Predicting the geographic distribution of *Lutzomyia longipalpis* (Diptera: Psychodidae) and visceral leishmaniasis in the state of Mato Grosso do Sul, Brazil

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To understand the geographic distribution of visceral leishmaniasis (VL) in the state of Mato Grosso do Sul (MS), Brazil, both the climatic niches of Lutzomyia longipalpis *and VL cases were analysed. Distributional data were obtained from 55 of the 79 counties of MS between 2003-2012. Ecological niche models (ENM) of* Lu. longipalpis *and VL cases were produced using the maximum entropy algorithm based on eight climatic variables.* Lu. longipalpis *showed a wide distribution in MS. The highest climatic suitability for* Lu. longipalpis *was observed in southern MS. Temperature seasonality and annual mean precipitation were the variables that most influenced these models. Two areas of high climatic suitability for the occurrence of VL cases were predicted: one near Aquidauana and another encompassing several municipalities in the southeast region of MS. As expected, a large overlap between the models for* Lu. longipalpis *and VL cases was detected. Northern and northwestern areas of MS were suitable for the occurrence of cases, but did not show high climatic suitability for* Lu. longipalpis. *ENM of vectors and human cases provided a greater understanding of the geographic distribution of VL in MS, which can be applied to the development of future surveillance strategies.*

Key words: *Lutzomyia longipalpis* - visceral leishmaniasis - Maxent - climatic niches

Visceral leishmaniasis (VL) is a severe systemic disease resulting from infection with *Leishmania infantum* (syn. *Leishmania chagasi*). In the Americas, domestic dogs are the main reservoirs of the parasite, which is transmitted mainly by *Lutzomyia longipalpis* (Diptera: Psychodidae) (Gontijo & Melo 2004), a synanthropic sandfly species with a wide geographic distribution in Brazil (Souza et al. 2009). Between 2003-2007, approximately 3,500 VL cases were reported per year in Brazil, which corresponds to 95% of all new cases of VL in the Americas (Alvar et al. 2012). Control measures directed against sandflies have been unsuccessful and the early detection and treatment of human cases remains the most important strategy for reducing case fatalities (Romero & Boelaert 2010).

In the state of Mato Grosso do Sul (MS), VL was restricted to the municipalities of Corumbá and Ladário until 1993 (Antonialli et al. 2007), but its presence is currently registered in 41 municipalities. An increase in the number of cases diagnosed in urban centres was observed, reflecting both disease expansion and urbanisation in the state as well as representing a threat to public health, considering the mean disease lethality

of 7.8% (Oliveira et al. 2006b). The Notifiable Diseases Information System recorded 1,919 cases of VL in MS between 2003-2011, which represents 75% of all cases recorded in Central-West Brazil. Most cases occurred in the municipalities of Aquidauana, Campo Grande and Três Lagoas, which are currently classified as areas of intense transmission of VL, where *Lu. longipalpis* has also been detected (Silva et al. 2007, MS/SVS 2013).

The geographic distribution of *Lu. longipalpis* extends from Mexico to Argentina (Deane & Deane 1962, Sherlock 1996, Souza et al. 2009, Salomón et al. 2011). This species occurs in areas with different climatic conditions, ranging from the semi-arid areas in the *Caatinga* biome to wetter areas in other Brazilian biomes and presents great adaptability to different habitats and environmental changes (Ximenes et al. 2000, Aguiar et al. 2003, Missawa & Lima 2007). In MS, *Lu. longipalpis* is the primary species captured in anthropic environments (Oliveira et al. 2006b, Almeida et al. 2010a, b) and is the principal vector of *L. infantum*. Transmission, however, can occur by *Lutzomyia cruzi* in Corumbá in northwest MS (Santos et al. 1998).

Ecological niche models (ENM) have been used to predict the occurrence areas of sandflies and leishmaniasis in Brazil. Peterson and Shaw (2003) predicted climate change effects on *Nyssomyia whitmani* distribution; specifically, under climate change scenarios, an extension of the geographic range of *Ny. whitmani* was predicted in Southeast Brazil, where cutaneous leishmaniasis appears to be re-emerging. Peterson et al. (2004) demonstrated that elevation and vegetation significantly

influence the distribution of sandflies in the state of São Paulo (SP). In the state of Bahia, predictive models estimated a higher incidence of VL in hot and dry areas (*Caatinga*), whereas areas of *Cerrado* and the Atlantic Forest were considered to be less suitable for disease transmission (Nieto et al. 2006). Through logistic regression, Zeilhofer et al. (2008) demonstrated that the spatial distribution of *Ny. whitmani* is associated with deforestation and with lower rates of gross domestic product in the state of Mato Grosso (MT).

Given the proven usefulness of spatial modelling techniques in understanding the distribution of sandflies and leishmaniasis and the fact that the predictive analysis of the geographic distribution of *Lu. longipalpis* and human cases of VL can aid in the development of monitoring and disease control programs in MS, the present study aims to analyse the climatic conditions of the occurrence areas of *Lu. longipalpis* and VL cases using ecological niche modelling.

MATERIALS AND METHODS

MS is located in the Central-West Region of Brazil, with an area of 358,200 km². It borders the states of Minas Gerais, SP and Paraná to the east and MT and Goiás to the north. Moreover, MS has international borders with Bolivia and Paraguay. The state has 78 counties, divided into 11 micro-regions, with a population of approximately two million inhabitants (IBGE/MPOG 2010). The Sedimentary Plateau Basin of the Paraná River and the South Pantanal Plains are the major geological units in MS. The biomes present in the state are the *Cerrado*, Pantanal and Atlantic Forests.

The climate of MS, according to the Köppen classification, varies between the Aw (tropical humid climate with a rainy summer and a dry winter) and the Cfa (humid mesothermal climate with no dry season) subtypes, with temperatures during the warmest month reaching over 22ºC and with low precipitation during its driest month. In MS, there is predominance of undulated elevation and of Oxisols (IBGE 1990).

Distributional data for *Lu. longipalpis* were obtained from entomological surveys carried out by the Sector of Entomology of the Vector Control Coordination of the State Department of Health and from entomology laboratories of the respective municipalities. The research was conducted between 2003-2012 in urbanised areas of 55 municipalities of MS, corresponding to 70% of the counties in this state. Samples were collected with CDC light traps that were installed at dusk inside houses and in domestic animal shelters (peridomiciles) and collections took place over a period of 12 h for three consecutive nights. In 23 of the 55 municipalities (42%), a survey of sandflies was performed via entomological monitoring over two years following the methods described in Almeida et al. (2010b). The number of points sampled varied among municipalities depending on the urban total area (between 4-12 sampling points), with capture effort ranging from 96-1,728 traps by municipality. For other municipalities, entomological surveys were conducted with an average capture effort of 50 traps. For identification of sandfly species, specific keys were used

(Galati 2003). Sandfly specimens were deposited in the Regional Laboratory of Entomology of Dourados, MS. The presence of *Lu. longipalpis* was recorded in 118 sites and their georeferencing had a precision of 0.0001º.

The present study included records of VL cases reported by local health services in the municipalities of MS during 2003-2012. The cases were georeferenced according to the coordinates of the municipality administrative centre with a reasonable degree of confidence (i.e., with an uncertainty of ≤ 5 km, to a precision of ± 0.01º). All records were georeferenced based on a consultation of Brazilian Institute of Geography and Statistic (IBGE) (ibge.gov.br/).To characterise climatic variation across MS, eight climatic variables were used: annual mean temperature, diurnal temperature range, temperature seasonality, maximum temperature in the warmest month, minimum temperature in the coldest month, annual precipitation, precipitation in the wettest month and precipitation in the driest month. These variables were obtained from the WorldClim project (worldclim.org) and were developed via the interpolation of mean monthly climatic data from meteorological stations over 30-50 years (1950-2000), depending on data availability at stations (Hijmans et al. 2005). These variables were selected to avoid the confounding effects of calibrating models in an overly dimensional environmental space (Peterson & Nakazawa 2008). The climatic database used in the analysis presented a spatial resolution of 5 x 5 km per pixel.

Models were produced using Maxent v.3.2.1 (Phillips et al. 2006), applying default parameters with the exception that we chose a random seed, with 10 replicate analyses based on bootstrap subsampling. We used the median output grids as the best hypothesis of potential range and imported them into ArcView v.3.3 for assessment and analysis. Distributional data for *Lu. longipalpis* and human cases of VL were separated into two sets: one for model calibration (75% of points) and the other for model evaluation (25% of points). For *Lu. longipalpis*, several sites were near one another and the model was produced by using a reduced number of spatially unique points, considering the spatial resolution of our data.

The quality of the models generated was evaluated using the receiver operating characteristics (ROC) curve, which correlates the sensitivity and specificity of model predictions across multiple thresholds (Phillips et al. 2006). The area under the ROC curve (AUC) provides a measure of model performance across many thresholds. AUC values close to one indicate high performance, whereas readings below 0.5 indicate the poor performance of a model (Elith et al. 2006). Model accuracy was assessed by examining the omission rates associated with test points (Anderson et al. 2002) and Maxent's jackknife test was used to identify variables that most influence model predictions. The contribution of each variable to the model was estimated through the permutation importance provided by Maxent (Phillips et al. 2006). To test model significance, we compared the predictive success of the models against null expectations using a cumulative binomial test. In particular, we assessed whether each test point fell in the areas that were identified by the model as suitable and compared this success rate with

overall proportions of pixels identified as suitable or unsuitable for that species.

Finally, to analyse the overlapping area between the models of *Lu. longipalpis* and VL cases, the raw Maxent outputs were converted into binary maps showing the presence or absence based on a threshold of 0.60 (values above this threshold were considered presence and they represented higher climate suitability). Then, the overlapping area was calculated using the ArcView Spatial Analyst extension.

RESULTS

Lu. longipalpis occurred in 37 (67%) of the 55 municipalities surveyed. The predictive model indicated wide potential distribution of *Lu. longipalpis* in MS. The highest climatic suitability for *Lu. longipalpis* was observed in the south of the state (Fig. 1). *Lu. longipalpis* was not registered in areas with high climatic suitability for vector occurrence in several municipalities of MS, such as Navirai, Itaquira and Juti. Temperature seasonality and annual mean precipitation were the variables that most influenced the distribution of *Lu. longipalpis* (Maxent's permutation importance = 79.7 and 4.9, respectively). The model revealed 0% error of omission and AUC = 0.992 and was statistically significant (p < 0.01) as indicated by the binomial test.

Two areas of high climatic suitability for the occurrence of VL cases were predicted: one near Aquidauana, Dois Irmãos do Buriti and Anastácio and another encompassing several municipalities in the southeast region of the state, such as Nova Alvorada do Sul, Angélica and Jatei (Fig. 1). Temperature seasonality and precipitation of the driest month were the variables that most influenced models of VL cases (Maxent's permutation importance = 67.4 and 7.3, respectively). The model showed 0% error of omission and AUC = 0.991 and was also statistically significant (p < 0.01).

A large area of overlap between the models of *Lu. longipalpis* and VL cases was detected. When considering all values of climatic suitability of the VL and *Lu. longipalpis* models, 100% of MS territory was suitable for the occurrence of vector and disease (Fig. 1). However, the northern and northwestern areas of MS were suitable for the occurrence of cases and did not show high climatic suitability for the occurrence of *Lu. longipalpis*. By superimposing the models of VL cases and *Lu. longipalpis*, considering the higher values of climatic suitability (> 0.60), areas climatically more favourable for the occurrence of *Lu. longipalpis* and VL cases were observed, representing 41% of MS territory (Fig. 2). This overlapping area included most municipalities of intense VL transmission (e.g., Aquidauana, Anastácio, Campo Grande, Três Lagoas) and several vulnerable municipalities for VL transmission in southeast MS (Fig. 2).

DISCUSSION

Predictive models indicated that almost the entire territory of MS has the potential for VL occurrence. In areas more suitable for the occurrence of VL cases and less suitable for the occurrence of *Lu. longipalpis*, transmission is possible by *Lu. cruzi* (Santos et al. 1998). This scenario should occur in Corumbá (northwest MS) and three other municipalities in northern MS (e.g., São Gabriel do Oeste, Camapuã and Cassilândia) where *Lu. cruzi* was detected. The models also indicated that in southern areas of MS, there is high climatic suitability for the occurrence of *Lu. longipalpis*, although the species has only been recorded in one municipality based on
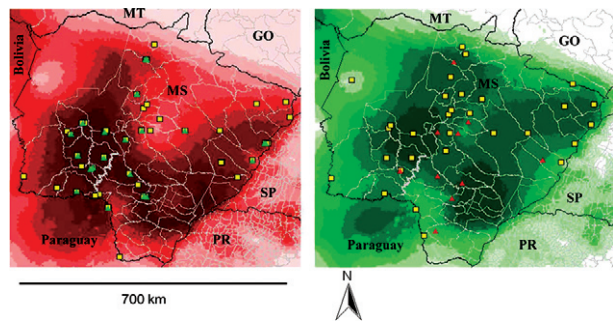

Fig 1: ecological niche models projected for the potential distribution of *Lutzomyia longipalpis* (left) and human cases of visceral leishmaniasis (right). Areas identified as suitable based on the records of the occurrence of *Lu. longipalpis* are shown on a red scale: light red (low suitability) to dark red (high suitability), while areas identified as suitable based only on VL cases are presented on a green scale: light green (low suitability) to dark green (high suitability) . The lines indicate the political boundaries of the states and its respective municipalities (GO: state of Goiás; MS: state of Mato Grosso do Sul; MT: state of Mato Grosso; PR: state of Paraná; SP: state of São Paulo). The records used to calibrate the models are represented by squares, while the records used to test the models are represented by triangles.

1: Aquidauana (136)
2: Miranda (27)
3: Bodoquena (9)
4: Porto Murtinho (2)
5: Bonito (28)
6: Anastácio (80)
7: Nioaque (7)
8: Guia Lopes da Laguna (10)
9: Jardim (33)
10: Bela Vista (15)
11: Dois Irmãos do Buriti (14)
12: Sidrolândia (9)
13: Maracaju (11)
14: Rio Brilhante (3)
15: Itaporã (0)
16: Dourados (4)
17: Caarapó (0)
18: Juti (0)
19: Navirai (0)
20: Amambai (0)
21: Nova Alvorada do Sul (0)
22: Campo Grande (1462)
23: Ribas do Rio Pardo (18)
24: Nova Andradina (0)
25: Angélica (0)
26: Ivinhema (0)
27: Novo Horizonte do Sul (0)
28: Jateí (0)
29: Taquarussú (0)
30: Bataiporã (0)
31: Anaurilândia (0)
32: Bataguassú (0)
33: Santa Rita do Pardo (0)
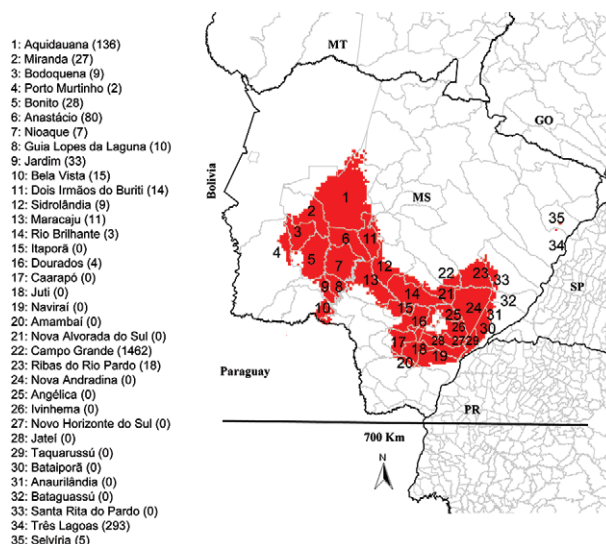34: Três Lagoas (293)
35: Selvíria (5)

Fig 2: overlapping ecological niche models of *Lutzomyia longipalpis* and human cases of visceral leishmaniasis (VL) in Mato Grosso do Sul (MS). The overlapping area is shown in red. The white areas indicate the absence or predicted climatic suitability smaller values (values < 0.60). The lines indicate the political boundaries of the states and its respective municipalities (GO: state of Goiás; MS: state of Mato Grosso; MT: state of Mato Grosso; PR: state of Paraná; SP: state of São Paulo). The numbers indicate the name of the municipalities of MS and the numbers between parentheses indicate notified human cases of VL in MS (2003-2012).

conducted entomological research. This result illustrates a practical application of ecological niche modelling by indicating suitable municipalities for vector occurrence in cases of entomological research with low capture effort or even unrealised sampling.

Lu. longipalpis was the most abundant species in the anthropic environment in municipalities of MS such as Campo Grande, where an increase of 60 times in the density of this species was registered compared to the last sandfly evaluation in 1999/2000 (Oliveira et al. 2006a). The factors associated with the increase in the occurrence and abundance of *Lu. longipalpis* in MS are only partially understood. Oliveira et al. (2012) found a significant positive linear correlation between the abundance of *Lu. longipalpis* and the percentage of vegetation cover; moreover, these researchers observed the highest abundance of sandflies in houses with chickens. The attractiveness of chickens to sandflies has often been described (Lainson & Rangel 2005).

Spatial analysis of VL in MS indicated that the expansion route of the disease has been from west to east, coinciding with the routes of three anthropogenic disturbances of the environment, such as the construction of a gas pipeline that began in 1998 and a federal highway and railroad that were constructed in the early XX century (Antonialli et al. 2007). The authors hypothesise that the intense migration of workers from the VL endemic region in the city of Corumbá explains the new epidemiological pattern of the establishment of this disease in MS. Our study indicated two areas of high climatic suitability for the occurrence of VL cases. The area near Aquidauana, Dois Irmãos do Buriti and Anastácio coincides with high predicted VL incidence rates in MS based on geostatistical models (Karagiannis-Voules et al. 2013). The incidence of VL in southeast MS is still low, but the present study indicates that climatic conditions are favourable for the occurrence of *Lu. longipalpis* in this area. Thus, municipalities near Nova Alvorada do Sul, Angélica and Jatei should be monitored.

The influence of climatic variables on the occurrence of sandflies and VL has been observed in other studies (Franke et al. 2002, Peterson & Shaw 2003). In this study, the variable that most influenced the models was temperature seasonality. Quintana et al. (2013) noted that precipitation seasonality and precipitation in the warmest quarter were the variables that most contributed to the ENM of *Lutzomyia neivai* and *Lutzomyia migonei* in Argentina. These variables, as well as temperature seasonality, are indicative of areas with well-defined dry and rainy seasons. In Brazil, the majority of VL cases have been recorded in northeastern and Central-West Brazil (MS/SVS 2013), which present a well-defined dry season. Ecological studies of sandflies in this region have indicated that the population density of these insects increases after the end of the rainy season (Sherlock 1996), which has also been observed for *Lu. longipalpis* in MS (Oliveira et al. 2003, Almeida et al. 2010a).

Other variables, such as deforestation and socioeconomic factors, which have been linked to the occurrence of other sandfly species (Zeilhofer et al. 2008), may also influence the occurrence of *Lu.* longipalpis. Environmental disturbances, rural-urban migration, increased population density, a higher amount of solid waste production without proper destination and the close proximity of residents to domestic animals are some of the factors that may facilitate the urbanisation of VL in municipalities with more than 100,000 inhabitants (MS 2006).

A limitation of the niche model for VL cases in MS was the use of the coordinates of the municipality administrative centre. A more refined analysis could be performed with the geographical coordinates of all confirmed human cases of VL in MS. Unfortunately, this information was not available for the present study. Future studies based on the specific location where infection occurred would increase the number of records analysed and allow for a better comparison between niche models of vector species and the occurrence of human cases. Moreover, studies analysing the effects of deforestation and demographic and socioeconomic variables for the occurrence of *Lu. longipalpis* and VL in MS may improve the predictive models presented in our study and aid in understanding the geography of disease transmission in the state. Analysis with higher spatial resolution, considering niche characteristics over future climate change scenarios (Moo-Llanes et al. 2013) and using different spatial analysis (Sarkar et al. 2010, Karagiannis-Voules et al. 2013), may refine the predictive models presented to better assess the risk factors for transmission of VL in MS.

Species distribution models may contribute to the planning of measures designed to control vector-borne diseases. Modelling enables the development of geographic predictions of pathogens transmitted by insect vectors, with important implications for human health. Accordingly, the methodological approach presented here may serve as a model for studies in other Brazilian states. ENM of vectors and human cases allows for a better understanding of the geographic distribution of VL in MS, which can be applied to the development of future strategies for the surveillance and control of this disease.

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