covariates at 2.5 min spatial resolution from Worldclim database [12]. All bioclim variables from one to nineteen were used in the analysis (Table 1). The topological data was extracted from one arc-second digital elevation model (DEM) of 30 m resolution, derived from NASA (Version 3.0) Shuttle Radar Topography Mission (SRTM) imagery available in the United States Geological Survey (USGS). The raster layers were then resampled to a fixed resolution of approximately 4 km² and stacked to a raster stack.

2.6. Fitting of model and Model prediction

The individual species distribution models (SDMs) and stacked species distribution models (SSDMs) were fitted using the R package “SSDM”. In our study, we used Maximum entropy model (Maxent) for the construction of the SDM. Maxent is a popular choice for SDM models using presence-only occurrences -only data. Finally, we built SSDMs using the same algorithm and the five *Leishmania* species. The outputs of the different species are aggregated in SSDM maps of local species richness, and a weighted endemism index (WEI) was used for the endemism map. All models were evaluated using ROC curves and the area under curves (AUC) for the produced thresholds were calculated.

2.7. Tree-cover loss analysis

To analyze the impact of deforestation on the distribution of the *Leishmania* species, we conducted a detailed spatiotemporal analysis using the R package “gfcanalytics” on the Hansan’s tree cover loss maps. We used the presence and generated pseudo-absence points at a spatial buffer of 15 km, for this analysis. Regression analysis was done for each *Leishmania* sp. to establish the relationship between deforestation and the presence of the pathogen.

3. Results

We georeferenced 470 exposures of *Leishmania* spp., which following thinning resulted in 107 geographically resampled, presence-only occurrences. We modelled four of the five *Leishmania* spp. observed in FG. The occurrences of *L. (V.) naiffi* were scarce (1/470; 0.93%) and could not be resampled to be used in the training process. The composition of the training data included *L. (L.) amazonensis* (8/107; 7.48%), *L. (V.) braziliensis* (18/107; 16.82%), *L. (V.) guyanensis* (76/107; 71.03%), and *L. (V.) lainsoni* (5/107; 4.67%). All models used MaxENT as the base algorithm.

3.1. *L. (L.) Amazonensis model*

The SDM model of *L. (L.) amazonensis* (Fig. 2) was evaluated and measured to have an AUC of 0.982 and a kappa of 0.034. The model sensitivity and specificity of 100% and 96% respectively. All bioclimatic variables excepting altitude and mean diurnal range contributed equally to the model construction (Table 1).

3.2. *L. (v.) braziliensis model*

The predictive risk of *L. (V.) braziliensis* is mapped in Fig. 3. The evaluation of SDM model observed an AUC of 0.632, kappa 0.003 and

<table>
<thead>
<tr>
<th>Bioclimatic variables</th>
<th>Amazonensis SDM</th>
<th>Braziliensis SDM</th>
<th>Guyanensis SDM</th>
<th>Lainsoni SDM</th>
<th>SSDM mean [95% CI]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Altitude</td>
<td>4.72</td>
<td>4.61</td>
<td>4.72</td>
<td>4.79</td>
<td>4.71 [4.7-4.72]</td>
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<td>4.78 [4.78-4.76]</td>
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<tr>
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<td>4.76 [4.75-4.77]</td>
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<td>Isothermality</td>
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<td>4.78</td>
<td>4.74</td>
<td>4.79</td>
<td>4.77 [4.77-4.77]</td>
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<tr>
<td>Temperature Seasonality</td>
<td>4.77</td>
<td>4.80</td>
<td>4.75</td>
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<td>4.64 [4.59-4.69]</td>
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<tr>
<td>Max Temperature of Warmest Month</td>
<td>4.77</td>
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<td>4.73</td>
<td>4.79</td>
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sensitivity and specificity of 60% and 66% respectively. Precipitation seasonality contribute to a lesser extent in comparison to the other bioclimatic variables (Table 1).

3.3. L. (v.) Guyanensis model

SDM model of L. (V.) guyanensis mapped the predictive risks of the potential sites of exposure (Fig. 4). The evaluation of the model observed an AUC, kappa, sensitivity, and specificity of 0.778, 0.036, 78% and 78% respectively. All bioclimatic variables excepting altitude and precipitation at the driest quarter contributed equally to the model construction (Table 1).

3.4. L. (v.) Lainsoni model

The predictive risk of L. (V.) lainsoni illustrated in Fig. 5. The evaluation of SDM model observed an AUC, kappa, sensitivity, and specificity of 0.976, 0.013, 100% and 95% respectively. All climatic variables contributed equally to the model construction. Temperature seasonality and precipitation at the driest quarter contributed to a lesser extent in comparison to the other bioclimatic variables (Table 1).

3.5. SSDM of Leishmania spp

The SDM models were stacked and outputs of the four Leishmania spp. were aggregated in SSDM maps of species richness and composition using the summing continuous habitat suitability maps stacking method (SSDM). The local Leishmania species richness and endemism maps are illustrated in Fig. 6. We observed a mean species richness error of 0.654 and prediction success, sensitivity and specificity of 70%, 80% and 67% respectively. The community similarity calculated by the Jaccard index was 4.8%. Precipitation and temperature seasonality along with altitude contributed to a lesser extent than other bioclimatic variables (Table 1).
3.6. Deforestation analysis

Occurrences of L. (V.) guyanensis (3050.21; 95% CI [2251.44, 3848.98]) and L. (V.) braziliensis (1261.04, 95% CI [781.22, 1740.86]) were found to have higher tree cover loss at the radius of 15 km than the pseudo-absence points. While the tree cover loss around the regions of L. (L.) amazonensis 368.66, 95% CI [160.22, 833.54] and L. (V.) lainsoni 111.26, 95% CI [158.81, 381.33] occurrence was not found to be significant in comparison with the pseudo-absence points.

4. Discussion

The major finding of this study is the presence of variations in the spatial patterns and clusters, suggesting that each Leishmania spp. has a specific climate niche and potentially different or exclusive vectors and reservoirs. The northeastern region of FG was found to have the highest species richness (Fig. 6). Cases with reported exposure from this region must be subject to molecular diagnosis for species identification prior to treatment, especially for travelers visiting this region. This can limit treatment failure especially with L. (V.) braziliensis usually fail to respond to pentamidine isethionate, the first-line treatment of L. (V.) guyanensis CL in FG and instead, relies on parenteral meglumine antimoniate or liposomal amphotericin B [13]. Evidence on the seasonal distribution of the Leishmania spp. is scarce but might provide an insight if there are temporal variations between the different species. Our species distribution modelling of the eco-epidemiological spatial patterns of the Leishmania spp. is the first to measure endemism based on bioclimatic factors in FG. Precipitation seasonality and altitude were found to
contribute the least in the distribution of the *Leishmania* spp. This can be explained by the influence of environmental predictors on the disease vector, sandflies, rather than the pathogen itself. The study also highlights the impact of deforestation on the increasing distribution of *L. (V.) braziliensis* in FG.

Studies prior to 1980 report no evidence of *L. (V.) braziliensis* in FG [14,15]. In the recent decade, there has been an increase in *L. (V.) braziliensis* cases in FG. Our results demonstrate that *L. (V.) braziliensis* was the second most common *Leishmania* spp. in FG with its distribution along the coast and clusters along the Oyapock, Maroni Rivers and deep in the primary rainforests at the tri-country border (shared between FG, Suriname and Brazil). Studies hypothesized that the increase in *L. (V.) braziliensis* cases was due to the increasing presence of humans in the deeply forested areas where transmission of *L. (V.) braziliensis* usually occurs [16] as cases occurred mostly amongst illegal gold miners and military personnel. This explains the cluster of cases at the tri-country border. Interestingly, our study findings note that the spatial distribution of *L. (V.) braziliensis* along the coast and more anthropized areas. This can be explained by the high tree cover loss in the proximity of *L. (V.) braziliensis*, which lead to increase contact of the vector and the species reservoir with humans.

Deforestation is a controversial risk factor incidence of *Leishmania*. Some studies suggest that continued deforestation lengthens the distance between habitations and forest, thus decreasing the contact between host and the vector [17], while others argue that deforestation increases the exposure of the vector to human dwellings [18,19]. Our study results supports the latter argument as *L. (V.) braziliensis* along with *L. (V.) guyanensis* were found to occur in regions with high tree cover loss. Increasing cases of *L. (V.) braziliensis* in the anthropized regions along with high tree cover loss suggests increased exposure of the vector with potential domestic reservoirs such as dogs leading to the establishment of a peridomestic life cycle [20], which was hypothesized in a previous study [21]. Reports from Brazil observe the frequent finding of cutaneous lesions due to *L. (V.) braziliensis* in dogs supporting the hypothesis that severe deforestation could lead to the formation of a peridomestic cycle between the infected wild animals (particularly rodents) and the domestic animals in the residential areas [22]. Domestic cats have also been identified as a reservoir of *L. (V.) braziliensis* in FG [23].

The impact of deforestation on *Leishmania* distribution is further emphasized by the restricted spatial spread of *L. (V.) lainsoni*, which was geographically limited to deeply forested regions inaccessible to anthropization. Tree cover loss was not found associated with *L. (V.) lainsoni* and *L. (L.) amazonensis*, as opposed to the two other widespread *Leishmania* spp... This can be explained by two distinct sylvatic cycles in *Leishmania* spp. occurring at different levels of the rainforest canopy [14]. The *L. (V.) guyanensis* cycle is located higher up in the canopy with the arboreal sand fly *Lutzomyia umbratilis* and the two-toed sloth as the vector and mammal reservoir of the canine, respectively. Tree cover loss destroys the natural habitat of the vector and sloth at the canopy, forcing their migration towards shorter trees and the rainforest periphery, thereby increasing the exposure to humans. While the other cycle with *L. (L.) amazonensis* occurs at ground level with *Lu. flaviscutellata* as the vector and *Proechymys cuvieri*, a rodent, as the main reservoir host [24]. Deforestation does not significantly disrupt the transmission at the ground level and thus, have no influence in the transmission dynamics of *L. (L.) amazonensis*.

Our results show that the climatic variables found to contribute to the spatial models of *Leishmania*. Precipitation seasonality, the tendency for a place to have more rainfall in certain months, was found to have lesser influence in the distribution of certain species and the stacked model. A decrease in rainfall was associated to increased cases two months later in FG [25]. Precipitation seasonality along with precipitation at the warmest and coldest quarter was also found to contribute the least to the distribution of the Phlebotomine sandflies in central Europe [26]. Altitude was found to contribute unanimously less across the models. This concurs with studies that demonstrate the higher risk of other *Leishmania* spp. in low-lying regions [27,28]. This demonstrates that the bioclimatic predictors influence the sandfly vectors rather than the pathogen. However, the new world sandflies of the genus *Lutzomyia* have been reported in a wide range of altitudes [29]. The decrease of *Leishmania* exposure in high altitudes in FG can be attributed to the poor access and absence of human settlements at these regions.

This study shows the use of SSDMs in analyzing the spatial patterns of different species and provides a future scope for studies exploring spatial variations in malarial strains or between the different clades of HIV in a region. Studies looking into the variations in the seasonal distribution of *Leishmania* spp. would help us understand better the transmission dynamics of the different species.

Fig. 6. Distribution of the four Leishmania sp. in French Guiana. Panel A: illustrating the local species richness of Leishmania sp.; Panel B: demonstrating the Endemism map of Leishmania.
5. Conclusion

In our study, we were able to characterize the spatial variations in the distribution of *Leishmania* species and map the predictive risk based on bioclimatic factors in FG. Our results establish that environmental predictors including temperature and rainfall influence the distribution of *Leishmania* spp. Along with tree cover loss, climatic covariates have influenced the extension of *L. (V.) braziliensis*, which was once found in the deeply forested regions, to the densely populated littoral regions. Tree cover loss was found to have a strong influence in the distribution in the of the two most common *Leishmania* spp., *L. (V.) guyanensis* and *L. (V.) braziliensis*. SDMs were found to be effective in detecting high-risk regions for the different *Leishmania* spp., in FG, which is particularly useful in the early testing and clinical management of the cases.

With increasing tree cover loss to accommodate the growing population and unpredictability in the climate, we can expect an upward trend in the cases of Leishmaniasis in the coming years in French Guiana. To effectively control and prevent the spread of Leishmaniasis, we need to identify the high-risk zones using risk maps as done in this study and enforce adequate monitoring of the vector and active surveillance for lesions in domestic animals in these regions. Other steps at a local scale include restricting deforestation for the construction of human habitation at the forest edge by better city planning. Our study demonstrates that the distribution of Leishmaniasis is spatially dependent on climatic covariates. The impact of climate change on the distribution of vector-borne zoonoses must be acknowledged and managed at a global scale by developing policy-ready management strategies for monitoring and control of the vectors.

Author Statement

SJ was involved in the conceptualization, methodology, formal analysis, writing of the original draft; MC in methodology, writing of the original draft; MG in resources, data curation and writing of the original draft; SS in in resources and data curation; GP in resources, data curation and writing- review and editing; PC in conceptualization, writing of the original draft; REG in conceptualization, writing of the original draft; MC in methodology, writing of the original draft; GP in conceptualization, writing of the original draft and supervision.

Declaration of Competing Interest

The authors declare no conflict of interest.

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