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MARÍLIA MELO FAVALESSO

CONDIÇÕES ECOLÓGICAS E PREDIÇÃO DE ÁREAS ADEQUÁVEIS PARA OCORRÊNCIA DE *Lonomia obliqua* Walker 1855 NO BRASIL

CASCAVEL-PR
Fevereiro/2018

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Condições ecológicas e predição de áreas adequáveis para Lonomia obliqua Walker 1885 no Brasil

Dissertação apresentada ao Programa de Pós-Graduação em Conservação e Manejo de Recursos Naturais em cumprimento parcial aos requisitos para obtenção do título de Mestra em Conservação e Manejo de Recursos Naturais, área de concentração Ciências Ambientais, linha de pesquisa Biologia Aplicada e Indicadores de Qualidade No Ambiente Terrestre. APROVADO(A) pela seguinte banca examinadora:

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Dedico o meu trabalho a todos os cientistas, em especial a minha orientadora.

Também dedico a minha família e aos meus amigos.

Dedicatória

“Toda a nossa ciência, comparada com a realidade, é primitiva e infantil – e, no entanto, é a coisa mais preciosa que temos” Albert Einstein (1879-1955)

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> *"A ciência é muito mais do que um corpo de conhecimento. É uma maneira de pensar. E isso é fundamental para o nosso sucesso. A ciência nos convida a aceitar os fatos, mesmo quando eles não estão de acordo com nossos preconceitos. Ela nos aconselha a levar hipóteses alternativas em nossas cabeças e ver quais são as que melhor correspondem aos fatos. Impõe-nos um equilíbrio perfeito entre a abertura sem obstáculos a novas ideias, por mais heréticas que sejam, e o mais rigoroso escrutínio cético de tudo – estabelecendo novas ideias e sabedoria. Precisamos da ampla apreciação desse tipo de pensamento. Funciona. É uma ferramenta essencial para uma democracia em uma era de mudança. Nossa tarefa não é apenas treinar mais cientistas, mas também aprofundar a compreensão pública da ciência."*
>
> CARL SAGAN, "Why we need to understand science", 1990

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Resumo geral

Lonomia obliqua Walker 1855 (Saturniidae: Hemileucinae) é uma espécie de mariposa de interesse sanitário no Brasil. Suas larvas são agentes etiológicos do lonomismo, uma forma de erucismo causado pelo contato dos seres humanos com as estruturas urticantes da espécie. Os sintomas mais preocupantes do lonomismo são os quadros hemorrágicos sistêmicos que podem conduzir a diversos desfechos, inclusive o óbito. As primeiras notificações oficiais de acidentes com a espécie datam do final da década de 80, no estado do Rio Grande do Sul. A partir de então, diversos acidentes têm sido documentados no Brasil, principalmente nas regiões sul e sudeste do país. Com o aumento do número de vítimas, autoridades sanitárias do estado de São Paulo, representadas pelo do Instituto Butantã, desenvolveram um soro *antilonômico*, o qual é distribuído pelo Ministério da Saúde em localidades com maior prevalência de acidentes. Hipóteses têm sido levantadas sobre a relação entre o crescimento dos casos de lonomismo e a ocupação humana; contudo, pouco se conhece sobre a distribuição espacial e aspectos ecológicos da espécie para possibilitar os testes destas hipóteses. Diante do exposto, o presente estudo objetivou produzir um mapa para a distribuição geográfica potencial de *L. obliqua* no Brasil, baseando-se na combinação de diferentes algoritmos ENM (*Ecological Niche Modeling*). Foram utilizados 38 pontos de ocorrência distribuídos pela área geográfica do Brasil e região de Misiones, na Argentina, os quais foram particionados para calibração e avaliação do modelo de distribuição. Foram selecionadas oito variáveis contínuas climáticas e de solo entre 16 previamente cogitadas. Diferentes metodologias ENM foram testadas e confrontados quanto a valores de índice TSS (*True Skill Statistic*). O mapa-modelo final foi composto por uma combinação de quatro algoritmos (Gower, Mahalanobis, Maxent e SVM), com amostragens de pseudo-ausências fora de um envelope bioclimático e número de pseudo-ausências igual ao de presenças. Esse mapa-modelo foi binarizado a partir do limiar LPT (*Lowest Presence Threshold*) e recortado somente para o Brasil. Segundo este mapa-modelo, as áreas preditas como adequáveis a *L. obliqua* estariam restritas as latitudes ~12º e ~32º, e as longitudes ~39º e ~57º. Também foi realizada uma caracterização das variáveis abióticas relacionadas ao nicho da espécie, sendo essas extraídas da área predita como adequada a presença da espécie no mapa-modelo. O percentual de classes de uso da terra também foi extraído, a fim de contribuir com as hipóteses que condicionam o aumento de acidentes em função da ocupação humana. Neste quesito, encontramos grande parte da área predita dentro de classes de solos agrícolas no Brasil, o que nos leva a ratificar as hipóteses atuais. Assim, a perda de habitat da espécie para os empreendimentos agrícolas aumenta o contato humano com a espécie, o que deve aumentar o número de notificações do lonomismo, gerando maior preocupação a nível epidemiológico e de conservação de habitat para essa espécie.

Palavras-chave: Animais venenosos; Modelagem de distribuição de espécies; Modelagem de nicho; Nicho fundamental; Taturana.

Ecological conditions and prediction of available areas for *Lonomia obliqua* walker 1855 in Brazil

General abstract

Lonomia obliqua Walker 1855 (Saturniidae: Hemileucinae) is a species of moth of sanitary interest in Brazil. Their larvae are etiological agents of lonomism, a form of erucism caused by the contact of the human beings with the stinging structures of the species. The most worrying symptoms of lonomism are the systemic hemorrhagic conditions that can lead to several outcomes, including death. The first official notifications of accidents with the species date back to the end of the 80s, in the state of Rio Grande do Sul. Since then, several accidents have been documented in Brazil, mainly in the south and southeast regions of the country. With the increase in the number of victims, health authorities in the state of São Paulo, represented by the "Instituto Butantã", developed an anti-lonomic serum, which is distributed by the Ministry of Health in places with a higher prevalence of accidents. Hypotheses have been raised on the relation between the growth of the cases of lonomismo and the human occupation; however, little is known about the spatial distribution and ecological aspects of the species to enable the testing of these hypotheses. In view of the above, the present study aimed to produce a map for the potential geographical distribution of *L. obliqua* in Brazil, based on the combination of different ENM (*Ecological Niche Modeling*) algorithms. A total of 38 occurrence points were distributed across the geographic area of Brazil and Misiones, Argentina, which were partitioned for calibration and evaluation of the distribution model. Eight continuous climatic variables and only 16 previously considered variables were selected. Different ENM methodologies were tested and compared to TSS (True Skill Statistic) index values. The final model-map was composed of a combination of four algorithms (Gower, Mahalanobis, Maxent and SVM), with pseudo-absences outside a bioclimatic envelope and a number of pseudo-absences equal to that of presences. This model map was binarized from the *Low Presence Threshold* (LPT) and cut only for Brazil. According to this model map, the areas predicted as suitable for *L. obliqua* would be restricted to latitudes ~12° and ~32°, and longitudes ~39° and ~57°. When evaluating new sites of occurrence of the specie in Rio Grande do Sul, it was possible to verify that all the municipalities were in areas predicted by the model-map. A characterization of the abiotic variables related to the niche of the specie was also carried out, being these extracted from the area predicted as adequate the presence of the specie in the model map. To help characterize these variables, we also extract categorical descriptors of climate, soil and vegetation (in %). The percentage of land use classes was also extracted in order to contribute to the hypothesis that condition the increase of accidents due to human occupation. In this question, we find a large part of the area predicted within classes of agricultural soils in Brazil, which leads us to ratify the current hypotheses. Thus, the loss of habitat of the species for the agricultural enterprises increases the human contact with the specie, which should increase the number of notifications of the lonomism, generating greater epidemiological concern and habitat conservation for this specie.

Keywords: Venomous animals; Modeling of species distribution; Niche modeling; Fundamental niche; Taturana.

1. Introdução geral

1.1. Caracterização geral

A ordem Lepidoptera constitui uma das maiores ordens de insetos conhecidos, com cerca de 500 mil espécies em todo o mundo (DUARTE et al., 2012). Somente no Brasil são conhecidas quase 26 mil espécies de lepidópteros, cerca de metade do total encontrado na região neotropical (DUARTE et al., 2012). São insetos que possuem asas recobertas de escamas na fase adulta (*Lepido* = escamas; *ptera* = asa), com corpo vermiforme na fase larval, e algumas espécies apresentando cerdas (MORAES, 2009). A ordem abrange os insetos conhecidos como borboletas e mariposas.

A importância dos integrantes da ordem Lepidoptera está relacionada tanto aos seus benefícios ambientais quanto às nocividades causadas por suas espécies (CORSEUIL; SPECHT; CRUZ, 2008). Há inúmeras espécies que prestam serviços ambientais, como a polinização e controle biológico, mas também há aquelas consideradas como pragas agrícolas ou nocivas à saúde pública (CORSEUIL; SPECHT; CRUZ, 2008).

Os lepidópteros de importância médica representam uma pequena parcela de espécies, com quatro principais famílias no Brasil: Megalopygidae, Saturniidae, Limacodidae e Arctiidae (MORAES, 2009). Dentre os acidentes com lepidópteros, aqueles causados pelo contato com as formas larvárias desses animais (também chamadas *lagartas urticantes*) são os mais frequentes, sendo que graves sintomas podem levar o enfermo a óbito.

No Brasil, destaca-se a mariposa *Lonomia obliqua*, pertencente à família Saturniidae, subfamília Hemileucinae. Esta espécie é o agente etiológico do lonomismo, uma forma de erucismo (acidentes com larvas) responsável por inúmeros acidentes hemorrágicos no sul da América do Sul (CHUDZINSKI-TAVASSI; ZANNIN, 2011). A espécie *Lonomia achelous* também causa acidentes hemorrágicos, porém esta ocorre ao norte do continente Sul-Americano (LEMAIRE, 2002a; CHUDZINSKI-TAVASSI; ZANNIN, 2011).

Os estágios larvais de *L. obliqua*, também denominados de “taturanas”, se aglomeram em espécies vegetais arbóreas onde passam por seis instares (LORINI, 1999). Durante estes estágios, a espécie apresenta coloração entre o marrom-claro e o marrom-claro-esverdeado, cores muito semelhantes às dos troncos das árvores hospedeiras (LORINI, 1999) (Figura 1). É nesta fase do ciclo de vida que a espécie apresenta “espinhos urticantes” (escolos) ao longo do corpo, sendo que o contato acidental dos seres humanos com essas estruturas desencadeia o lonomismo (CHUDZINSKI-TAVASSI; ZANNIN, 2011). Ao tocar a pele, os escolos se fragmentam e liberam o conteúdo do veneno. As formas de acidentes mais comuns são pelo toque de crianças e de adultos andando em matas, praças e pomares (CRUZ; BARBOLA, 2016; LORINI, 2008).

Figura 1 - Colônia de *Lonomia obliqua* em tronco de hospedeiro desconhecido.
Fonte: Divulgação: CIT/UFSC.

Os principais sintomas do lonomismo variam entre ardor e dores locais severas, reações alérgicas associadas à dermatite urticante, problemas respiratórios, osteocondrites, coagulopatias, insuficiência renal e hemorragia intracerebral (CHUDZINSKI-TAVASSI; ZANNIN, 2011). A gravidade dos sintomas é altamente variável, sendo dependente principalmente do grau de contato humano com as larvas (ABELLA et al., 1999). Sem o tratamento emergencial adequado, as vítimas podem morrer rapidamente, podendo o óbito ser oriundo da hemorragia cerebral aguda, ou mesmo da insuficiência renal aguda (DIAZ, 2005).

Os registros de acidentes com larvas de *L. obliqua* começaram de maneira alarmante no final da década de 80, na região sul do Brasil, em áreas rurais dos estados do Rio Grande do Sul e de Santa Catarina (CHUDZINSKI-TAVASSI;

ZANNIN, 2011). Hoje já existem registros de casos de acidentes com *L. obliqua* nos estados do Paraná, São Paulo e Minas Gerais (ALMEIDA et al., 2013; CHUDZINSKI-TAVASSI; ZANNIN, 2011; CRUZ; BARBOLA, 2016; GAMBORGI et al., 2012) e também na Argentina, na província de Missiones (SÁNCHEZ et al., 2015). Chudzinski-Tavassi e Zannin (2011) reportaram um total de 4003 acidentes com a espécie nos estados do Rio Grande do Sul, Santa Catarina e Paraná, sendo registrados 25 óbitos. Os autores realizaram a consulta em artigos científicos publicados e centros de atendimentos à saúde pública. Um adendo importante é que autores têm colocado que os acidentes são subnotificados no Brasil, principalmente em áreas agrícolas. Assim, não existem registros precisos sobre o número de óbitos no Brasil.

Atualmente, o único tratamento seguro e eficaz para o lonomismo é o soro antilonômico, produzido pelo Instituto Butantã de São Paulo (DIAS DA SILVA et al., 1996). Apesar da distribuição do soro antilonômico pelo Ministério da Saúde para todo o Brasil, apenas o Rio Grande do Sul apresenta um banco de dados ativo de acidentes com o gênero, arquivado em metadados do DATASUS (<www.tabnet.datasus.gov.br>). Somente na última década (2007-2017), o estado reportou um total de 926 casos de acidentes lonômicos, com registro de 3 óbitos.

A Secretaria de Vigilância em Saúde (SVS) do Brasil atribui os óbitos decorrentes do lonomismo ao resultado do atraso de atendimento às vítimas, em especial pela falta de conhecimento do tratamento adequado e seguro pelos profissionais da saúde (SVS, 2009). Diferente dos demais acidentes com animais peçonhentos, os acidentados com sintomas de lonomismo procuram tardiamente o atendimento médico, cerca de 12 horas ou mais após o contato com as larvas de *L. obliqua* (WEN; DUARTE, 2009). Considerando que as alterações hematológicas se manifestam entre 1 e 72 horas após o contato com as larvas (TORRES; ABELLA, 2008), essa procura tardia pode ser fatal. Ademais, não existe diagnóstico com sintomatologia específica para o lonomismo, sendo este realizado apenas a partir do histórico do paciente e da descrição do espécime que desencadeou os sintomas (WEN; DUARTE, 2009). Sendo assim, o conhecimento sobre áreas de ocorrência da espécie tem importância epidemiológica, pois auxiliará os agentes da área da saúde na recepção dos enfermos, identificando sintomas em função da determinação de áreas adequáveis a ocorrência da *L. obliqua*.

Desta forma, o conhecimento de potenciais áreas de ocorrência de *L. obliqua* pode ser considerado como uma ferramenta útil para a compreensão dos aspectos eco-epidemiológicos de acidentes com a espécie. Até o presente momento, os registros das principais áreas de acidente com *L. obliqua* são áreas antrópicas de fronteira com áreas de floresta primária (áreas próximas a fragmentos de floresta, pomares e parques dentro de cidades) (LORINI, 1999). Assim, impactos antrópicos devem aumentar o risco de acidentes pelo contato com a lagarta em função da diminuição da distância entre as pessoas e a espécie (GAMBORGI et al., 2012). As principais hipóteses relacionam a redução da vegetação nativa e o crescimento das fronteiras agrícolas e das áreas urbanas (ABELLA et al., 1999; LEMAIRE, 2002; LORINI, 1999; MORAES et al., 2002).

É provável que as constantes mudanças ambientais resultantes dos fatores antrópicos sobre o uso e a ocupação da terra desequilibrem as relações ecológicas de *L. obliqua* com o seu meio, fazendo com que a espécie se desloque para novos ambientes a fim de concluir o seu ciclo de vida (ABELLA et al., 1999; LEMAIRE, 2002b; MORAES et al., 2002). Este fato é decorrente do hábito herbívoro da espécie, sendo o estágio larval a única fase em que se alimenta. A hipótese de Lemaire (2002) é que a espécie tem migrado do seu meio original para locais com espécies comercias em fronteiras agrícolas. Um fato evidenciado para esta hipótese é o relato de Lorini (1999), que demonstrou ter encontrado *L. obliqua* em espécies comerciais como goiabeira, figo e pereira. Não estando mais restrita às florestais naturais, *L. obliqua* traz maior perigo ao ser humano, o qual tem se tornando mais suscetível a acidentes (GARCIA, 2013).

Outra hipótese atribuída ao aumento de acidentes com a espécie está relacionada à supressão dos seus inimigos naturais, principalmente insetos da ordem Diptera e Hymenoptera (LORINI; CORSEUIL; CORSEUIL, 2001; MORAES et al., 2002). Tal fato parece também estar associado aos impactos antrópicos, sendo possivelmente decorrente do aumento do uso de inseticidas, culminando na redução da população destes insetos e diminuindo também o controle biológico de *L. obliqua*.

Em relação à distribuição geográfica, Lemaire (2002) coloca a ocorrência de *L. obliqua* no Brasil desde o sul do Rio Grande do Sul, até o norte da Bahia, mas sem exatidão quanto aos municípios entre estes estados (Figura 2). Assim, a distribuição exata de *L. obliqua* continua sendo uma incógnita a ser explorada, em especial sob o enfoque epidemiológico.

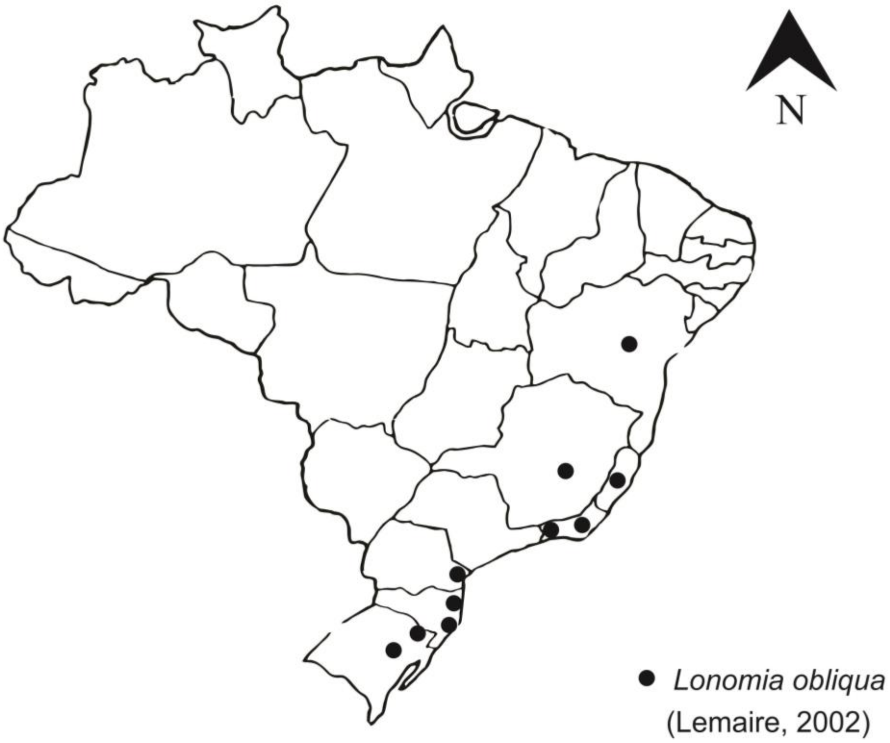

Figura 2 - Distribuição geográfica de *L. obliqua* no Brasil segundo Lemaire (2002).

1.2. Aspectos biológicos e ecológicos de *Lonomia obliqua*

De maneira geral, a *L. obliqua* apresenta quatro estágios de vida: ovo, larva, pupa e adultos. Os adultos apresentam acentuado dimorfismo sexual, sendo o macho menor do que a fêmea (LORINI, 1999) (Figura 3a e 3b). A fase adulta dura no máximo duas semanas e serve apenas para reprodução e ovoposição, pois possui peças bucais atrofiadas e não se alimenta neste período (LORINI, 2008). As fêmeas são menos ágeis do que os machos, com asas largas ornamentadas, e algumas com grandes prolongamentos caudais (LEMAIRE, 2002b). Após a cópula, a fêmea voa até a copa da árvore hospedeira onde realizará a ovoposição sobre as folhas (LORINI, 2008).

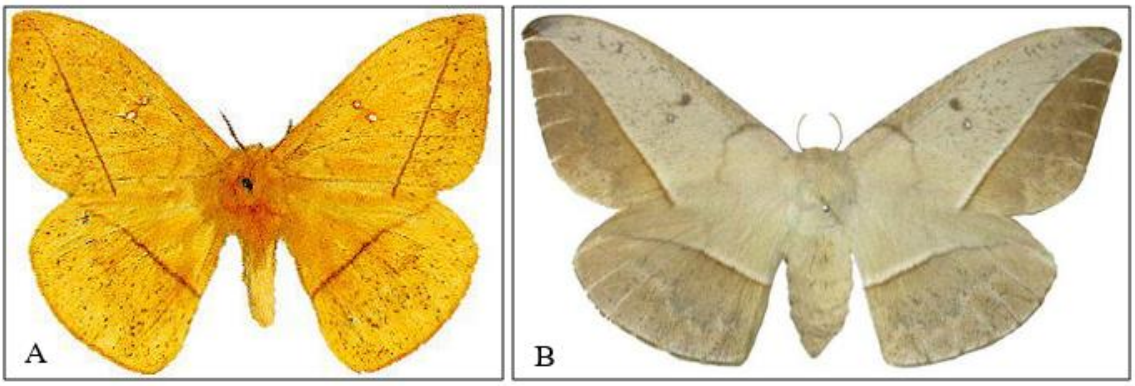

Figura 3 - Exemplares adultos de *Lonomia obliqua* (A - Macho; B - Fêmea).
Fonte: <http://www.pucrs.br/uni/poa/fabio/labento/lepidoptera/saturniidae/saturniidae.html>

Cada fêmea adulta realiza 2,8 ovoposições, em média, com fecundidade de 111,9 ovos (LORINI, 1999). Após a eclosão, as larvas em primeiro instar migram da copa da árvore em direção ao chão, aonde irão empupar, passando por seis instares sempre na mesma árvore hospedeira (Figura 4) (LORINI, 2008).

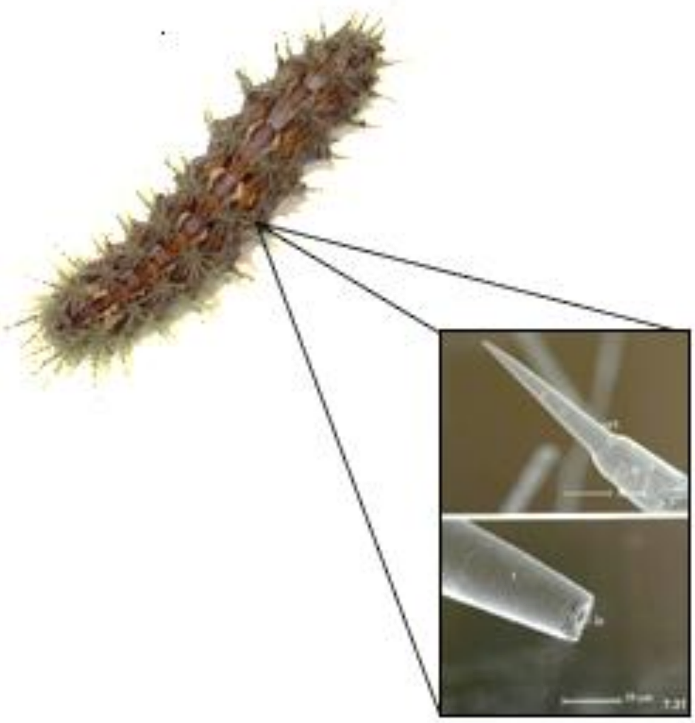

Figura 4 - Larva de *Lonomia obliqua* e cerdas urticantes.
Fonte: Larva - INMET, Argentina; Cerdas - Lorini (2008).

Durante o dia, as larvas descansam sobre o tronco da árvore hospedeira, se deslocando até a copa durante a noite para se alimentar das folhas (LORINI, 2008). Quanto mais jovens são as larvas, mais próximas à copa elas se encontram durante o dia (LORINI, 2008). As formas larvais ocorrem, geralmente, nos períodos mais quentes do ano (primavera e verão), empupando no solo entre o outono e o inverno (LORINI, 2008).

Como todas as espécies, *L. obliqua* está inserida na cadeia biológica natural, possuindo inimigos naturais que provocam a redução de sua população (LORINI, 2008), e desempenhando papel como predadora de espécies vegetais, onde também se hospeda (Quadro 1) (LORINI, 1999; MORAES et al., 2002).

Quadro 1 - Inimigos naturais e espécies vegetais hospedeiras de *L. obliqua.*

Relação ecológica	**Nome científico**	**Ordem: Família**	**Referência**
Inimigos naturais	*Belvosia viedemanni*	Diptera: Tachinidae	LORINI (1999)
	Enicospilus sp.	Hymenoptera: Ichneumonidae	LORINI (1999)
	Leschenaultia sp.	Diptera: Tachinidae	LORINI (1999)
	Moreira wiedemanni	Diptera: Tachinidae	LORINI (2008)
	Lespesia affinis	Diptera: Tachinidae	MORAES et al. (2002)
	Alcaeorrhynchus grandis	Pentatomidae: Hemiptera	MORAES et al. (2002)
	Hexamermis sp.	Nematoda: Memithidae	MORAES et al. (2002)
	LOOBMNPV	Vírus múltiplo nucleopolydrovis	MORAES et al. (2002)
	Isaria javanica	Fungi: Sordariomycetes	SPECHT et al. (2009)
Hospedeiros	*Alcornia sp.*	Malpighiales: Euphorbiaceae	DUARTE et al. (1990)
	Cedrela fissilis	Sapindales: Meliaceae	DUARTE et al. (1990)
	Erytrina crista-galli	Fabales: Fabaceae	BIEZANKO; SETA (1939)
	Eucalyptus spp.	Myrtales: Myrtaceae	BERNARDI et al., (2011)
	Ficus carica	Rosales: Moraceae	LORINI (1993)
	Ficus elástica	Rosales: Moraceae	LORINI 1999
	Ficus subtiplinervia	Rosales: Moraceae	DUARTE et al. (1990)
	Persea gratíssima	Laurales: Lauracea	LORINI (1993)
	Platanus acerifolia	Proteales: Platanaceae	LORINI (1999)
	Pyrus communis	Rosales: Rosaceae	LORINI (1999)
	Prunus domestica	Rosales: Rosaceae	LORINI (1999)
	Prunus pérsica	Rosales: Rosaceae	LORINI (1993)
	Psidium guayava	Myrtales: Myrtaceae	LORINI (1999)
	Rollinia emarginata	Magnoliales: Annonaceae	DUARTE et al. (1990)
	Tabebuia pulcherrima	Bignoniaceae	DUARTE et al. (1990)

Em relação às condições abióticas, poucos são os trabalhos que buscaram elucidar as associações de ocorrência de *L. obliqua* com variáveis ambientais como temperatura e pluviosidade. Lorini et al. (2004) reproduziu exemplares em laboratório a partir de uma temperatura média de 18,6ºC (mínimo = 13ºC; máximo = 24ºC), com umidade relativa média de 80,6% (mínimo = 64%; máximo = 92%) e fotofase de 12 horas. Lemaire (2002) coloca a espécie como residente de florestas primárias. Garcia (2013) em seu trabalho sobre as condições socioambientais de ocorrência de *L. obliqua*, tendo como base os munícipios de ocorrência da espécie no sul do Brasil, atribui a ocorrência de acidentes a uma variação de temperatura entre 20 e 25ºC e umidade próxima a 70%. A mesma autora ainda frisa que alterações ambientais resultantes do fenômeno *La niña* fizeram com que a umidade relativa do solo aumentasse, sendo uma condição propícia ao empupamento. A melhor condição de desenvolvimento do espécime garante que este atinja a fase adulta e, por conseguinte, aumente o sucesso de manutenção da prole, aumentando assim o número de larvas que promovem acidentes.

1.3. Nicho ecológico e distribuição de espécies

Considerando-se que o único tratamento seguro, eficaz e disponível para o lonomismo é o soro antilonômico produzido pelo Instituto Butantã, tornam-se necessários estudos que busquem elucidar condições de nicho de *L. obliqua*, bem como sua distribuição ao longo do território brasileiro. Uma vez que há falta de informações sobre as áreas potenciais de ocorrência da *L. obliqua*, a *modelagem de nicho ecológico e distribuição de espécies* se torna uma excelente ferramenta, permitindo estimar as condições ambientais que possibilitam a ocorrência e distribuição geográfica potencial da espécie (PETERSON et al., 2011)

O nicho ecológico está diretamente relacionado às tolerâncias ecológicas da espécie, descrevendo seu condicionamento no ambiente. As espécies tendem a se estabelecer em regiões geográficas que apresentem condições ambientais propícias a sua sobrevivência e reprodução. Essas mesmas condições também afetam a distribuição, com resultado sobre a dispersão das populações (PETERSON et al., 2011).

Grinnell (1917) foi o primeiro a utilizar a palavra nicho, definindo este conceito como “unidade de distribuição, dentro da qual cada espécie é mantida por suas limitações estruturais e instintivas”. Esse termo é hoje chamado de nicho espacial (ODUM; BARRET, 2015), sendo um reflexo das variáveis abióticas também chamadas de cenopoéticas. Dessa maneira, Grinell (1917) definiu o nicho em função de variáveis ambientais e distribuição de espécies em grande escala, sem considerar a presença de interação entre as espécies. Mais tarde, Elton (1927) introduziu o termo de nicho como uma associação do *status* funcional de um organismo com a sua comunidade, dando enfoque para as interações bióticas que as espécies apresentam com o meio, o que Hutchinson (1957) definiu mais tarde como variáveis bionômicas, as quais são medidas, principalmente, em uma escala local.

Hutchinson (1957) descreveu o conceito de nicho como o conjunto de condições e recursos que uma espécie necessita para tolerar e persistir no ambiente a fim de cumprir o seu modo de vida. Existe uma faixa ideal dentre diferentes fatores que favorecem a permanência de uma dada espécie no ambiente, ou uma dimensão de nicho para um dado organismo, considerando-se, portanto, o nicho como um hipervolume de n-dimensões. Hutchinson (1957) ainda conceitua a ocorrência de uma espécie como um reflexo do seu nicho ecológico **fundamental** (relativo às limitações fisiológicas das espécies em função das variáveis cenopoéticas) e reduzido pelo nicho **realizado** (interação da espécie com variáveis bionômicas).

Dessa forma, o termo “nicho ecológico” possui múltiplos significados que são definidos conforme o propósito ou o problema biológico abordado, mas, evidentemente, sempre relacionado ao espaço geográfico (seja ao nível de paisagem, ou local) (LIMA-RIBEIRO; DINIZ-FILHO, 2013).

A estrutura teórica de nicho ecológico representada pelo diagrama BAM (PETERSON et al., 2011; SOBERÓN, 2007; SOBERÓN; PETERSON, 2005) não apenas resume todas as teorias aqui relatadas, como as relaciona com o espaço geográfico que pode ser ocupado pela espécie (Figura 5).

De maneira geral, o círculo ‘A’ no diagrama BAM representa o espaço geográfico que apresenta as condições cenopoéticas necessárias para a reprodução e o crescimento da espécie. O círculo ‘B’ representa as condições bionômicas ideais; e o círculo ‘M’ representa os locais acessíveis à espécie dado a sua capacidade de dispersão. As áreas onde ‘A ∩ B’ representam o espaço ambiental adequado para a sobrevivência da espécie, porém, se a sua capacidade de distribuição (M) é limitada, ela pode não chegar a habitar determinada localidade. Dessa forma, as regiões que de fato são ocupadas pela espécie são definidas por ‘A ∩ B ∩ M’. Da mesma forma, qualquer região em que ‘M $\notin$ $(A \cap B)$’ será um sumidouro. Logo, a espécie poderá se dispersar para essas localidades, mas não habitá-las de fato. A região ‘M’ do diagrama BAM não é um atributo do nicho ecológico da espécie, em vista que este é definido pelo meio cenopoético e bionômico, mas sim um fator de limitação da espécie no espaço geográfico (LIMA-RIBEIRO; DINIZ-FILHO, 2013). Outro fato importante é que nem sempre o diagrama

BAM será como o apresentado na figura 5, podendo se configurar com diferentes estruturas conforme as propriedades no nicho da espécie e sua capacidade de dispersão (SOBERÓN; PETERSON, 2005).

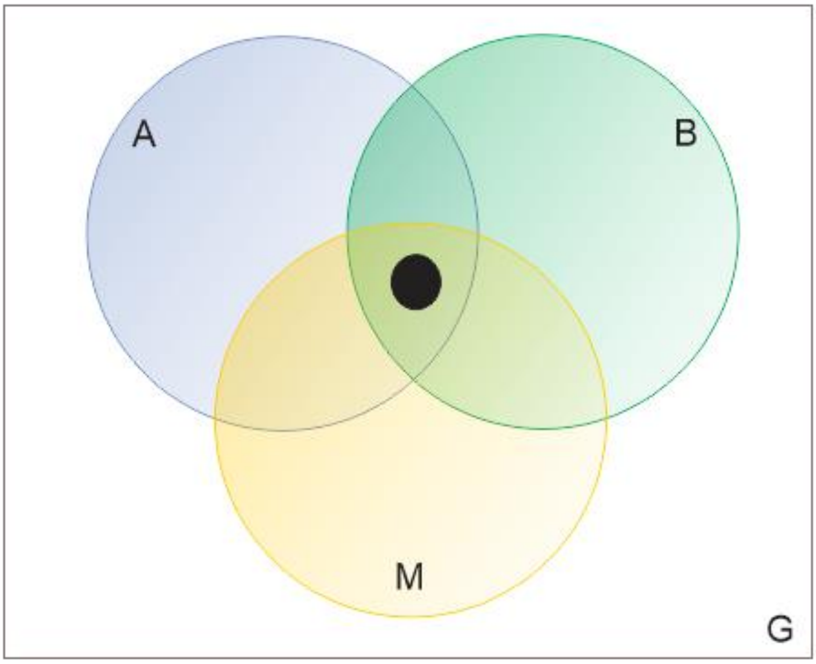

Figura 5 - Representação do diagrama BAM onde: G - representa todo o espaço geográfico de interesse; A - representa toda a região com condições cenopoéticas favoráveis ao estabelecimento, sobrevivência e reprodução da espécie; B - representa toda a região com condições bionômicas; M - representa toda a área acessível à espécie segundo a sua capacidade de dispersão; • - consiste na área ambiental e geográfica ideal para a espécie.

Os conceitos definidos por Hutchinson, tendo como base o nicho de Grinell e de Elton, são de suma importância para compreender como uma espécie se distribui no espaço, e sua dependência por variáveis ambientais, estabelecendo-se assim em regiões geográficas distintas. Com base nestes conceitos, a distribuição geográfica das espécies é reflexo de como o nicho se manifesta. Assim, é possível ponderar essa distribuição entre um espaço “virtual” de variáveis ambientais, onde o nicho efetivamente existe (E-) e um espaço físico geográfico (G), caracterizando a Dualidade de Hutchinson (COLWELL; RANGEL, 2009). Dessa forma, os dois espaços E e G) podem ser sobrepostos, sendo que um ponto no espaço ‘E’ pode refletir uma gama de variáveis ambientais que moldam o nicho n-dimensional de uma espécie. Esse mesmo ponto, quando sobreposto a ‘G’, pode corresponder às áreas geográficas com as condições propícias para a ocorrência da espécie, refletindo assim as várias localidades em ‘G’ onde o nicho da espécie existe (E).

A *modelagem de nicho ecológico* (chamado de ENM – *Ecological Niche Modeling*) (PETERSON; SOBERÓN, 2012) é conceitualmente baseada nas teorias de nicho ecológico e seus sucessores. Ela é sustentada pelas informações que conhecemos sobre a espécie (tendo como base a sua ocorrência geográfica), pelas variáveis ambientais preditoras (o nicho), e pelos algoritmos matemáticos para a modelagem de nicho e predição de ocorrência da espécie em determinada área geográfica pré-definida. Ferramentas estatísticas de ENM utilizam algoritmos matemáticos para caracterizar o nicho de uma espécie em função de variáveis ambientais. Após a caracterização, o modelo é aplicado à extensão geográfica de interesse para se encontrar as possíveis áreas de distribuição da espécie com base na adequabilidade ambiental dessas áreas. Neste contexto, poucos são os trabalhos de ENM que utilizam variáveis bionômicas, como definido por Elton (1927), para a modelagem de nicho, logo que em largas extensões geográficas ocorrem mudanças drásticas na maneira em que a espécie interage com outras no ambiente.

Com a criação de uma função matemática para caracterizar o nicho da espécie torna-se possível predizer a sua distribuição ao longo de uma determinada região geográfica pré-estabelecida, ou ainda, sobrepor ‘E’ em ‘G’. Por conseguinte, existem diversas técnicas de *modelagem de nicho* e *distribuição de espécie* tendo como base algoritmos matemáticos.

As técnicas de ENM’s podem se dividir em três tipos: 1) as que utilizam somente a presença da espécie conhecida para definir as variáveis que predizem o seu nicho; 2) as que utilizam a presença conhecida da espécie e pseudo-ausências (locais que não sabemos se a espécie ocorre) para a criação do modelo; e 3) técnicas que utilizam tanto as presenças conhecidas, como as ausências. Os dois primeiros são mais utilizados em função da dificuldade de se encontrar áreas exatas de ausência de espécies no espaço geográfico (LIMA-RIBEIRO; DINIZ-FILHO, 2013).

Os algoritmos de presença somente utilizam métricas de envelope (BIOCLIM) e de distância ambiental (como a distância de Mahalanobis e Gower). No primeiro caso, as métricas de envelope assumem total independência entre a influência das variáveis ambientais sobre as espécies e estabelece um envelope retilíneo que delimita as condições ambientais adequadas à sobrevivência das espécies. As distâncias ambientais, por outro lado, assumem a existência de um “ótimo” ecológico para a sobrevivência de cada espécie e o determinam a partir do centroide das condições ambientais relacionadas aos pontos de ocorrência conhecidos da espécie. Essas distâncias são interpretadas como índices de similaridade das variáveis locais onde a espécie é conhecida, colocando-as em função de outros locais onde não sabemos se a espécie é presente, ou não (LIMA-RIBEIRO; DINIZ-FILHO, 2013).

Os algoritmos que utilizam pseudo-ausências são mais complexos e computacionalmente extensivos quando comparados aos de presença somente. As chamadas pseudo-ausências são informações ambientais extraídas de pontos geralmente amostrados aleatoriamente da região geográfica de interesse (chamados de *background*), representando condições ambientais distintas quando comparadas àquelas onde a espécie é conhecida. Essas pseudo-ausências não indicam, por definição, que o ambiente é realmente inadequado à sobrevivência das espécies modeladas, como é assumido com os dados reais de ausência. Técnicas de modelagem com pseudo-ausências também tendem a apresentar regiões com adequabilidade ambiental mais restrita quando comparada a modelos de somente presença (LIMA-RIBEIRO; DINIZ-FILHO, 2012).

Um método de pseudo-ausências bastante utilizado é o baseado no conceito de máxima entropia, implementado pelo algoritmo Maxent. O método estima a adequabilidade ambiental para a presença de uma espécie em distribuição uniforme sob a restrição de que os valores ambientais estejam de acordo com os valores empíricos observados nos pontos de ocorrência (MARCO-JÚNIOR; SIQUEIRA, 2009), sendo este um dos métodos de modelagem mais utilizado na literatura.

Outro algoritmo que tem ganhado espaço nos trabalhos de modelagem de nicho é o SVM (*Suport Vector Machine* - Máquinas de vetores de suporte), que se caracteriza por ser um conjunto de métodos de aprendizagem supervisionados e relacionados que pertencem à família de classificadores lineares generalizados. Marco-Júnior & Siqueira (2009) colocam o algoritmo como um dos mais interessantes no momento de construção de ENM’s pelo fato de que essa metodologia minimiza o risco empírico relacionado aos dados, otimizando o desempenho mesmo em situações em que os dados de entrada são duvidosos.

Dados de ocorrências conhecidos das espécies utilizados durante a modelagem são chamados de dados de ‘calibração’, ou ‘treino’. Uma próxima etapa neste tipo de estudo é a introdução de novos dados de ocorrência de espécies chamados de ‘testes’. Estas novas ocorrências são sobrepostas ao mapa modelo de distribuição gerado e visam verificar se essas ocorrências caem em locais preditos para a presença da espécie pelo modelo criado. Como em muitos casos a amostragem de novos dados é inviável, assim como a partição dos dados originais em função de um “n” relativamente baixo (n<100), técnicas de partição de dados para avaliação podem ser empregadas. Essas técnicas dividem os dados de ocorrência em treino e teste de maneira aleatória, com a criação de um número variado de réplicas nos algoritmos. Assim, dados de treino podem ser utilizados como teste em algumas réplicas, e vice-versa (PETERSON et al., 2011).

Com a partição dos dados para teste e sobreposição desses pontos no mapa modelo final, torna-se então possível calcular alguns índices de avaliação do modelo. Os índices mais utilizados na literatura para avaliação de modelos são o índice AUC, a sensibilidade (% de acertos do modelo) e o índice TSS (ACHO QUE VALERIA UMA REFERÊNCIA AQUI...). Os dois últimos são dependentes do limiar de decisão (ou *threshold*), que binariza as réplicas geradas em áreas adequadas para a espécie (acima de um valor de adequabilidade ambiental), e áreas inadequadas para a espécie (abaixo do valor de adequabilidade ambiental). Inúmeras técnicas de decisão de limiar são encontradas na literatura hoje.

Sendo assim, uma vez que estas ferramentas fornecem auxílio no âmbito de prever áreas ambientalmente adequadas à distribuição de espécies, estas serão utilizadas para encontrar possíveis áreas de ocorrência de *L. obliqua* no Brasil.

2. Objetivo geral

Essa pesquisa se propõe a conhecer a distribuição potencial da *L. obliqua* no Brasil, bem como caracterizar as variáveis cenopoéticas relacionadas ao nicho fundamental da espécie. Também realizamos uma descrição de variáveis categóricas para *uso e ocupação da terra no Brasil*, a fim de contribuir com as hipóteses que relacionam a ocupação humana e seus impactos com os acidentes lonômicos.

2.1 Objetivos específicos

- Estimar as variáveis cenopoéticas relacionadas ao nicho fundamental de *L. obliqua*.
- Utilizar algoritmos de modelagem de nicho e distribuição de espécies para encontrar as áreas ambientalmente adequáveis para a presença de *L. obliqua* nos biomas em que a espécie já foi amostrada no Brasil.
- Apresentar um descritivo das variáveis descritoras de parte do nicho fundamental recortadas dentro da área predita.
- Apresentar um descritivo dos tipos de categorias de uso da terra recortadas dentro da área predita.

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4. Capítulo 1:

Condições ecológicas e potenciais áreas de ocorrência de *Lonomia obliqua* Walker 1855 no Brasil

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Potential occurrence areas and ecological conditions of *Lonomia obliqua* Walker 1855 (Saturniidae: Hemileucinae) in Brazil

Abstract

Lonomia obliqua Walker 1855 (Lepidoptera: Saturniidae) is a species of moth whose larvae are responsible for the lonomism, a form of envenomation that has been occurring in Brazil since the 1980s. The knowledge about spatial distribution and some ecological aspects of this species is still very incomplete due to the lack of studies. In this regard, different Ecological Niche Modeling (ENM) methods have been tested for constructing a model map for the potential distribution of this species in Brazil. According to the selected ENM model mapping, the adjusted range for L. obliqua occurs at latitudes between 12°-32° and longitudes between 57°-39°. In addition, we found that the environment considered suitable fundamental niche for the specie is characterized by warmer summers with higher rainfall index, and winters with lower temperatures and rainfall index. Although the specie is associated with savanna, deciduous forest and ombrophilous forest, great part of the predicted area was also found to be characterized by agriculture and silviculture. On the whole, the map and information obtained herein may help as a tool for Brazilian public health agencies to appropriately direct preventive strategies and antivenom availability to those places where people are at high risk of lonomism. This study also provides an addendum on the habitat loss for L. obliqua, suggesting that conservation actions need to be implemented for this species.

Keywords: Caterpillar; ecological niche modeling; venomous insect.

1. Introduction

Lonomia obliqua is a species of Lepidoptera whose larval stage (caterpillar) is of broad medical interest due to its role as an etiological agent of lonomism, a form of envenomation caused by the human contact with the stinging structures of this species stage. This envenomation is frequently characterized by systemic hemorrhage, which can reach vital organs and lead the patient to death (Chudzinski-Tavassi and Alvarez-Flores, 2013).

The first reports of official accidents with *L. obliqua* in Brazil started to be documented in the late 1980s, in southern Brazil, when the lonomism began to reach alarming epidemiological proportions (Duarte et al., 1990). Nowadays, Rio Grande do Sul (Lorini, 2008), Santa Catarina (Zannin et al., 2003) and Paraná (Garcia and

Danni-Oliveira, 2007; Rubio, 2001) are the states with the highest proportion of cases, following, with minor cases, São Paulo (Chudzinski-Tavassi and Alvarez-Flores, 2013) and Minas Gerais (Cerbino et al., 2004; Jader, 2007). Up to the present date thousands of lonomism cases have been recorded in the country (Chudzinski-Tavassi and Alvarez-Flores, 2013), but it is worth mention that, for the authors, the accidents are underreported in Brazil, especially in agricultural areas, not having an accurate record of the number of deaths in the country.

Due to the severity of envenoming, Brazilian sanitary authorities - represented by the Butantan Institute - started the production of *L. obliqua* antivenom, which has been used since 1996 and it is considered the sole antivenom available worldwide for caterpillar envenomation, avoiding complications observed in most severe cases, and the patients' death consequently (Dias da Silva et al., 1996). Despite the availability of treatment in Brazil, mortality rates continue to occur, mainly due to the victims' delay to seek medical treatment (Moraes, 2009), or even the lack of knowledge by health professionals to make the diagnosis (Moraes, 2009; SVS, 2009).

There are few studies on the geographical distribution and ecological aspects of *L. obliqua* in the literature. In a review study on the subfamily Hemileucinae (Saturniidae), Lemaire (2002) presented the distribution of *L. obliqua* in Brazil between Rio Grande do Sul and Bahia, a geographical area almost entirely occupied by the Atlantic Forest biome. Although the author presents some occurrence areas for the species in this geographic space, the potential occupation of the species in Brazil is unknown, which makes this information of paramount importance to provide subsidies in terms of public health. Other studies in the literature are related with local climatic data, in attempt to characterize the environmental variables associated with the occurrence of lonomism (e.g. Gamborgi et al., 2012; Garcia, 2013).

Lonomia obliqua has also undergone an expansion in its distribution area (e.g. recent cases of lonomism in the province of Misiones - Argentina - Sánchez et al., 2015). It is suspected that this insect has moved from its natural habitat, the primary forest, to other regions that also involve arboreal hosts, with emphasis on areas with commercial plant species (Lemaire, 2002). Hypothesis has related this recent distribution to the loss of native vegetation due to the anthropic occupation. With this displacement, there has been a decrease in physical space between *L. obliqua* and human beings, which can be contributing to more cases of lonomism.

In this context, the present study aimed to: a) model the geographical distribution of *L. obliqua* in Brazil with presentation of the species environmental suitability map; b) perform a description of the variables that compose part of the species fundamental niche extracted from the predicted area; and c) perform a description of the land-use classes for the predicted area in order to contribute to the hypothesis that relates species distribution and lonomic accidents with anthropic occupation.

2. Material and methods

For the creation of a model map showing the distribution of *L. obliqua* in Brazil, Ecological Niche Modeling (ENM) methods were used. These methods estimate associations among environmental variables and known points of species occurrence, to infer under which conditions its population can survive (Peterson 2014). From this, values of environmental suitability are traced in areas where the species occurrence is unknown (Peterson 2014). This study aimed to assess a data survey to indicate the occurrence points of *L. obliqua* for the construction of an ENM for the species in Brazil; with the subsequent description of scenopoetic variables as well as occupation and land–use variables. A description of the methodology is summarized in Figure 1.

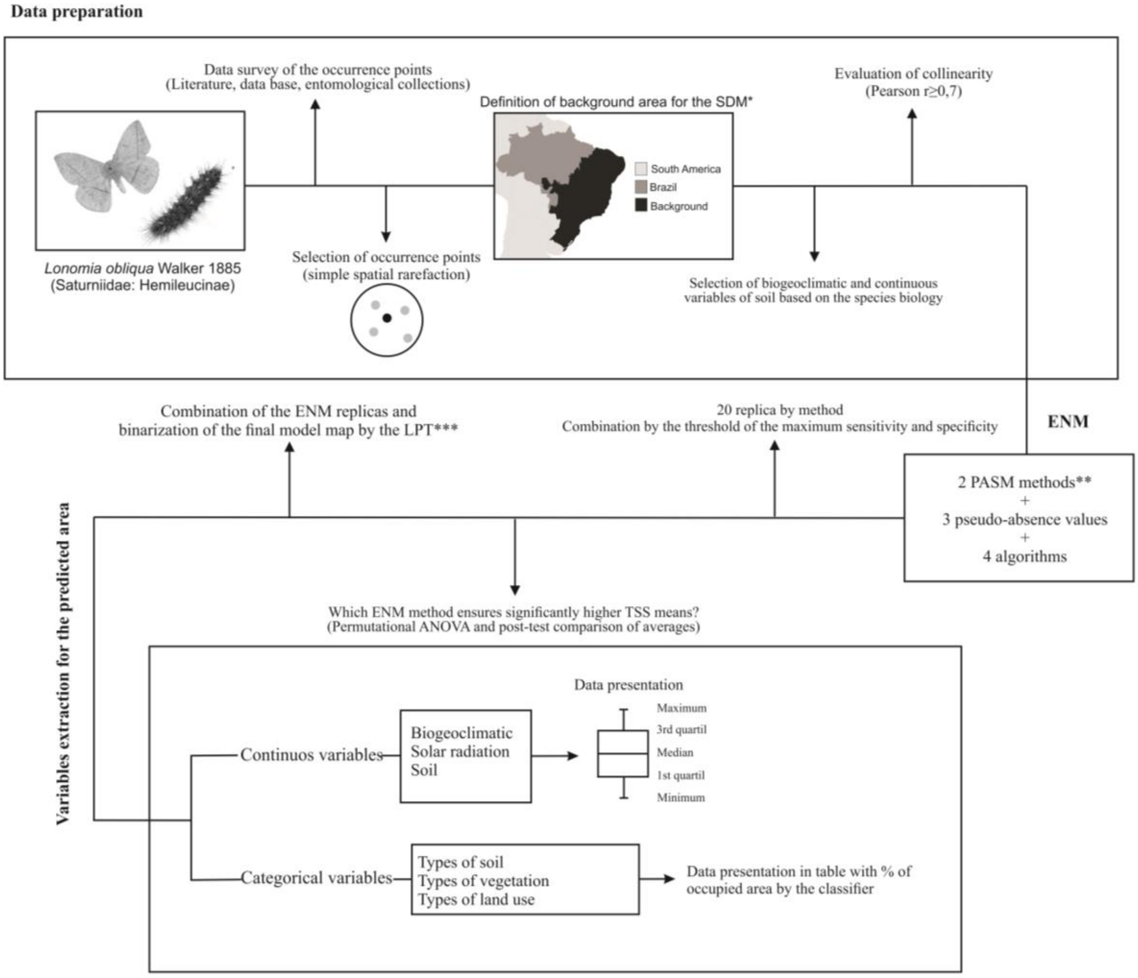

Figure 1 - Flowchart summarizing the methodology used in the present study.

2.1. Occurrence points of L. obliqua

The occurrences points of *L. obliqua* were consulted on data available at online databases (Global Biodiversity Information Facility - GBIF: <http://www.gbif.org>; SpeciesLink: <http: //splink.cria.org.br>), in technical-scientific literature, and through entomological collections of Embrapa Cerrado and the National Institute of Tropical Medicine of Argentina (INMeT) (Appendix A).

In order to select the *L. obliqua* occurrence points, the following inclusion criteria were considered: a) to include a geographical coordinate location, or address of the place, park or region of the city where the specimen was sampled; b) to be included in the period between 1990 and 2017, due to the fact that official notifications of accidents with the species started only in 1989 (Duarte et al. 1990) in cases of accident data. From these definitions, 96 occurrence points of *L. obliqua* were selected, ranging from 1994 to 2017.

The occurrence points were filtered to obtain a single point in a radius of 2.5 arc-minutes (~5 km). In this analysis, a ray (or buffer) was drawn around the occurrence points, selecting only one point when two or more than two were found within the same space. The objective of the analysis was to avoid the redundancy of environmental information in the model, which may generate a subprediction of the potential distribution of the species.

2.2. Environmental Data

After selecting the points, the environmental variables for the ENM were selected. The first environmental variables selected were biogeoclimatic from the WorldClim 2 database (<http://www.worldclim.org/> - Fick and Hijmans, 2017) (Table 1). Only those that presented mean quarter and/or annual results were considered. The selection of variables in the quarters is based on the species biological cycle, which passes through the caterpillar stage in warmer months, and the pupa stage in the colder months of the year (e. g. Garcia, 2013; Garcia and Danni-Oliveira, 2007; Salomão De Azevedo, 2011). Thus, temperature and rainfall variables that met these criteria were observed. From the same database, solar radiation was also considered (table 1). This variable can influence not only on *L. obliqua* host plants, but also on its larval gregarious behavior, as shown in studies with other Saturniidae species (Klok and Chown, 1999). The other niche variables considered in this study were the continuous soil variables available at the SoilGrids database (<http://soilgrids.org/>)

(Table 1). These variables were selected due to the fact that the pupal phase of *L. obliqua* occurrs under plant litter (Lorini, 1999) and even under soil (unpublished data).

Database	Variables	Abbreviation
WordClim	Annual Mean Temperature (TºC)	AMT
	Mean Temperature of Warmest Quarter (TºC)	MTWQ
	Mean Temperature of Coldest Quarter (TºC)	MTCQ
	Annual precipitation (mm)	AP
	Precipitation of Warmest Quarter (mm	PWQ
	Precipitation of Coldest Quarter (mm	PCQ
	Solar radiation (kJ m-2 day-1)	SR
SoilGrids	Bulk density (fine earth) in Kg/m^3	BLDFIE
	Cation exchange capacity of soil (cmol/Kg);	CECSOL
	Clay contente (0-2µm) in mass fraction (%)	CLYPPT
	Coarse fragments volumetric in %	CRFVOL
	Soil organic carbon content (fine earth fraction) (g/Kg)	ORCDRC
	pH x 10 in H_2O	PHIHOX
	pH x 10 in KCL	PHIKCL
	Silt content (2-30µm) mass fraction (%)	SLTPPR
	Sand content (50 - 200µm) mass fraction (%)	SNDPPT

Table 1 - Abbreviation of climatic and soil variables.

All variables presented WGS 1984 projection, with pixel size of 2.5 arc-minutes (~5 km). The relatively large size of the pixel was fixed as a function of some presence data being related to parks, covering areas larger than 30 arc-seconds (~1 km) and smaller than 5 arc-minutes (~10 km).

2.3. Selection of environmental data

Using an excessive number of points in the same locality can promote obliquities in an ENM, as well as using an excessive number of variables can also promote an overadjustment in the model, oversizing the niche and causing the modeling algorithms to fail in finding new places for the species occurrence (Peterson, 2014). Thus, to avoid overadjustment in the ENM, the niche predictors' variables were submitted to a previous selection from the Pearson correlation coefficient analysis. Collinear variables were regarded as having an $r \geq 0.70$ among them, excluding those with an excessive number of associations with other variables.

2.4. Study area

The modeling area (or background) was related to the Pampa, Atlantic Forest, Caatinga and Cerrado biomes. This geographical extension was based on the occurrence points of the species. As *L. obliqua* has a winged life stage, even if short (approximately 7 days according to Lorini, 1999), the main barriers to the species dispersal are related to environmental conditions. Besides, no historical occurrences were found outside these biomes in the databases investigated in this work. Thus, the criteria related to dispersion and historical occurrences were considered for the background definition (Barve et al., 2011).

2.5. ENM

Currently there are various ENM algorithms available for the prediction of environmentally suitable areas for the species distribution, and the combined use of these several algorithms (or ensemble) tends to increase the models’ reliability, therefore considering a wide range of species distribution patterns (Araújo and New, 2007). In addition, there is no consensus on which modeling methodologies present the best adjustment (Peterson and Soberon, 2012; Qiao et al., 2015). For this reason, the present study tested different modeling methodologies in order to find the one that best represents the distribution of *L. obliqua* for the selected geographical area.

Four algorithms were used based on different modeling methods. The Domain (or Gower distance) (Carpenter et al., 1993) and Mahalanobis (Farber and Kadmon,

2003) methods are based on environmental distances and the presence of species only. The Maxent algorithm (Phillips et al., 2004) and Support Vector Machine (SVM) (Tax and Duin, 2004) are models based on mechanical learning and records of presence and pseudo-absence.

We tested two pseudo-absences sampling methodologies (PASM): 1) randomly in the background, excluding the known occurrence points in the studied area; and 2) those found in environmentally different areas of the species occurrence sites defined by the modeling through BIOCLIM algorithm (bioclimatic envelope) (Nix, 1986). The first methodology is one of the most utilized for ENM (Iturbide et al., 2015) and considers that absence points can occur in any area of the background. The second methodology draws an "envelope" around the values of environmental variables based on what is already known about the species, considering that the locations with variable values outside this "envelope" are the ones that present the greatest possibility of showing a low environmental suitability (Barbet-Massin et al., 2012). With the definition of the envelope, random sampling of the pseudo-absence points was performed.

We also tested three different sample sizes of pseudo-absences: equal to the number of presences, 10 times the number of presences and 100 times the number of presences. We tested different numbers of pseudo-absences in order to find a value that would result in better accuracy for the species distribution in the machine-learning model. The environmental distance models, Gower and Mahalanobis, do not use pseudo-absences when constructing the niche model, since this information is utilized only for the construction of the evaluation index.

During the modeling, the occurrence data were partitioned into two subsets, with 73.68% of the points used in the models’ training and 26.32% used for the test. These percentages were selected to use discrete frequencies of presence records during modeling. Since training and testing are subsets of the same occurrence points, both were randomized 20 times in order to minimize the spatial structure among the data sets, thus providing less oblique assessments. These points randomly collected for the test in each replica were then compared to the generated training models, followed by the calculation of the True Skill Statistic (TSS) index as suggested by Allouche et al. (2006).

The TSS index is threshold dependent. The threshold is a cut in the predicted values of environmental suitability in each replica of generated model, classifying the areas for the species distribution as environmentally adequate (1) and environmentally inadequate (0). Based on the recommendations of Liu et al. (2016, 2013), the threshold was determined by the maximum sensitivity and specificity to transform the continuous maps into binary systems.

The TSS index shows a variation between -1 and 1, considering that close to zero or negative values indicate that the forecasts are not different from a randomly generated model, whereas forecasts with values closer to 1 are considered excellent to define the species distribution (Allouche et al., 2006). This index is calculated based on the components of the standard confusion matrix that represents the correspondence and mismatches between observations and predictions (Eq.1):

TSS = Sensitivity + Specificity - 1 (Equation 1)

In which the sensitivity is considered the relative frequency of presence hits while the specificity corresponds to the relative frequency of the pseudo-absence hits in the test.

In order to compare the different ENM methodologies and choose the one that results in TSS index values closer to +1, a Variance Analysis (ANOVA) with 999 permutations was performed, followed by the test of mean’s clustering, via Bootstrap, with 1000 permutations (Ramos and Ferreira, 2009). The independent variables considered for the tests were: 1) Pseudo-Absence Sampling Method (PASM); 2) Number of pseudo-absences; and 3) Modeling algorithms.

With the selection of the sampling methodology and the number of pseudo-absences that promoted the best models in accordance with all the used algorithms, the replicas of each algorithm were combined based on the methodology proposed by Araújo and New (2007). Each replica, already binarized by the maximum sensitivity and specificity threshold, was combined with a final mean result. Values close to 1 represented the most suitable environment for the species in accordance with all replicas of ENMs.

For the construction of the final *L. obliqua* distribution model, the TSS index was presented with the average ± standard deviation. This map was cut only for the background area within Brazil, despite the use of species occurrence data from the province of Misiones - Argentina. In this last model, the data were dichotomized by the Lowest Presence Threshold (LPT), which represents the minimum value predicted for the training sites.

The final distribution model was evaluated using data provided by the Entomological Collection of the University of Passo Fundo (CEUPF), in Rio Grande do Sul, which performs sampling of *L. obliqua* larvae for breeding of this species in lab conditions and venom preparation. As these data are only concerning to the municipalities of the sampled specimens, we conducted a qualitative evaluation to verify if the predicted zones by the distribution map were coincident with the cities where *L. obliqua* had been sampled.

2.6. Predictor variables of the ecological niche

In order to draw a distribution map, it was previously necessary to develop a mathematical model for the species niche regardless of the algorithm used. The model is subsequently applied to the bottom area for the classification of environmental suitability. In the present study, the presentation of such model is impracticable, since different mathematical algorithms were combined in order to create the distribution map. Thus, to obtain information about the variation of the variables that characterize part of the *L. obliqua* niche, the variation of all descriptors previously considered for the predicted area was extracted and their information presented through descriptive statistics (quartiles).

Variable categories were also extracted from the predicted area: the percentage of vegetation classes for Brazil (<www.ibge.com.br>), and the percentages of land-use types, referring to the year 2000 and obtained from the IBGE database (<www.ibge.com.br>). These variables were considered with the purpose of contributing with biological information and also with the hypotheses that relate lonomic accidents with the use and occupation of the land.

2.7. Software

Simple spatial rarefaction was performed by the ArcGis 10 program (ESRI, 2011) with the SDMtoolbox package (Brown, 2014), through selection of flat projection "Continent: South America equidistant Conic".

The statistical analyzes were implemented in software R (R Core Team, 2017). The models were generated with the packages raster (Hijmans, 2016), rgdal (Bivand et al., 2017), dismo (Hijmans et al., 2017), kernlab (Karatzoglou et al., 2004), rJava (Urbanek, 2016) and vegan (Jari Oksanen et al., 2017). For the permutational ANOVA, the vegan package was used, and for the mean comparison test via Bootstrap, the ExpDes.pt package was utilized (Ferreira et al., 2013). .

3. Results

The total of 38 occurrence points were selected for the ENM of *L. obliqua* through simple spatial rarefaction (Figure 2). The collinearity of the environmental variables was also evaluated (Table 2): MTWQ, PCQ, Radiation, CECSOL, CLYPPT, CEFVOL, ORCDRC and PHIHOX.

From such selections, the bioclimatic envelope model was generated (Figure 2). After the envelope definition, 480 model replicas were generated for *L. obliqua* distribution (2 PASM * 3 pseudo-absence values * 4 algorithms * 20 replicas) and then compared to the TSS index results for each methodological combination (Appendix B).

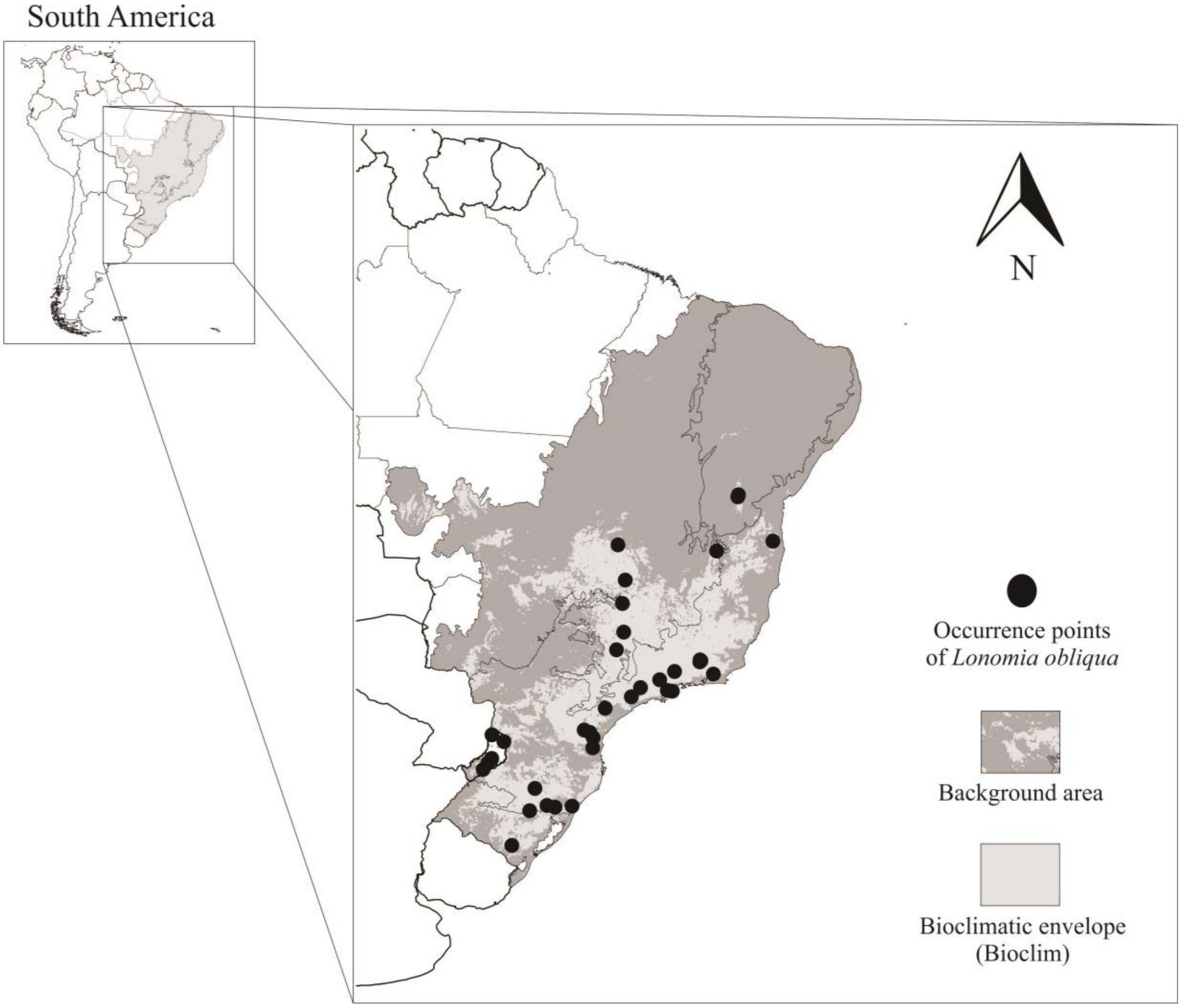

Figure 2 - Background area selected for the ENM, with respective occurrence points of *L. obliqua* and suitable area for the species according to the bioclimatic envelope.

	Climate							Soil							
	AMT	**MTWQ**	MTCQ	AP	**PWQ**	**PCQ**	**SR**	BLDFIE	**CECSOL**	CLYPPT	**CRFVOL**	**ORCDRC**	**PHIHOX**	PHIKCL	SLTPPT
MTWQ	**0.93**														
MTCQ	**0.98**	**0.86**													
AP	-0.29	-0.24	-0.29												
PWQ	-0.52	-0.54	-0.54	0.61											
PCQ	-0.18	-0.01	-0.21	0.34	-0.14										
SR	0.66	0.59	0.66	-0.61	**-0.75**	-0.11									
BLDFIE	**0.74**	0.65	**0.72**	-0.6	-0.51	-0.33	**0.72**								
CECSOL	-0.58	-0.41	-0.61	0.06	0.02	0.42	-0.27	-0.61							
CLYPPT	**-0.74**	-0.69	**-0.72**	0.39	0.46	0.19	-0.55	**-0.74**	0.56						
CRFVOL	-0.11	-0.07	-0.11	-0.35	-0.25	0.12	0.2	-0.03	0.37	0.01					
ORCDRC	**-0.72**	-0.59	**-0.72**	0.49	0.38	0.4	-0.6	**-0.87**	0.69	0.68	0.12				
PHIHOX	0.3	0.31	0.25	**-0.77**	-0.51	-0.17	0.63	0.56	0.1	-0.3	0.4	-0.44			
PHIKCL	0.33	0.28	0.3	**-0.72**	-0.4	-0.3	0.57	0.59	-0.01	-0.27	0.32	-0.52	**0.88**		
SLTPPT	-0.42	-0.24	-0.44	0.13	-0.12	0.47	-0.13	-0.48	**0.72**	0.46	0.22	0.59	0.07	-0.12	
SNDPPT	**0.73**	0.62	**0.72**	-0.34	-0.3	-0.33	0.46	**0.75**	**-0.71**	**-0.93**	-0.1	**-0.75**	0.2	0.25	**-0.75**

Table 2 – Pearson's correlation matrix among continuous environmental variables for use in ENM (**r ≥ 0.7**).

The following methods had significant effects on the TSS index values: PASM variables (F_1=146.23; p=0.001), number of pseudo-absences (F_1=12.09; p=0.002), modeling algorithms (F_3=69.51; p=0.001), interactions between PASM and number of pseudo-absences (F_1=5.74; p=0.016), PASM and algorithms ($F_{0.34}$=8.78; p=0.001) and number of pseudo-absences and algorithms (F_3=10.55; p=0.001).

From these results, it was possible to verify that regardless the number of pseudo-absences or algorithms, the PASM by the bioclimatic envelope guaranteed higher TSS index means when compared to the random sampling method (Figure 3A and 3B). In relation to the number of pseudo-absences and algorithms, the TSS index means were significantly higher when a similar number of attendances were used (n=38) (Figure 3C). The difference was also observed in the value of TSS index means within the algorithms, with value reduction after increasing the number of pseudo-absences in SVM (Figure 3C).

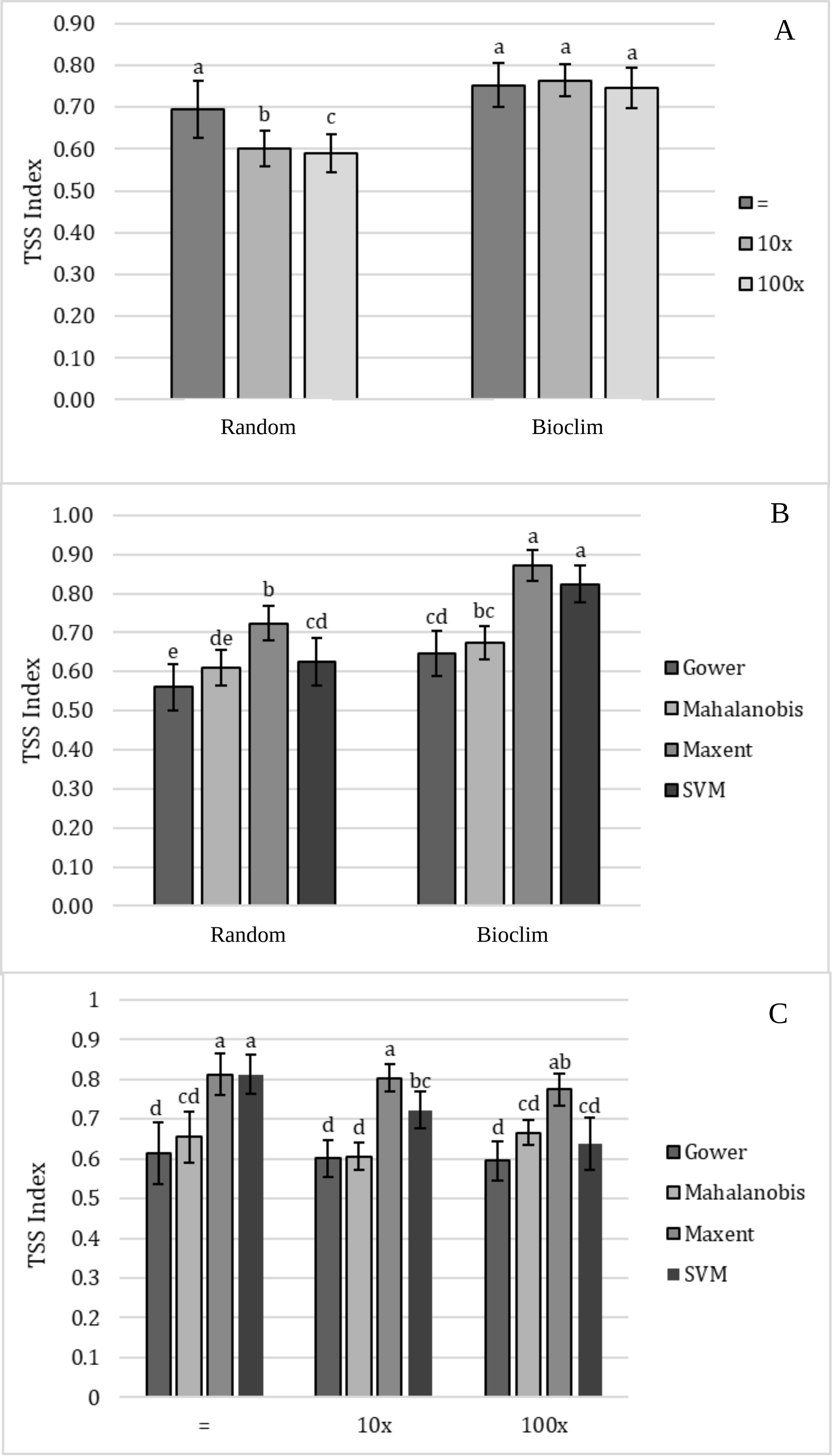

Figure 3 - TSS index values for comparison among the ENM methodologies from the Bootstrap method (1000 randomizations). A – PASM vs. Number of pseudo-absences; B – PASM vs. Algorithms; C – Number of pseudo-absences vs. Algorithms. The averages of TSS index are classified as “a” (higher values) to “d” (lower values).

Thus, the final model for the distribution of *L. obliqua* followed the criteria: PASM methodology outside the bioclimatic envelope, number of pseudo-absences equals to the presences (n=38) and a combination of replicas for all modeling algorithms (Figure 4A). The resulting model map was then binarized by the LPT. The final TSS index of this distribution model was 0.7525±0.1118.

The final model was evaluated through a qualitative analysis with *L. obliqua* occurrence data in cities of Rio Grande do Sul, verifying that the model predicted the species occurrence areas in all municipalities sampled by the Entomology Laboratory of the University of Passo Fundo (Figure 4B).

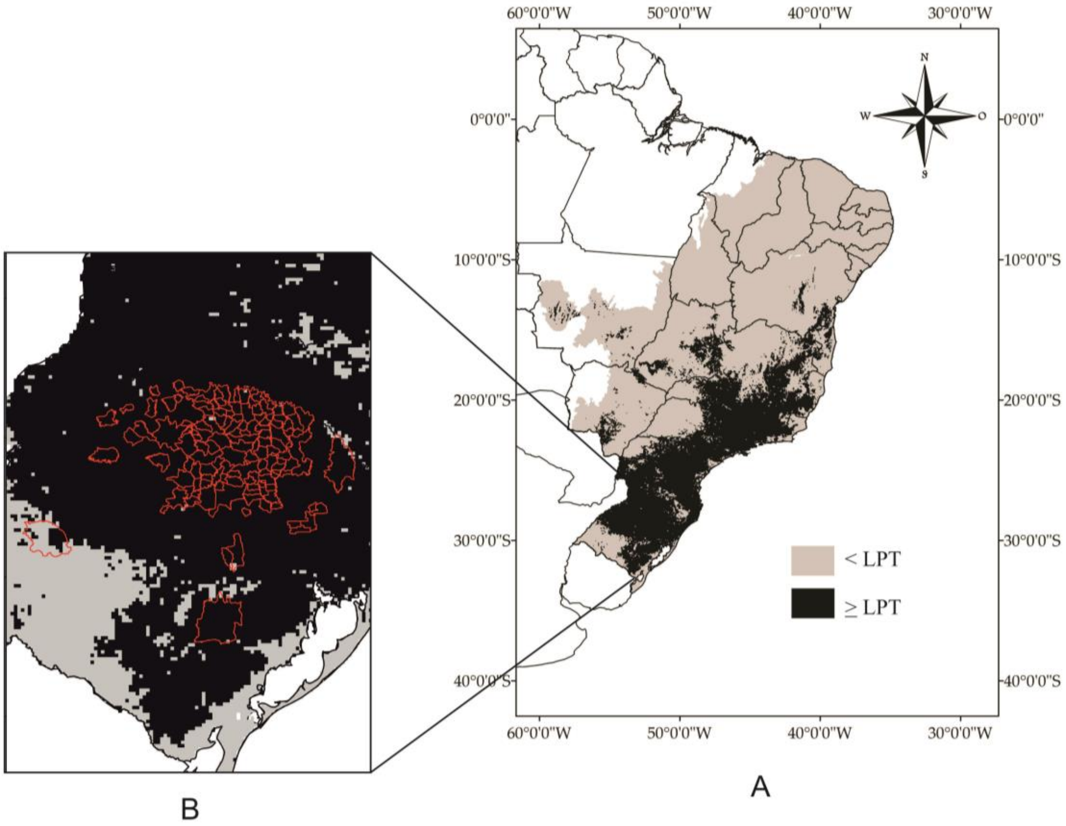

Figure 4 – A) ENM map predicting the distribution of *L. obliqua* in Brazil binarized by the Lowest Presence Threshold (LPT); B) Municipalities of Rio Grande do Sul where individuals of *L. obliqua* were sampled (Source: CEUPF - Entomological Collection of the University of Passo Fundo).

According to the model map for the distribution of *L. obliqua* binarized by the LPT, the adjusted range for the species occurs between latitudes ~12° and ~32° and longitudes ~57° and ~39° in Brazil (figure 4). The main Brazilian states that presented environmentally suitable areas for the species occurrence were Rio Grande do Sul, Santa Catarina, Paraná, São Paulo, Minas Gerais, Rio de Janeiro and Espírito Santo. There were few predicted suitable areas for the species occurrence in the states of Bahia, Goiás, Federal District, Mato Grosso and Mato Grosso do Sul.

The variation of the climatic and soil variables extracted from the predicted area are presented in Table 3. The variables descriptors of forest types and land-use classes are presented in Table 4.

Variables		Descriptive statistics				
		Minimum	1st quartil	Median	3rd quartil	Maximum
Climatic	AMT (ºC)	11.66	18.49	19.83	21.16	24.49
	MTWQ (ºC)	14.41	21.67	22.72	23.66	26.32
	MTCQ (ºC)	8.63	14.31	16.38	18.38	23.20
	AP (mm)	606	1359	1507	1642	3124
	PWQ (mm)	194	429	492	600	1095
	PCQ (mm)	5	65	158	354	573
	SR (kJ m-2 day-1)	12798	14614	15188	15699	17483
Soil	BLDFIE (Kg/m^3)	842	978	1244	1286	1384
	CECSOL (cmol/Kg)	5	11	14	17	30
	CLYPPT (%)	18	33	36	42	64
	CRFVOL (%)	0	1	2	3	10
	ORCDRC (g/Kg)	14	21	29	44	103
	PHIHOX	46	51	53	55	62
	PHIKCL	39	44	46	47	55
	SLTPPR (%)	9	16	19	26	37
	SNDPPT (%)	11	35	46	50	70

Table 3 - Descriptive statistics (by quartiles) of the continuous variables extracted from the entire predicted area for *L. obliqua*.

Classes	Categories	Relative freq. (%)
Forests	Caatinga	29.46
	Semi-deciduous seasonal forest	22.51
	Mixed ombrophilous forest	14.65
	Areas of ecological tension	12.90
	Dense ombrophilous forest	11.76
	Deciduous seasonal forest	6.11
	Others	2.61
Land-use	Mosaic of forest vegetation with agricultural areas	29.58
	Mosaic of agriculture with forest remnants	17.51
	Agricultural area	12.72
	Planted grassland	8.12
	Silviculture	6.97
	Natural grassland	6.67
	Forest vegetation	6.62
	Others	11.81

Table 4 - Relative frequency (%) of vegetation and land use categories for the predicted area for *L. obliqua*.

4. Discussion

An ecological niche model (ENM) was developed in this study to estimate the distribution of *L. obliqua* in Brazil. For the calibration and evaluation of the model, we used data available in online databases, provided by entomological collections and institutions linked to public health. In order to find the one that resulted in better TSS scores, we tested different niche modeling techniques. Thus, besides providing a model map for the potential distribution of L. obliqua in Brazil, we also contributed with data related to its fundamental niche, which is of relevance regarding the ecological process of the species dispersion and eventually the occurrence of lonomism.

In relation to ENM methods, a series of studies have established that the pseudo-absence selection method directly affects the performance of models (e.g. Hertzog et al., 2014; Iturbide et al., 2015; Senay et al., 2013; Wisz and Guisan, 2009). In this study the pseudo-absences sampled via bioclimatic envelope resulted in the best TSS index values. The pseudo-absences sampled randomly from the background area may increase the risk of including adequate environments for the species occurrence as a pseudo-absence, underestimating the fundamental niche and species distribution (Anderson and Raza, 2010). In contrast, it is easier to classify the actual presences correctly (expressed in the sensitivity index) and the actual pseudo-absences (expressed in the specificity index) when they have already been defined in relevant scenopoetic conditions, as happened with the bioclimatic envelope.

Several studies have shown that the model type is an important source of uncertainties in the results, as well as the selection of variables, collinearity of data and selection of pseudo-absences (e.g. Guisan and Zimmermann, 2000; Qiao et al., 2015). Considering the parsimony method, we chose the replicas with only 38 pseudo-absences points, utilizing the BIOCLIM methodology. In this way, a simpler and faster execution methodology – with also a high TSS index value - was used to develop the model.

The model-map obtained in this study shows a distribution area for *L. obliqua* relatively broad as proposed by Lemaire (2002), which restricts the species to the regions of Atlantic Forest. An explanation for finding new areas of occurrence by ENM can be translated as commission errors in the models (or "overestimation"). However, both commission and omission errors (or failures to hit occurrence points taken for the evaluation of the model) can be minimized according to the choice of the threshold decision, which in this study was the criteria of maximum sensitivity and specificity for the replicas’ combination, supporting more accurate predictions. Besides, even when using external data, such as new occurrences of *L. obliqua* in municipalities of Rio Grande do Sul state, the selected ENM model-map was able to satisfactorily predict suitable areas within such municipalities.

A second stage of this study aimed to describe the abiotic conditions present in the entire area predicted as suitable for *L. obliqua* in Brazil. This shows higher temperatures in the warmest quarter (minimum= 14.41ºC, maximum= 26.32ºC, median= 22.72ºC) and lower temperatures in the coldest quarter (minimum= 8.63ºC, maximum= 23.20ºC, median= 16.38ºC) in relation to the annual values (minimum= 11.66ºC, maximum= 24.49ºC, median= 19.83ºC). Regarding the temperature values, those corresponding to the warmer months – in which occurs the development of the larval stage of *L. obliqua* - are approximately equal to the values considered ideal for the development of most insects (mean~25ºC, minimum=15ºC, maximum=38ºC) (Rodrigues, 2004). In addition, the temperature values used for the breeding of *L. obliqua* larvae under controlled environments (25±1ºC - Lorini et al., 2007; 18.6ºC - Lorini et al., 2004) are within the minimum and maximum range reported herein Futhermore, this everage variation is similar to that reported by some ecoepidemiological studies conducted in southern Brazil. Garcia (2013) showed a mean temperature variation between 20ºC and 25ºC, whereas Gamborgi et al. (2012) calculated an range of temperature mean between 21ºC and 34.6ºC. However, these areas are restricted to a few localities, and the greatest temperature variation observed is related to spatial and temporal amplitude.

Precipitation presents higher values in the warmest quarter (minimum= 194mm, maximum= 1095mm, median= 492mm) than in the coldest quarter (minimum= 5mm, maximum= 573mm, median= 158mm), but both periods presented values below the annual precipitation (minimum= 606mm, maximum= 3124mm, median= 1507mm). Garcia (2013) showed values between 1500 mm and 2000 mm annually for the southern region of Brazil in months with the greatest notification of lonomic accidents. These values are in line with the predicted annual precipitation values obtained from the model mapping in this study.

About solar radiation and soil variables investigated here, it is important to notice that there are no comparative parameters in published studies for *L. obliqua*.

Therefore, this study constitutes the first description of these data variation in possible areas for the occurrence of this species. Noteworthy, we selected the solar radiation as a variable that could affect *L. obliqua* niche since Klok and Chown (1999) showed that it has influence on the body thermoregulation of larvae of *Imbrasia belina*, which also belongs to the family Saturniidae and subfamily Hemileucinae. Furthermore, it is widely known the influence of solar radiation on plant species, and since *L. obliqua* larvae are dependent on arboreal hosts, they may be indirectly influenced by this variable. The median value of solar radiation obtained in this study for the predicted area is close to the mean value described for the Brazilian territory (14.795-24.658 Kj m^{-2} day^{-1}) (Pereira et al., 2006).

The region of the predicted area is characterized by a large amount of clay particles (75% of the area with values ≥33% of clay in the mass fraction) and by having less than 70% and 37% of sand and silt, respectively. According to Embrapa (2006), with such compositions, much of the area predicted can be classified as clayey soil. This type of soil tends to present a smaller number of coarse fragments (EMBRAPA, 2006), coincidentally, these were detected in less than 10% in the predicted area. Furthermore, higher values of cation exchange capacity and remarkably more acidic pH values, characteristics of clay soils (Instituto da Potassa & Fosfato, 1998), were also identified in that area.

The great amount of organic matter, which is fundamental for the high level of cation exchange capacity in clay soils, comes mainly from the nutrient cycling of vegetation in the superficial layer (Binkley and Fisher, 2013). Organic matter is essential for life in soil as a source of energy due to the release of carbon by the microbiota (Binkley and Fisher, 2013). Thus, the soil in the predicted area is associated with primary vegetation that offers organic matter to the soil, wich is common in forests like caatinga, seasonal, ombrophilous, and areas of ecological tension (ecotones).

Orderly, “forests” are defined as a thickening of tall trees that form a canopy. This feature is fundamental for the development of *L. obliqua*, which depends on host trees for oviposition and larvae development. These hosts are also responsible for providing food to the larvae, which is the only stage of the species’ life-cycle that feeds on. During the warmer periods of the year – corresponding to the larval period of *L. obliqua* –, a wide evapotranspiration of the soils occurs under the forest canopy, causing the decrease of the temperature in the soil and the increase of air humidity (Binkley and Fisher, 2013). This evapotranspiration is essentially conditioned by the soil type in forest areas, which are more porous, contributing with evaporation of water from the superficial layer to the air (Binkley and Fisher, 2013). Under the canopy, air stay warmer and higher relative humidity. Lorini et al. (2007, 2004) showed that the development of *L. obliqua* larvae under laboratory conditions is conditioned by humidity variations ranging from 62 to 80%, demonstrating the importance of humidity for the development of this species.

It is during the winter, with temperatures below 15ºC, that insects usually go into hibernation (Rodrigues, 2004), and this is coincident with the pupation period for *L. obliqua*. Precipitation decreases during this period, causing treetop reduction with greater biomass deposition on the soil (Binkley and Fisher, 2013). With the canopy opening, solar radiation tends to directly affect the soil, resulting in increase of the temperature in the superficial layer due to the absence of leaching and evapotranspiration (Binkley and Fisher, 2013). The heat loss is also low in the organic layer, which presents a low thermal conductivity, but retains the humidity required to maintain life in the soil (Binkley and Fisher, 2013).

It is likely that at the end of the rainy season, with the soil still humid, that the prepupa of *L. obliqua* excavates the substrate for the pupation. Literature reports show that this phase of life occurs under plant litter, but some unpublished statements suggest that it may occur under the soil surface layer. Under these substrates, at certain conditions of temperature and humidity, the species must continue its development, especially because the pupa does not present a protective cocoon (Lemaire, 2002). The dependence of the species on humidity, even during the pupal stage, is in accordance with the observation by Lorini (1999), who stated that it was necessary to periodically wet the pupation substrate for the closure of its life cycle in lab conditions. Garcia (2013) also mentions an increase in soil humidity due to higher rainfalls as a result of the climatic phenomenon *La niña*, with the hypothesis that this climatic conditioning has increased the number of accidents in the southern region of Brazil. Finally, at the end of the dry season, with the decrease of temperature and humidity in the soil and with the increase of these characteristics in the air, it is probable that the species reach the adult stage, moving from the substrate to the treetop of host trees in which it will reproduce and ovoposite.

In relation to the land-use pattern, there are many classes related to agricultural enterprises within the predicted area, especially on mosaic areas with forests. These results corroborate the fact that there are more cases of lonomism in agricultural environments when compared to other types of land-use patterns (SVS, 2009), as well as they support the hypothesis of an increase in the number of such accidents due to the decrease of forests remnants and the proximity of the latter to agricultural areas (Abella et al., 1999; Lemaire, 2002; Lorini, 1999).

Additionally, a significant portion of the predicted area is occupied by silviculture, which also supports the hypothesis of Lemaire (2002), which considers that *L. obliqua* would be migrating from areas of primary vegetation to areas with commercial crop tree species. Larvae of *L. obliqua* have already been found in trees of the genus *Eucalyptus* sp. (Bernardi et al., 2011), one of the commercial species most cultivated in Brazil in the twentieth century (Bacha and Barros, 2004). In 2000, Brazil became the sixth country with the largest area occupied by tree monocultures, with 5 million hectares covered by mixed ombrophilous forest and seasonal semi-deciduous forest (Bacha and Barros, 2004). Considering that both type of forests account for approximately 35% of the predicted area as being suitable for the species occurence, it is evident that the exploitation of commercial species are related to the increase of lonomism cases.

The results presented here are alarming not only from an epidemiological point of view, but also from the point of the species conservation, since *L. obliqua* has been losing its primary habitat due to anthropic impacts on the use. The loss of the closed-canopy forest and tree caatinga has operated as the main threat to the persistence of forest biotas (Dirzo and Raven, 2003), which has a direct impact on the life cycle of *L. obliqua*, causing its displacement to new environments and consequently resulting in lonomic accidents. A very clear example of the consequence that anthropic changes can bring on this health problem is the fact that *L. obliqua* had already been sampled in Rio Grande do Sul in 1932, but accidents with this species only began to be reported in the late 1980s (Lorini, 2008), about 10 years after the amplification of agricultural crops and forest extractivism in that region (Conceição, 1986).

5. Conclusion

The map generated in this study may support other researches aiming at sampling individuals of *L. obliqua* for their breeding and venom extraction. This map may also help as a tool for Brazilian public health agencies to appropriately direct preventive strategies and antivenom availability to those places where people are at high risk of Ionomism. From the predicted map, we notice that the environment considered adequate for the species occurrence – corresponding to part of its fundamental niche – is characterized by warmer summers with higher rainfall index, and winters with lower temperatures and rainfall index. The species would also be associated with caatinga, deciduous forest and ombrophilous forest. From this information, hypothesis has been made about the dependence of the species on higher temperatures and humidities, so that *L. obliqua* can complete its life cycle, passing through both the larval phase in arboreal hosts and the pupal phase under substrates with clay characteristics. In relation to the land-use pattern, this work corroborates the hypothesis that brings an increase of Ionomism cases due to the anthropic impacts related to agriculture and silviculture, mainly because great part of the predicted area is characterized by these two descriptors. Finally, this study also provides an addendum on the habitat loss for *L. obliqua*, suggesting that conservation actions need to be implemented for this species.

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7. Supplementary material

A table with all the occurrence points considered in this study is provided in Appendix A; Combinations of ENM methods can be found in Appendix B; Variations of climate and soil variables are available in Appendixes C, D and E.

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APPENDIX A

Occurrence points of *L. obliqua*

Coordinate	**Biome**	**City**	**State**	**Country**	**Sample date**	**Reference**
-12.9294, -41.3292	Cerrado	Mucugê	BA	Brazil	2012	Embrapa cerrado (email)
-12.9294, -41.3292	Cerrado	Mucugê	BA	Brazil	2012	Embrapa cerrado (email)
-12.9458, -41.3254	Cerrado	Mucugê	BA	Brazil	2012	Embrapa cerrado (email)
-13.0454, -41.3499	Cerrado	Mucugê	BA	Brazil	2012	Embrapa cerrado (email)
-13.0454, -41.3499	Cerrado	Mucugê	BA	Brazil	2012	Embrapa cerrado (email)
-15.4, -39.4833	Atlantic Forest	Camacan	BA	Brazil	2004	Embrapa cerrado (email)
-15.5833, -47.7	Cerrado	Planaltina	DF	Brazil	1997	Embrapa cerrado (email)
-15.5833, -47.7	Cerrado	Planaltina	DF	Brazil	1997	Embrapa cerrado (email)
-15.5833, -47.7	Cerrado	Planaltina	DF	Brazil	1997	Embrapa cerrado (email)
-15.6, -47.7333	Cerrado	Planaltina	DF	Brazil	2000	Embrapa cerrado (email)
-15.6, -47.7333	Cerrado	Planaltina	DF	Brazil	2006	Embrapa cerrado (email)
-15.6, -47.7333	Cerrado	Planaltina	DF	Brazil	2006	Embrapa cerrado (email)
-15.93, -42.48	Cerrado	Vereda Funda	MG	Brazil	2010	Embrapa cerrado (email)
-15.93, -42.48	Cerrado	Vereda Funda	MG	Brazil	2010	Embrapa cerrado (email)
-17.466944, -47.343056	Cerrado	Catalão	GO	Brazil	1999	Camargo and Schmidt (2009)
-18.7167, -47.5	Cerrado	Iraí de Minas	MG	Brazil	1994	Embrapa cerrado (e-mail)
-20.230000, -47.430000	Cerrado	Pedregulho	SP	Brazil	2001	GBIF (access 2017-01-15)
-21.17, -47.81	Cerrado	Ribeirão Preto	SP	Brazil	2013	species link (acess 2017-02-15)
-21.17, -47.81	Cerrado	Ribeirão Preto	SP	Brazil	2014	species link (acess 2017-02-15)
-21.17, -47.81	Cerrado	Ribeirão Preto	SP	Brazil	2015	species link (acess 2017-02-15)
-21.17, -47.81	Cerrado	Ribeirão Preto	SP	Brazil	2015	species link (acess 2017-02-15)
-21.711256, -43.362922	Atlantic Forest	Juiz de Fora	MG	Brazil	2004-2006	Almeida et al. (2013)
-21.753296, -43.316516	Atlantic Forest	Juiz de Fora	MG	Brazil	2004-2006	Almeida et al. (2013)

APPENDIX A (continuation) – Occurrence points of *L. obliqua*

Coordinate	Biome	City	State	Country	Sample date	Reference
-21.808359, -43.376280	Atlantic Forest	Juiz de Fora	MG	Brazil	2004-2006	Almeida et al. (2013)
-22.320000, -44.720000	Atlantic Forest	Serra da Mantiqueira	Between MG and RJ	Brazil	2011	GBIF (access 2017-01-15)
-22.320000, -44.720000	Atlantic Forest	Serra da Mantiqueira	Between MG and RJ	Brazil	2011	GBIF (access 2017-01-15)
-22.320000, -44.720000	Atlantic Forest	Serra da Mantiqueira	Between MG and RJ	Brazil	2011	GBIF (access 2017-01-15)
-22.320000, -44.720000	Atlantic Forest	Serra da Mantiqueira	Between MG and RJ	Brazil	2011	GBIF (access 2017-01-15)
-22.320000, -44.720000	Atlantic Forest	Serra da Mantiqueira	Between MG and RJ	Brazil	2011	GBIF (access 2017-01-15)
-22.4625, -42.6531	Atlantic Forest	Cachoeiras de Macacú	RJ	Brazil	2010	species link (access 2017-02-15)
-22.4625, -42.6531	Atlantic Forest	Cachoeiras de Macacú	RJ	Brazil	2010	species link (access 2017-02-15)
-22.766701 , -45.516701	Atlantic Forest	Campos do Jordão	SP	Brazil	2001	species link (access 2017-02-15)
-22.770000, -45.520000	Atlantic Forest	Pindamonhangaba	SP	Brazil	2001	GBIF (access 2017-01-15)
-22.770000, -45.520000	Atlantic Forest	Pindamonhangaba	SP	Brazil	2001	GBIF (access 2017-01-15)
-22.770000, -45.520000	Atlantic Forest	Pindamonhangaba	SP	Brazil	2002	GBIF (access 2017-01-15)
-23.1833 , -46.533299	Atlantic Forest	Atibaia	SP	Brazil	2002	species link (access 2017-02-15)
-23.3333 , -45.099998	Atlantic Forest	São Luís do Paraitinga	SP	Brazil	2001	species link (access 2017-02-15)
-23.366699 , -44.833301	Atlantic Forest	Ubatuba	SP	Brazil	2001	species link (access 2017-02-15)
-23.6667 , -47.016701	Atlantic Forest	Cotia	SP	Brazil	2001	species link (access 2017-02-15)
-24.270000, -48.400000	Atlantic Forest	Ribeirão Grande	SP	Brazil	2001	GBIF (access 2017-01-15)
-24.273322, -48.416936	Atlantic Forest	Iporanga	SP	Brazil	2015	Biological collection of Oswaldo Cruz Institute (access 2017-01-13)
-24.273322, -48.416937	Atlantic Forest	Iporanga	SP	Brazil	2015	Biological collection of Oswaldo Cruz Institute (access 2017-01-13)
-24.273322, -48.416938	Atlantic Forest	Iporanga	SP	Brazil	2013	Biological collection of Oswaldo Cruz Institute (access 2017-01-13)
-24.273322, -48.416939	Atlantic Forest	Iporanga	SP	Brazil	2014	Biological collection of Oswaldo Cruz Institute (access 2017-01-13)
-24.273322, -48.416940	Atlantic Forest	Iporanga	SP	Brazil	2015	Biological collection of Oswaldo Cruz Institute (access 2017-01-13)
-25.4333, -49.5333	Atlantic Forest	Campo Largo	PR	Brazil	2002	Embrapa cerrado (e-mail)
-25.4333, -49.5333	Atlantic Forest	Campo Largo	PR	Brazil	2002	Embrapa cerrado (e-mail)
-25.559544, -49.231099	Atlantic Forest	Curitiba	PR	Brazil	2004	Gouveia (2004)

APPENDIX A (continuation) – Occurrence points of *L. obliqua*

Coordinate	**Biome**	**City**	**State**	**Country**	**Sample date**	**Reference**
-25.685437, -54.438402	Atlantic Forest	Foz do Iguaçu	PR	Brazil	2008	Riella et al. (2008)
-25.835814, -49.048667	Atlantic Forest	Tijucas do Sul	PR	Brazil	2010-2011	Santos et al. (2015)
-26.380000, -49.080000	Atlantic Forest	Jaraguá do Sul	SC	Brazil	2011	GBIF (access 2017-01-15)
-26.380000, -49.080000	Atlantic Forest	Jaraguá do Sul	SC	Brazil	2011	GBIF (access 2017-01-15)
-26.666667, -53.550000	Atlantic Forest	West of SC	SC	Brazil	2001-2003	Cherem and Kammers (2008)
26°03`03.23" S 53°48`09.37" O	Atlantic Forest	San Antonio	Misiones	Argentina	2017	National Institute of Tropical Medicine
-27.265 , -53.8605556	Atlantic Forest	Derrubadas	RS	Brazil	2001	species link (access 2017-02-15)
-27.265 , -53.8605556	Atlantic Forest	Derrubadas	RS	Brazil	2001	species link (access 2017-02-15)
-27.265 , -53.8605556	Atlantic Forest	Derrubadas	RS	Brazil	2001	species link (access 2017-02-15)
-28.5347222 , -52.1533333	Atlantic Forest	Vila Maria	RS	Brazil	1995	species link (access 2017-02-15)
-28.5347222 , -52.1533333	Atlantic Forest	Vila Maria	RS	Brazil	1995	species link (access 2017-02-15)
-28.5347222 , -52.1533333	Atlantic Forest	Vila Maria	RS	Brazil	1995	species link (access 2017-02-15)
-28.5347222 , -52.1533333	Atlantic Forest	Vila Maria	RS	Brazil	1995	species link (access 2017-02-15)
-28.5347222 , -52.1533333	Atlantic Forest	Vila Maria	RS	Brazil	1995	species link (access 2017-02-15)
-28.5347222 , -52.1533333	Atlantic Forest	Vila Maria	RS	Brazil	1996	species link (access 2017-02-15)
-28.5347222 , -52.1533333	Atlantic Forest	Vila Maria	RS	Brazil	1996	species link (access 2017-02-15)
-28.5347222 , -52.1533333	Atlantic Forest	Vila Maria	RS	Brazil	1996	species link (access 2017-02-15)
-29.4386111 , -51.5113889	Atlantic Forest	Salvador do Sul	RS	Brazil	1994	species link (access 2017-02-15)
-29.4386111 , -51.5113889	Atlantic Forest	Salvador do Sul	RS	Brazil	1994	species link (access 2017-02-15)
-29.4386111 , -51.5113889	Atlantic Forest	Salvador do Sul	RS	Brazil	1994	species link (access 2017-02-15)
-29.4386111 , -51.5113889	Atlantic Forest	Salvador do Sul	RS	Brazil	1994	species link (access 2017-02-15)
-29.4386111 , -51.5113889	Atlantic Forest	Salvador do Sul	RS	Brazil	1995	species link (access 2017-02-15)
-29.4386111 , -51.5113889	Atlantic Forest	Salvador do Sul	RS	Brazil	1995	species link (access 2017-02-15)
-29.4386111 , -51.5113889	Atlantic Forest	Salvador do Sul	RS	Brazil	1995	species link (access 2017-02-15)
-29.4808333 , -50.1744444	Atlantic Forest	São Francisco de Paula	RS	Brazil	2001	species link (access 2017-02-15)

APPENDIX A (continuation) – Occurrence points of *L. obliqua*

Coordinate	**Biome**	**City**	**State**	**Country**	**Sample date**	**Reference**
-29.4808333 , -50.1744444	Atlantic Forest	São Francisco de Paula	RS	Brazil	2001	species link (access 2017-02-15)
-29.4808333 , -50.1744444	Atlantic Forest	São Francisco de Paula	RS	Brazil	2003	species link (access 2017-02-15)
-29.5380555556 , -51.0808333	Atlantic Forest	Morro Reuter	RS	Brazil	2005	species link (access 2017-02-15)
-29.7177778 , -52.4258333	Atlantic Forest	Santa Cruz do Sul	RS	Brazil	1995	species link (access 2017-02-15)
-29.7177778 , -52.4258333	Atlantic Forest	Santa Cruz do Sul	RS	Brazil	1995	species link (access 2017-02-15)
-29.7177778 , -52.4258333	Atlantic Forest	Santa Cruz do Sul	RS	Brazil	1995	species link (access 2017-02-15)
-29.7177778 , -52.4258333	Atlantic Forest	Santa Cruz do Sul	RS	Brazil	1995	species link (access 2017-02-15)
-29.7177778 , -52.4258333	Atlantic Forest	Santa Cruz do Sul	RS	Brazil	1995	species link (access 2017-02-15)
-29.7177778 , -52.4258333	Atlantic Forest	Santa Cruz do Sul	RS	Brazil	1995	species link (access 2017-02-15)
-29.7177778 , -52.4258333	Atlantic Forest	Santa Cruz do Sul	RS	Brazil	1995	species link (access 2017-02-15)
-29.7177778 , -52.4258333	Atlantic Forest	Santa Cruz do Sul	RS	Brazil	1995	species link (access 2017-02-15)
-31.564167, -53.433056	Pampa	Pinheiro Machado	RS	Brazil	2005-2007	Bernardi et al. (2011)
-31.5667, -53.3833	Pampa	P. Machado	RS	Brazil	2004	Embrapa cerrado (e-mail)
-31.7, -25.3333	Pampa	Pelotas	RS	Brazil	1997	Embrapa cerrado (e-mail)
S 26° 56,327’ - W 54° 27,080’	Atlantic Forest	Colonia El Saltito	Misiones	Argentina	2015	National Institute of Tropical Medicine
S 26°57.648’ W 054° 27.592’	Atlantic Forest	San Vicente	Misiones	Argentina	2015	National Institute of Tropical Medicine
S 27° 00,120’; W 54° 28,535’	Atlantic Forest	San Vicente	Misiones	Argentina	2015	National Institute of Tropical Medicine
S 27° 12,058’ - W 54° 39,334’	Atlantic Forest	25 de Mayo	Misiones	Argentina	2014	National Institute of Tropical Medicine
S 27° 12,211’ - W 054° 39,347’	Atlantic Forest	25 de Mayo	Misiones	Argentina	2015	National Institute of Tropical Medicine
S 27° 31,356’ - W 54° 55,146’	Atlantic Forest	Pje. Villa Unión/Eldoradito	Misiones	Argentina	2015	National Institute of Tropical Medicine
S 27° 31,544’ - W 54° 54,516’	Atlantic Forest	Campo Ramón	Misiones	Argentina	2015	National Institute of Tropical Medicine
S 27° 31,544’ - W 54° 54,516’	Atlantic Forest	Campo Ramón	Misiones	Argentina	2015	National Institute of Tropical Medicine

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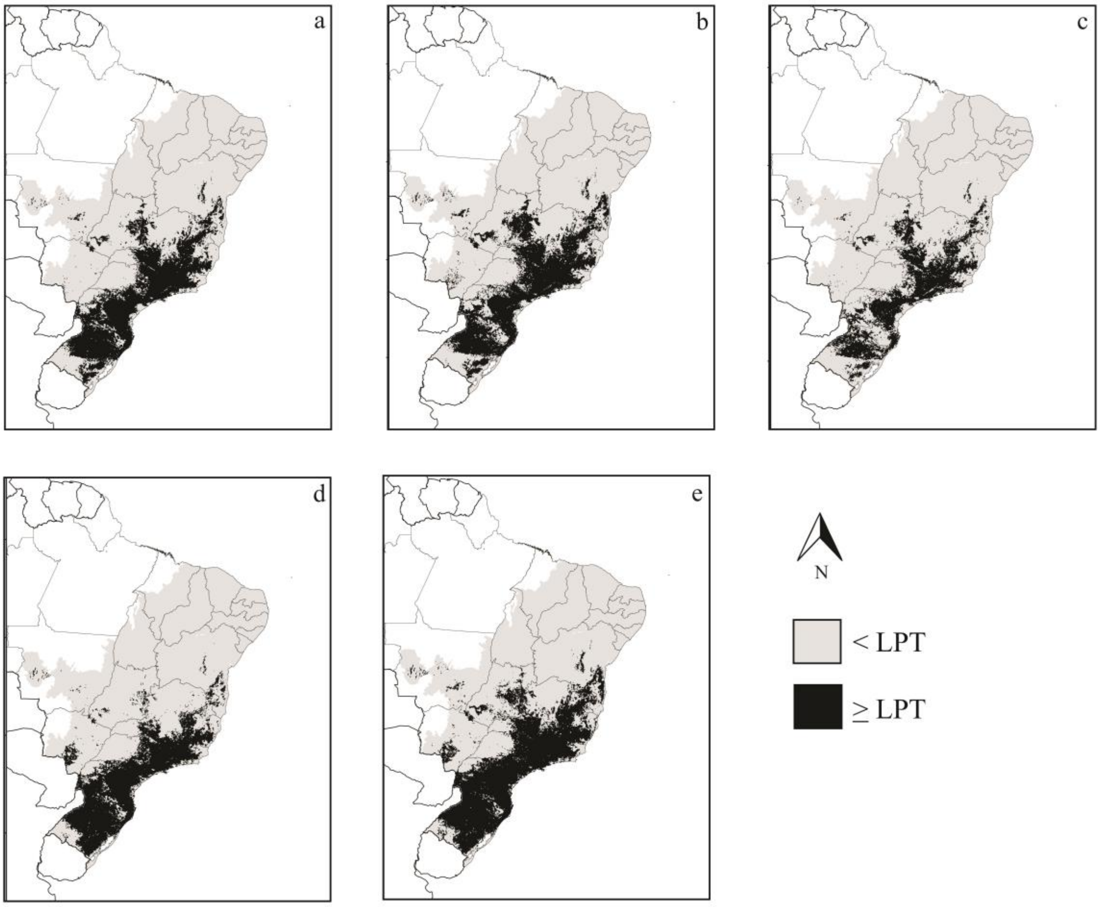
APPENDIX B

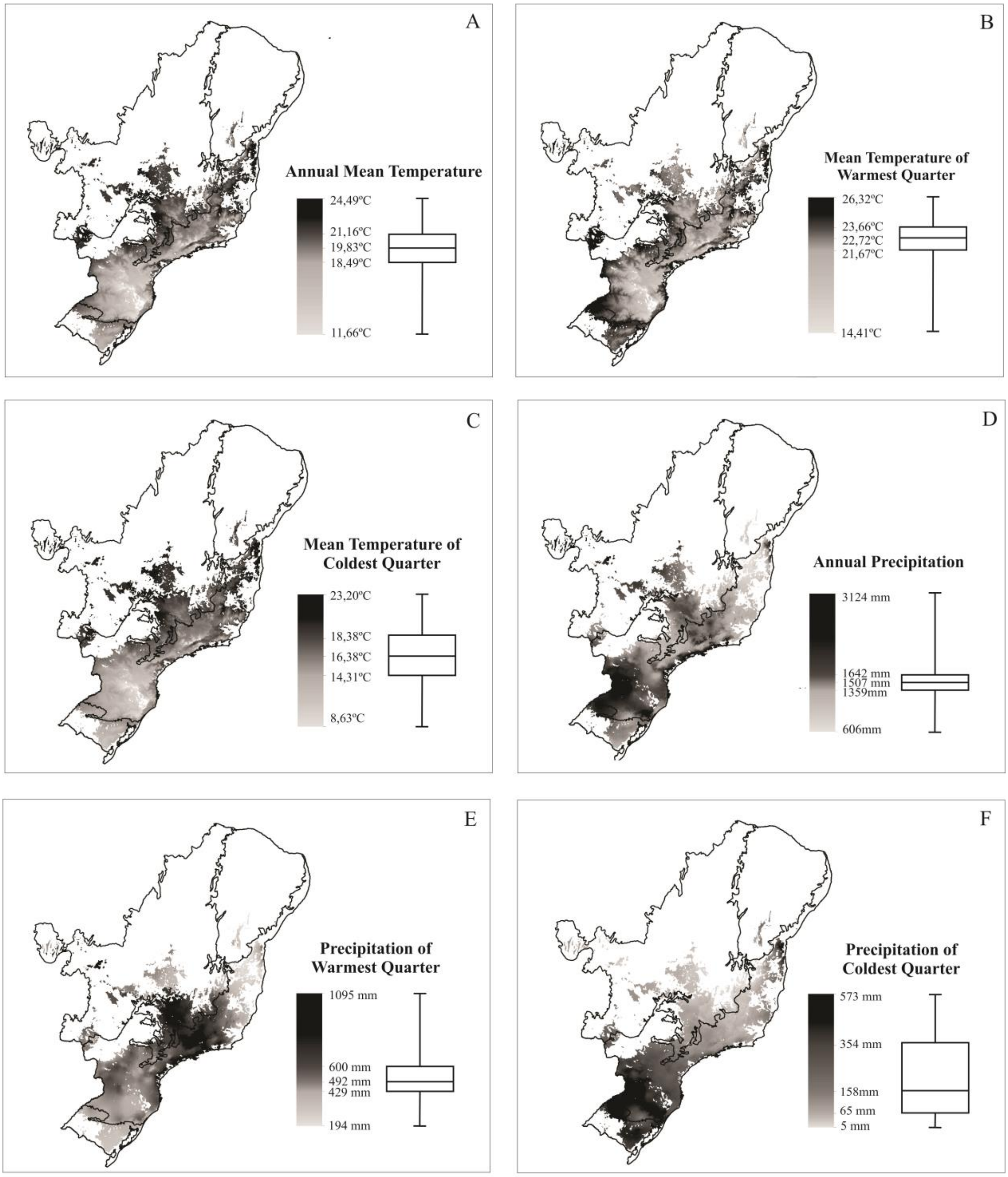
Combination of other ENM methodologies



a – Random vs. 38; b – Random vs. 380; c – Random vs. 3800; d – Bioclim vs. 380; e – Bioclim vs. 3800.

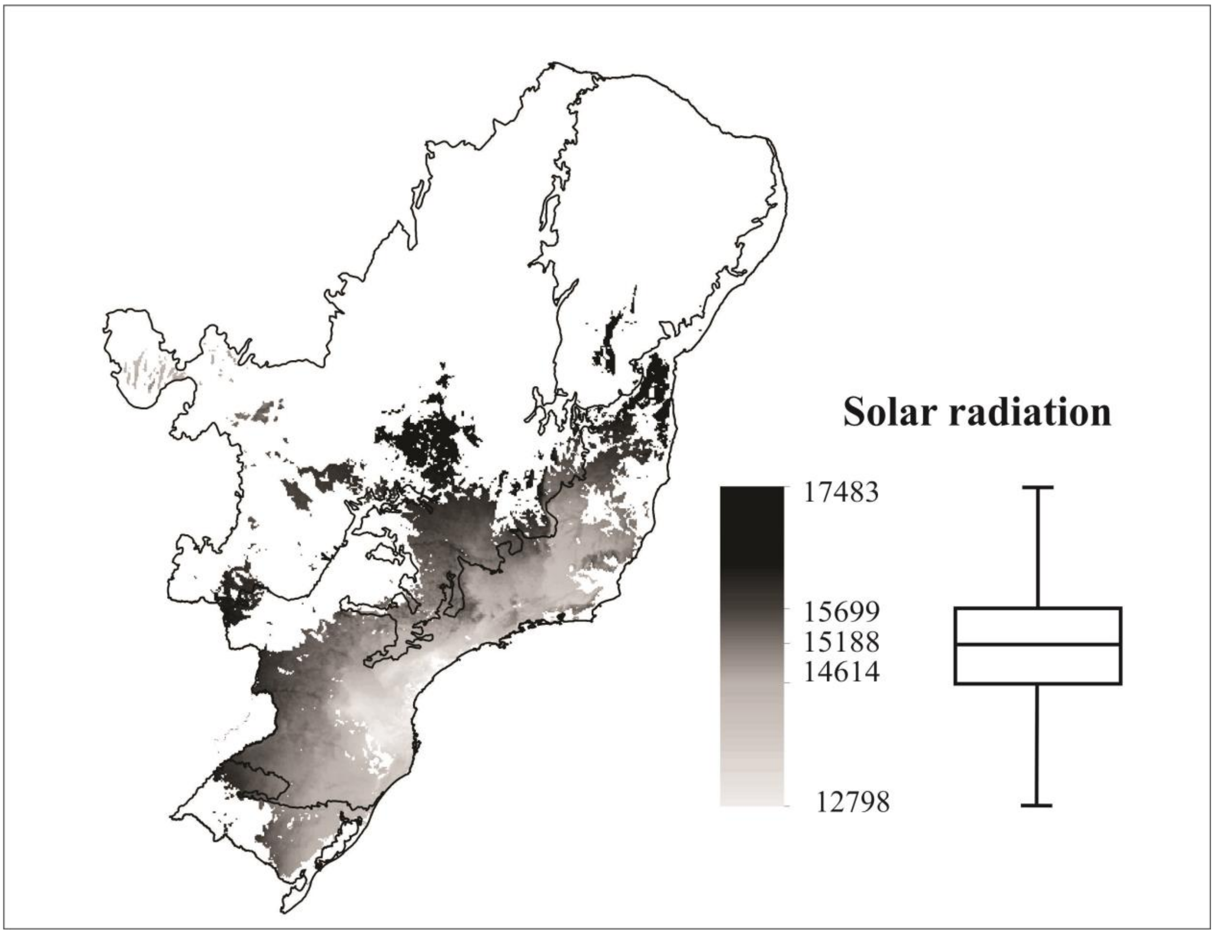
APPENDIX C

Variation of the biogeoclimatic variables within the predicted area for the distribution of *L. obliqua* in Brazil.



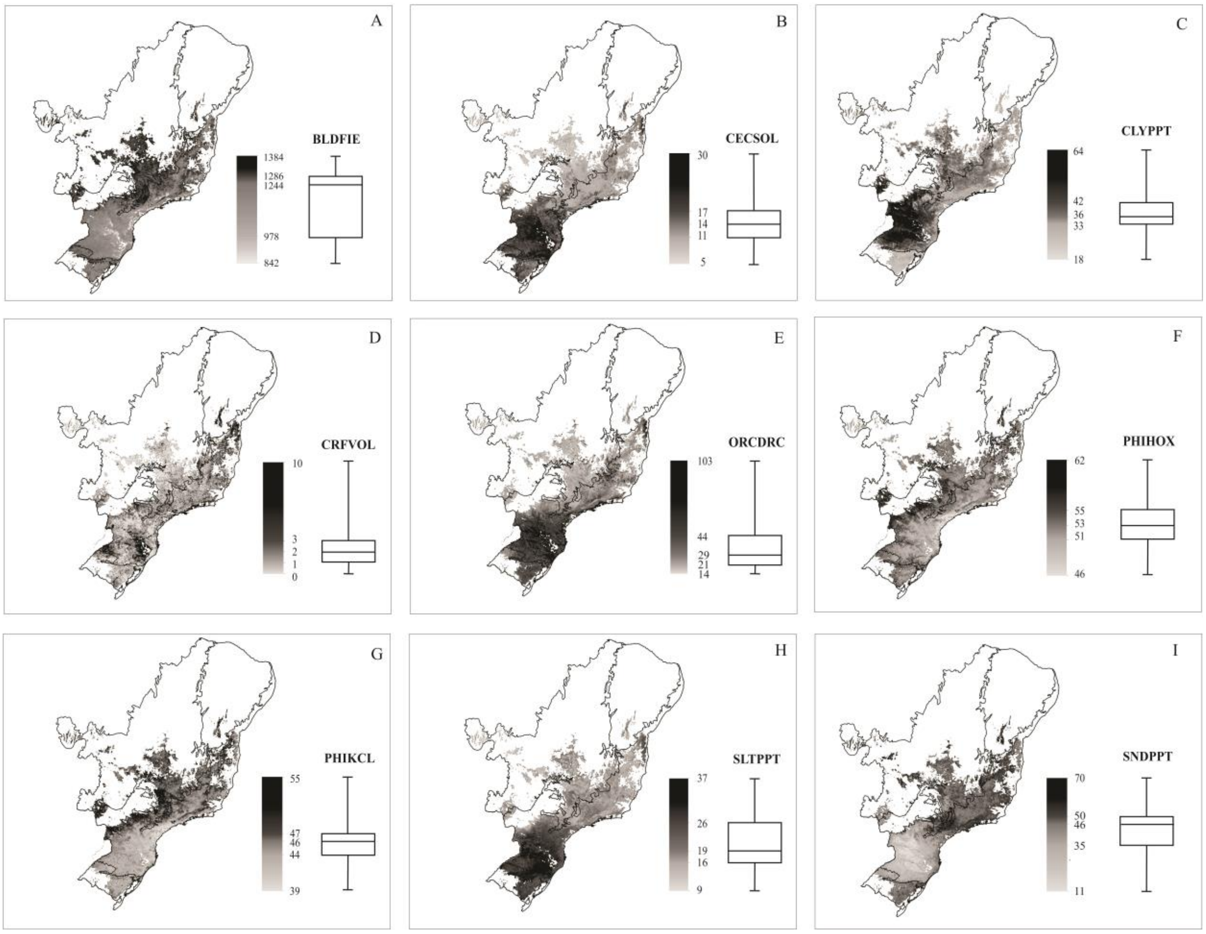
APPENDIX D

Variation of the solar radiation within the predicted area for the distribution of *L. obliqua* in Brazil.



APPENDIX E

Variation of soil variables within the predicted area as suitable for *L. obliqua.*



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