UNIVERSIDAD AUTÓNOMA DEL ESTADO DE MÉXICO

DOCTORADO EN CIENCIAS AGROPECUARIAS Y RECURSOS NATURALES

RESPUESTA DE LA HERPETOFAUNA A LA PERTURBACIÓN DEL HÁBITAT: UN ANÁLISIS MULTIESCALAR

TESIS

QUE PARA OBTENER EL GRADO DE DOCTOR EN CIENCIAS AGROPECUARIAS Y RECURSOS NATURALES

PRESENTA:

ANDREA GONZÁLEZ FERNÁNDEZ

COMITÉ DE TUTORES:

Tutor académico: Dr. Francisco Javier Manjarrez Silva
Tutores adjuntos: Dr. Víctor Arroyo Rodríguez
Dr. Gustavo Llorente

El Cerrillo, Piedras Blancas, Toluca, Estado de México.

Enero, 2019

Agradecimientos

Agradezco a las instituciones que me han apoyado en el trabajo: la Universidad Autónoma del Estado de México y CONACYT (Consejo Nacional de Ciencia y Tecnología). Al IGECEM por las imágenes SPOT. A todo el equipo con el que he estado trabajando: Armando Sunny, Maristella D'Addario, Javier Manjarrez, Víctor Arroyo-Rodríguez, Fabiola Ramírez-Corona, Arlene Aguilera-Hernández y Uri García-Vázquez. Agradezco también a los compañeros e investigadores que en algún momento me han apoyado con consejos o me han ayudado en alguna parte de la metodología: Francisco Reyna-Sáenz, Fabiola Judith Gandarilla-Aizpuro, Carmen Galán-Acedo, Gustavo Llorente y Andrés Lira-Noriega.

Índice

I. RESUMEN

El cambio de uso de suelo y el cambio climático están amenazando el mantenimiento de la biodiversidad a nivel global. Estos cambios ambientales afectan negativamente parámetros poblacionales como la distribución, abundancia o diversidad genética de muchas especies, incrementando su riesgo de extinción. Los anfibios y reptiles se encuentran entre los grupos de vertebrados más amenazados. El Eje Neovolcánico Transversal (ENT) es una de las provincias biogeográficas de México con mayor riqueza de especies, muchas de ellas endémicas; sin embargo, también es una de las zonas más perturbadas. En esta tesis evalúo la respuesta de la herpetofauna del ENT a los cambios en variables ambientales medidas a diferentes escalas espaciales. El primer capítulo analiza la distribución potencial (presente y futura) de las cinco especies de *Thamnophis* que se encuentran en el ENT y las variables (climáticas y de uso de suelo) más importantes que explican dicha distribución. Los hallazgos indican que las cinco especies responden negativamente al aumento de la cobertura de vegetación árida. Para *T. scalaris* y *T. scaliger*, la distancia al bosque de oyamel (*Abies religiosa*) fue la variable más importante: su probabilidad de ocurrencia disminuyó en sitios más alejados a estos bosques. Aunque una proporción mayoritaria de la distribución de estas especies se encuentra en el ENT, los resultados sugieren que todas las especies de *Thamnophis* sufrirán reducciones en sus distribuciones potenciales dentro de esta provincia biogeográfica en el futuro. Por lo tanto, este primer capítulo contribuye a entender la persistencia de este grupo herpetofaunístico ante un escenario de cambio ambiental. El segundo capítulo evalúa la abundancia y diversidad genética de una especie de salamandra (*Pseudoeurycea robertsi*) microendémica del Nevado de Toluca, y su respuesta a cambios en la cantidad y configuración del hábitat, a escala local y de paisaje. Este estudio demuestra que la abundancia de *P. robertsi* está más estrechamente relacionada con variables locales y del paisaje que la diversidad genética. La cantidad de hábitat a nivel local (i.e. volumen de troncos caídos) fue el predictor más importante, y se asoció positivamente con la abundancia de individuos y el número de alelos. La configuración del paisaje que rodea a los sitios de muestreo también juega un papel importante. En particular, la densidad de borde tuvo un efecto negativo en todas las variables de respuesta. En ambos estudios, el bosque de oyamel tuvo una importancia clave; en el primero para la presencia de las culebras *T. scalaris* y *T. scaliger*, en el segundo ya que se trata del macrohábitat de la salamandra *P. robertsi*. La preservación de los bosques de oyamel con áreas núcleo bien conservadas y con elevada disponibilidad de troncos caídos debe ser considerada una prioridad de conservación en el ENT. En este sentido, esta provincia biogeográfica tiene la mayor extensión de bosques de oyamel del país (91%), y en este estudio se demuestra que la extensión de estos bosques se ha mantenido estable entre 2002 y 2011. Sin embargo, las nuevas políticas llevadas a cabo en los últimos años en materia ambiental, las cuales reducen las restricciones en áreas de gran importancia ecológica, comprometen potencialmente la preservación de estos bosques y de las especies que de ellos dependen a largo plazo.

Palabras claves: *Thamnophis, Pseudoeurycea robertsi*, modelado de nicho ecológico, diversidad genética, ecología del paisaje, *Abies religiosa*.

II. INTRODUCCIÓN GENERAL

El cambio de uso de suelo y el cambio climático están amenazando el mantenimiento de la biodiversidad a nivel global (Newbold et al. 2016), alterando las comunidades, los ecosistemas y los servicios ambientales asociados a éstos (Parmesan & Yohe 2003, Cardinale et al. 2012). Las actividades productivas como la agricultura, la cría de ganado, la extracción de madera y la urbanización han transformado una gran proporción de la superficie del planeta (Foley et al. 2005). Estos cambios ambientales pueden afectar negativamente los parámetros poblacionales de muchas especies como la distribución, abundancia o diversidad genética (Hoffmann & Willi 2008, Ehrlén & Morris 2015), incrementando su riesgo de extinción.

Los anfibios son el grupo de vertebrados más amenazado (Catenazzi 2015) y sus poblaciones están disminuyendo rápidamente en todo el mundo (Stuart et al. 2004; Mendelson III et al. 2006, Eigenbrod et al. 2008), su baja vagilidad y restricciones fisiológicas los hace especialmente vulnerables a alteraciones en el ambiente (Huey 1982) como la pérdida del microhábitat térmicamente adecuado (Nowakowski et al. 2018). El 32.5% de las 5743 especies de anfibios descritas se encuentran amenazadas (Mendelson III et al. 2006). En cuanto a los reptiles, los declives de muchas poblaciones son similares a los que experimentan los anfibios en términos de amplitud taxonómica, distribución geográfica y gravedad (Gibbons et al. 2000).

México es uno de los cinco países más ricos en biodiversidad del mundo (Groombridge & Jenkins 2000); sin embargo, está sufriendo un rápido y extensivo cambio de uso de suelo (Mas et al. 2004) debido principalmente a la expansión de las actividades agropecuarias (Arredondo-León et al. 2008). Entre los años 1976 y 2000 más de 20,000 km^2

de bosque templado fueron talados en el país (deforestación anual del 0.25%; Mas et al. 2004). Los bosques templados se distribuyen en áreas de gran elevación como el Eje Neovolcánico Transversal (ENT). Esta provincia biogeográfica está conformada por un conjunto de sistemas montañosos y volcanes de diferentes edades, alineados en una franja que cruza el territorio mexicano de este a oeste. Se trata de una zona de transición entre las regiones Neártica y Neotropical que resulta en un solapamiento de las biotas de ambas regiones (Suárez-Atilano 2015). Su historia geológica y su posición geográfica la convierten en una zona muy compleja con 30 tipos climáticos distintos y diferentes comunidades vegetales como los bosques de coníferas (*Pinus* sp. y *Abies religiosa*), bosques de encino (*Quercus* sp.), bosques mesófilos, pastizales alpinos, zonas de matorral subalpino y zonas de vegetación riparia (Espinoza & Ocegueda 2007). Por estos motivos, el ENT preserva una de las comunidades herpetofaunísticas más ricas del país y la más importante en número de especies endémicas de anfibios y reptiles (Flores-Villela & Canseco-Márquez 2007, Sunny et al. 2017). Desafortunadamente, es una de las zonas más perturbadas de México ya que contiene las mayores áreas urbanas del país (CONAPO 2010).

El objetivo general del trabajo fue entender la respuesta de la herpetofauna del ENT a los cambios en variables ambientales medidos a diferentes escalas espaciales. El primer caso de estudio se enfocó en un grupo de especies relacionadas taxonómicamente y con una distribución relativamente amplia por lo que las variables de respuesta que se utilizaron fueron registros de presencia, que son los más fáciles de obtener si queremos abarcar una extensión geográfica amplia. Para este estudio se utilizaron variables ambientales a 1 km de resolución, tanto climáticas como de uso de suelo, abarcando todo el Altiplano Mexicano. El objetivo consistió en determinar la distribución potencial presente y futura de las cinco especies de *Thamnophis* que se encuentran en el ENT y las variables más importantes que explican su distribución, con el fin de evaluar la persistencia de este grupo herpetofaunístico ante un escenario de cambio ambiental. En el segundo caso se estudió un anfibio, la salamandra *Pseudoeurycea robertsi*. Esta especie tiene una distribución muy restringida, ya que es microendémica del Nevado de Toluca (un volcán del ENT). Para este estudio se utilizaron registros de abundancia y diversidad genética de toda su distribución geográfica y variables ambientales a gran resolución (10 metros). El objetivo fue evaluar la influencia relativa de la cantidad y configuración del hábitat, a escala local y de paisaje, en la abundancia y diversidad genética de *P. robertsi*.

III. CAPÍTULO 1 (PRIMER ARTÍCULO)

Este artículo fue publicado en la revista *Peer J* el 11 de Abril de 2018. (https://peerj.com/articles/4618/?utm_source=TrendMD&utm_campaign=PeerJ_TrendMD_1&utm_medium=TrendMD)

PeerJ

Present and future ecological niche modeling of garter snake species from the Trans-Mexican Volcanic Belt

Andrea González-Fernández[1,*], Javier Manjarrez[1], Uri García-Vázquez[2], Maristella D'Addario[3] and Armando Sunny[3,*]

[1] Facultad de Ciencias, Universidad Autónoma del Estado de México, Toluca, Estado de México, México
[2] Facultad de Estudios Superiores Zaragoza, Universidad Nacional Autónoma de México, Ciudad de México, México
[3] Centro de Investigación en Ciencias Biológicas Aplicadas, Universidad Autónoma del Estado de México, Toluca, Estado de México, México
[*] These authors contributed equally to this work.

ABSTRACT

Land use and climate change are affecting the abundance and distribution of species. The Trans-Mexican Volcanic Belt (TMVB) is a very diverse region due to geological history, geographic position, and climate. It is also one of the most disturbed regions in Mexico. Reptiles are particularly sensitive to environmental changes due to their low dispersal capacity and thermal ecology. In this study, we define the important environmental variables (considering climate, topography, and land use) and potential distribution (present and future) of the five *Thamnophis* species present in TMVB. To do so, we used the maximum entropy modeling software (MAXENT). First, we modeled to select the most important variables to explain the distribution of each species, then we modeled again using only the most important variables and projected these models to the future considering a middle-moderate climate change scenario (rcp45), and land use and vegetation variables for the year 2050 (generated according to land use changes that occurred between years 2002 and 2011). Arid vegetation had an important negative effect on habitat suitability for all species, and minimum temperature of the coldest month was important for four of the five species. *Thamnophis cyrtopsis* was the species with the lowest tolerance to minimum temperatures. The maximum temperature of the warmest month was important for *T. scalaris* and *T. cyrtopsis*. Low percentages of agriculture were positive for *T. eques* and *T. melanogaster* but, at higher values, agriculture had a negative effect on habitat suitability for both species. Elevation was the most important variable to explain *T. eques* and *T. melanogaster* potential distribution while distance to *Abies* forests was the most important variable for *T. scalaris* and *T. scaliger*. All species had a high proportion of their potential distribution in the TMVB. However, according to our models, all *Thamnophis* species will experience reductions in their potential distribution in this region. *T. scalaris* will suffer the biggest reduction because this species is limited by high temperatures and will not be able to shift its distribution upward, as it is already present in the highest elevations of the TMVB.

Subjects Conservation Biology, Zoology, Climate Change Biology, Environmental Impacts, Spatial and Geographic Information Science
Keywords Climate change, Environmental niche models, *Thamnophis*, Potential distribution, Land-use change

Submitted 15 December 2017
Accepted 24 March 2018
Published 11 April 2018

Corresponding authors
Andrea González-Fernández, andreagofe@gmail.com
Javier Manjarrez, jsilva@ecologia.unam.mx
Armando Sunny, sunny.biologia@gmail.com

Academic editor
Jose Maria Cardoso da Silva

Additional Information and Declarations can be found on page 13

DOI **10.7717/peerj.4618**

How to cite this article González-Fernández et al. (2018), Present and future ecological niche modeling of garter snake species from the Trans-Mexican Volcanic Belt. **PeerJ 6:e4618; DOI 10.7717/peerj.4618**

INTRODUCTION

Land use and climate change are affecting the abundance and distribution of species, altering biological communities, ecosystems, and their associated services to humans (*Parmesan & Yohe, 2003*; *Cardinale et al., 2012*; *Kortsch et al., 2015*; *Nadeau, Urban & Bridle, 2017*). Both factors are the main contributors to the global decline of reptiles (*Ribeiro et al., 2009*; *Schneider-Maunoury et al., 2016*; *Sunny, González-Fernández & D'Addario, 2017*). In fact, some studies indicate that 20% of the world's reptile species are threatened (*Böhm et al., 2013*) because they are particularly sensitive to environmental changes due to their low dispersal capacity and thermal ecology (*Huey, 1982*; *Castellano & Valone, 2006*; *Ribeiro et al., 2009*; *Russildi et al., 2016*). Studies predicting biological responses to land use and climate change are therefore necessary to assess the potential impacts of these changes and develop management decisions and conservation strategies (*Jimenez-Valverde & Lobo, 2007*; *Nadeau, Urban & Bridle, 2017*) to mitigate negative impacts. Information concerning species' distributions is essential in these cases (*Liu, White & Newell, 2013*). Through species occurrence data and environmental information, we can generate environmental niche models that can be used to predict the location of particular areas where environmental conditions are favorable for the presence of the species of study (*Suárez-Atilano, 2015*).

The Trans-Mexican Volcanic Belt (TMVB) is a set of mountain ranges and volcanoes of different ages, aligned on a strip that crosses Mexico from west to east. It is a transition area between Nearctic and Neotropical regions that results in an overlap of biotas from both regions (*Suárez-Atilano, 2015*). Its geological history and geographic position make it a very complex area with 30 distinct climatic types and several different vegetation communities, such as coniferous forests (*Pinus* sp. and *Abies* sp.), oak forests (*Quercus* sp.), mesophyll forests, alpine pastures, subalpine scrub, and riparian vegetation zones (*Espinoza & Ocegueda, 2007*). For these reasons, the TMVB has the second highest herpetological richness in Mexico and is the most important biogeographic region of the country in number of endemic amphibian and reptile species (*Flores-Villela & Canseco-Márquez, 2007*; *Sunny, González-Fernández & D'Addario, 2017*). Due to the complex characteristics of the TMVB, the montane taxa of this region have been exposed to a sky-island dynamic through climate fluctuations (*Mastretta-Yanes et al., 2015*), consequently, the high-altitude-adapted species could be especially vulnerable to climate change as they may be limited by future rising temperatures (*Sunny, González-Fernández & D'Addario, 2017*). Moreover, the TMVB is one of the most disturbed regions in the country as it contains the biggest metropolitan areas of Mexico (*CONAPO, 2010*; *Sunny, González-Fernández & D'Addario, 2017*).

Garter snakes are among the most abundant snake species in North America (*Rossman, Ford & Seigel, 1996*; *De Queiroz, Lawson & Lemos-Espinal, 2002*) and they are distributed from Canada to Costa Rica (*Rossman, Ford & Seigel, 1996*; *Manjarrez, 1998*; *De Queiroz, Lawson & Lemos-Espinal, 2002*). However, we lack information on the ecology and current conservation status of most *Thamnophis* species that are endemic to, or primarily distributed in, Mexico (*Manjarrez, Venegas-Barrera & García-Guadarrama, 2007*). They are also the most abundant snake genus in the TMVB (*Flores-Villela, Canseco-Márquez*

& Ochoa-Ochoa, 2010), thus they have an important ecological role in the ecosystem (*Montoya, Pimm & Solé, 2006*). These garter snakes also have great ecological plasticity in reproduction, feeding, and thermal ecology (*Seigel, 1996*).

For this study, we chose the five *Thamnophis* species that occur in the TMVB. *Thamnophis melanogaster* is endemic to the Mexican Central Plateau. It is a semiaquatic species that inhabits the edges of water bodies and specializes in underwater foraging, preying on aquatic animals such as fish, tadpoles and leeches (*Rossman, Ford & Seigel, 1996*). *Thamnophis scalaris* is endemic to high elevations across the TMVB (*Rossman, Ford & Seigel, 1996*). It lives in grasslands and the periphery of forests, and feeds mainly on earthworms, although it can eat vertebrates such as mice and lizards (*Uribe-Peña, Ramirez-Bautista & Casas-Andreu, 1999*; *Manjarrez, Venegas-Barrera & García-Guadarrama, 2007*). *Thamnophis scaliger* is a poorly-known montane species, endemic to central Mexico (*Rossman, Ford & Seigel, 1996*). It inhabits forests where it feeds on frogs, salamanders and lizards (*Uribe-Peña, Ramirez-Bautista & Casas-Andreu, 1999*). *Thamnophis cyrtopsis* extends from the southern United States to Guatemala, although is mainly distributed in Mexico (*Hammerson, 2013*). It is an amphibian specialist in aquatic habitats from subtropical deciduous and mixed forests (*Rossman, Ford & Seigel, 1996*). *Thamnophis eques* is widely distributed over the Mexican Plateau, reaching southern Arizona and New Mexico (*Rossman, Ford & Seigel, 1996*). It is a generalist predator because it feeds on both aquatic and terrestrial prey—mostly frogs, tadpoles, and fish, supplemented by lizards and mice (*Drummond & Macías García, 1989*; *Manjarrez, 1998*).

Despite their widespread distribution and relatively high abundance in comparison to other reptiles, this group has suffered critical reductions in the last 10 years (*Canseco-Márquez & Mendoza-Quijano, 2007*; *Hammerson, 2013*; *Vázquez Díaz & Quintero Díaz, 2007*; *Hammerson, Vázquez Díaz & Quintero Díaz, 2007*). Thus, knowledge regarding their ecological niche and their present and future potential distribution is key to better understanding the causes of their population decline. Moreover, due to the geological history and geographic position of the TMVB, the animals of this region constitute a cenocron (a group of animals originated in a defined area that have coexisted for a long period, thus sharing a common biogeographic history and a distribution pattern; *Halffter & Morrone, 2017*), therefore, changes in garter snake species' distributions in the TMVB may represent future changes in other species' distributions of this region. We expect that land use and climate change will reduce the future potential distribution of these five garter snake species (i.e., a reduction in the suitable area available for each species). In this study, we aimed to answer the following four questions. (1) Which climatic, topographic and land use variables determine each species' distribution? (2) Considering land use and climate change, what are the present and future potential distributions of each species? (3) What changes in suitable available area in the TMVB will each species undergo in the future? (4) What changes in suitable available area in the entire country will each species undergo in the future?

MATERIALS AND METHODS

We modeled the potential distribution of the five *Thamnophis* species that occur in the TMVB (*T. cyrtopsis*, *T. eques*, *T. melanogaster*, *T. scalaris,* and *T. scaliger*). Occurrence records were obtained from fieldwork (60% or more; Table S1) and online databases such as Global Biodiversity Information Facility (GBIF) and iNaturalist. We selected only the records from the last 20 years for the analysis, as extensive land use changes occurred in Mexico during the 1990s (*FAO, 1993*) and we included some land use variables (such as extent of induced grasslands and agriculture areas) in the analysis. Maps of occurrence data for all five species were generated to check for obvious errors. We also filtered these data to eliminate duplicated observations from the same pixel (1-km resolution). We defined a polygon (background) for each species that represents the accessibility area (*Suárez-Atilano, Burbrink & Vázquez-Domínguez, 2014*; *Suárez-Atilano et al., 2017*). These polygons were generated considering biogeographic regions with geographical records or records near their borders (*Sunny, González-Fernández & D'Addario, 2017*). Three of the species are endemic to Mexico and the other two have only a marginal distribution outside the country (*Rossman, Ford & Seigel, 1996*; *Hammerson, 2013*), therefore all polygons are large representative regions of species distribution ranges. We obtained bioclimatic variables from WorldClim (*Hijmans et al., 2005*); topographic and land cover variables were obtained from the National Institute of Statistics and Geography (*INEGI, 2013*). We reclassified the land use map (series V, 1:250,000, generated during the period 2011 to 2013; *INEGI, 2013*) in different exclusive classes that were converted to raster and transformed from categorical to continuous using a resample method that averages the value of the surrounding pixels to assign a new value to each pixel. All layers were processed in a raster format, with 1-km resolution, using ARC GIS 10.5 and the packages RASTER (*Hijmans, 2016*) and RGDAL (*Bivand, Keitt & Rowlingson, 2017*) for R software (version 3.4.0; *R Development Core Team, 2017*). After a bibliographic review and Pearson correlation analysis to discard highly correlated variables ($R^2 > 0.8$, *Suárez-Atilano, 2015*) we selected the following variables: elevation, percent natural grasslands, percent human-induced grasslands, percent arid vegetation, percent *Pinus* forest, distance to *Pinus* forest, percent *Quercus* forest, distance to *Quercus* forest, percent *Abies* forest, distance to *Abies* forest, distance to water sources, percent agriculture, minimum temperature of the coldest month, maximum temperature of the warmest month, precipitation of the wettest month, and precipitation of the driest month.

We used the maximum entropy modeling software (MAXENT; *Phillips, Anderson & Schapire, 2006*) which estimates species' distributions by finding the distribution of maximum entropy (the most spread out, or closest to uniform), subject to constraints imposed by a known distribution of the species, and by the environmental conditions across the study area (*Anderson & González Jr, 2011*). First, we ran the model for each species in MAXENT with 10 replicates and we selected the most important variables that explained the distribution of each species (*Anderson, Lew & Peterson, 2003*; *Chefaoui, Hortal & Lobo, 2005*; *Suárez-Atilano et al., 2017*). We only used linear and quadratic features because we had less than 80 records of *T. scaliger* (*Merow, Smith & Silander, 2013*) and, for an easier comparison, we used the same methodology for all species. All analyses were performed using the logistic output for an easier interpretation and a convergence threshold of 1×10^{-5} with 500 iterations (*Pearson, 2007*; *Suárez-Atilano, 2015*). We modeled again, this time with only the most important variables for each species (*Guisan & Zimmerman, 2000*; *Guisan & Thuiller, 2005*; *Araujo & Guisan, 2006*) and projected these models to the future using both clumping (restricting the variables to the range of values encountered during model calibration) and extrapolation methods (*Merow, Smith & Silander, 2013*). We obtained the future bioclimatic variables CCSM4 for the year 2050 considering the climate change scenario rcp45 (middle-moderate) from WorldClim. Land use and vegetation variables for the year 2050 were generated using the module LAND CHANGE MODELER FOR ECOLOGICAL SUSTAINABILITY in IDRISI SELVA 17.0 software (*Clark Labs, 2012*) and land use and vegetation layers from years 2002 and 2011 (series III and V; *INEGI, 2005*; *INEGI, 2013*). We also used elevation, slope (obtained from the elevation layer), and distance to urban settlements, for a better prediction of land use change. We designated present urban areas (from the present distribution maps) and future urban areas (from the future distribution maps) as areas of zero habitat suitability. We did not include distance to urban areas as a variable in the models because this can generate a bias, as these areas are more easily accessed by observers (*Araujo & Guisan, 2006*). We generated present and future potential distribution maps for each species. We preferred to show the continuous maps because binary outputs can obscure important biological detail (*Liu, White & Newell, 2013*). To evaluate model performance, we used partial Receiving Operating Characteristic (partial ROC) analyses (*Peterson, Papes & Soberón, 2008*; *Osorio-Olvera, 2016*) as recommended based on criticisms of area under the curve analyses (AUC) (*Lobo, Jiménez-Valverde & Real, 2008*; *Peterson, Papes & Soberón, 2008*). While AUC evaluates only the environmental niche model (under the omission-commission framework) performance, partial-ROC allows for statistical significance from the AUC itself, based on a null distribution of expectations created via bootstrapping replacement of 50% of the total available points and 1,000 resampling replicates (*Suárez-Atilano, 2015*). One-tailed significance of the difference between AUC and the null expectations was assessed by fitting a standard normal variate (the z-statistic) and calculating the probability that the mean AUC ratio was ≤ 1. We used 75% of occurrence localities for model training and 25% for model testing (*Suárez-Atilano, 2015*). We used the platform NICHE TOOLBOX for partial-ROC calculations (*Osorio-Olvera, 2016*). We generated the species potential distribution binary maps using Max SS threshold (*Liu, White & Newell, 2013*), a threshold selection method based on maximizing the sum of sensitivity and specificity. This is considered an adequate method to use when reliable absence data are unavailable (*Liu, White & Newell, 2013*). For each species, we used these binary maps to calculate the present and future high-suitability areas (*Suárez-Atilano, 2015*) in both all of Mexico, and the TMVB only, to assess whether the distribution of each species will decrease or increase in the future.

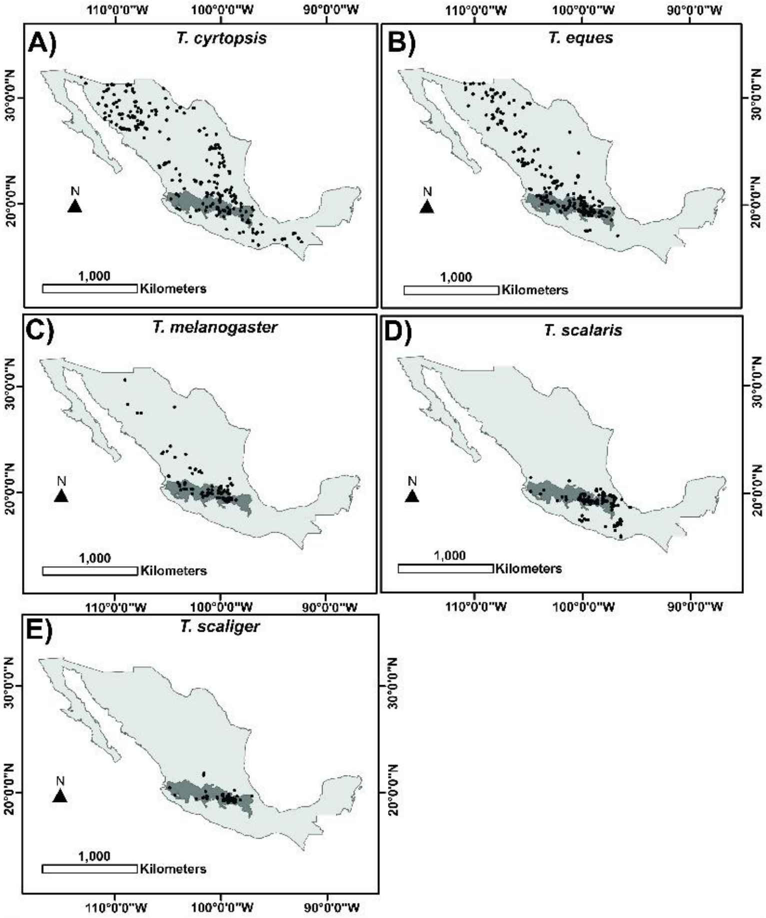


Figure 1 Occurrence records used to build the distribution model for each *Thamnophis* species, showing the Trans-Mexican Volcanic Belt (TMVB) in dark gray.

Full-size DOI: 10.7717/peerj.4618/fig-1

RESULTS

After filtering the data, we worked with 267 records of *T. cyrtopsis* (Fig. 1A), 274 of *T. eques* (Fig. 1B), 103 of *T. melanogaster* (Fig. 1C), 186 of *T. scalaris* (Fig. 1D), and 76 of *T. scaliger* (Fig. 1E). The most important variables for each *Thamnophis* species are summarized in Table 1. In all cases these variables together explained 60% or more of the species' potential distribution. It is important to note that arid vegetation had an important negative effect

Table 1 **Contribution percent of the most important variables that explain the distribution of each *Thamnophis* species.**

Variables	*T. cyrtopsis*	*T. eques*	*T. melanogaster*	*T. scalaris*	*T. scaliger*
Minimum temperature of the coldest month	33.7	19.2	11.3		26.5
Maximum temperature of the warmest month	5.2			36	
Elevation		28.4	27.3		
Arid vegetation	26.4	15.6	11.5	4.9	5.3
Agriculture		9.6	12.9		
Distance to *Quercus* forest	8.5				
Distance to *Abies* forest				44.9	40.6
Total	73.8	72.8	63	85.8	72.4

on habitat suitability for all species (Fig. S1) and the minimum temperature of the coldest month was important in four of the five models. This latter variable was the most important to explain *T. cyrtopsis* potential distribution, which was the species with the lowest tolerance to minimum temperatures (5 °C). Habitat suitability for *T. scalaris* and *T. cyrtopsis* steadily decreased when maximum temperatures increased. Low agriculture percentages were positive for *T. eques* and *T. melanogaster* but, at higher values (above 30%), agriculture had a negative effect on habitat suitability for both species. Elevation was the most important variable to explain *T. eques* and *T. melanogaster* potential distribution. It was a positive variable for *T. melanogaster*, while habitat suitability for *T. eques* was optimal near 2,500 m above sea level (masl). Distance to *Abies* forests was the most important variable to explain *T. scalaris* and *T. scaliger* potential distribution. It had negative effects on these species (as distance to *Abies* forests increase, habitat suitability decrease), which means that proximity to *Abies* forests was positive for both species. Distance to *Quercus* forests had a negative effect on habitat suitability for *T. cyrtopsis*, which means that proximity to these forests was positive for the species.

Between 2002 and 2011, there was an increase of almost 16,000 km^2 in agriculture and about 5,000 km^2 in urban areas (Fig. 2). There was also an increase in human-induced grasslands. A reduction in arid vegetation and natural grasslands occurred, mainly because of its conversion to agriculture lands. The area of *Pinus* and *Quercus* forests also fell, but *Abies* forests held steady. For the year 2050, an increase of 82,865 km^2 in agriculture areas is expected according to the model (Figs. 3A, 3B). The urban areas will increase by 20,392 km^2 (Figs. 3C 3D), most of it taking place in the area around Toluca city (Figs. 3C, 3D), and induced grasslands will increase by 24,796 km^2 (Figs. 3E, 3F). Potential distribution maps for each species are in Figs. 4A–4J. We found no differences in future projections between extrapolation and clumping methods. Partial-ROC bootstrap tests showed significant ratio values of empirical AUC over null expectations (mean AUC ratios $\geq$1.5 and *p*-values <0.001 in all cases; Fig. S1).

All species had a high proportion of their potential distribution in the TMVB. However, according to high-suitability area calculations for present and future, all *Thamnophis* species will experience reductions in their distribution in this region (Table 2). *T. scaliger* was only distributed in the TMVB, while *T. scalaris* had a small part of its potential distribution in the Sierra Madre del Sur. *T. cyrtopsis*, *T. eques*, and *T. melanogaster* were

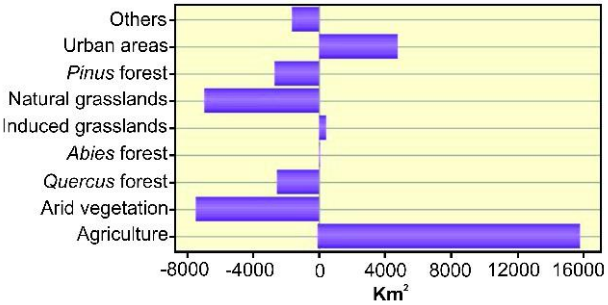


Figure 2 **Land use change by category (km²) between years 2002 and 2011.**
Full-size DOI: 10.7717/peerj.4618/fig-2

also distributed in the Sierra Madre Occidental, Sierra Madre Oriental, Sierra Madre del Sur, and Oaxaca mountain ranges. Unlike the TMVB, these biogeographic regions will not suffer important reductions in suitable habitat for *T. eques* and *T. melanogaster* in the future. *T. cyrtopsis* will suffer important reductions in all its potential distribution, which also includes Chiapas Highlands. The potential distribution of *T. melanogaster* will increase in the future, considering the entire country, and *T. scalaris* will suffer the biggest reduction of the five species (reductions of 54.08% for the TMVB, and 54.30% for all of Mexico, Table 2; Figs. 4A–4J).

DISCUSSION

Environmental variables

Although current records and literature support the idea that grasslands and water sources are essential for *Thamnophis* species in Mexico (*Jones, 1990*; *Manjarrez & Drummond, 1996*; *Venegas-Barrera & Manjarrez, 2011*), these variables were not selected by the model as important to explain the distribution of the species. Both variables are more related to the microhabitat of the species, but for this study, we modeled the macrohabitat. Although most records of *Thamnophis* are in grasslands or near water sources (lakes, ponds and streams), these habitat features are present throughout most of the country, including areas where the species is not present, therefore, these variables are not limiting the species at a macro level. The percent of arid vegetation (which can be interpreted as the opposite of water sources) was a negative limiting factor for all species (Table 1). Distances to forests were more important for explaining the presence of *Thamnophis* species than the percent of these forests. This was especially important for *T. scalaris* and *T. scaliger* as distance to *Abies* forest was the most important variable determining their potential distribution. These results are consistent with our fieldwork observations, as we found only a few individuals inside forests; the majority were found in grasslands near coniferous forests. This could be because coniferous forests occur in a moist (1,000–3,800 mm annual precipitation) and cold microclimate (2°–24 °C; *Sáenz-Romero et al., 2012*; *Sunny, González-Fernández &*

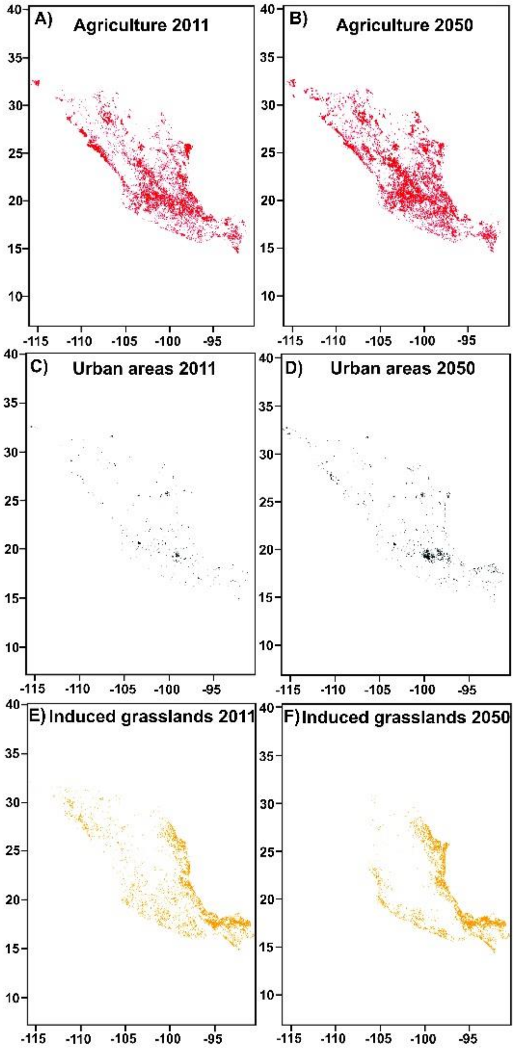


Figure 3 Present (2011) and future (2050) maps of (A, B) agriculture, in red; (C, D) urban, in gray; and (E, F) induced grasslands, in orange.

Full-size DOI: 10.7717/peerj.4618/fig-3

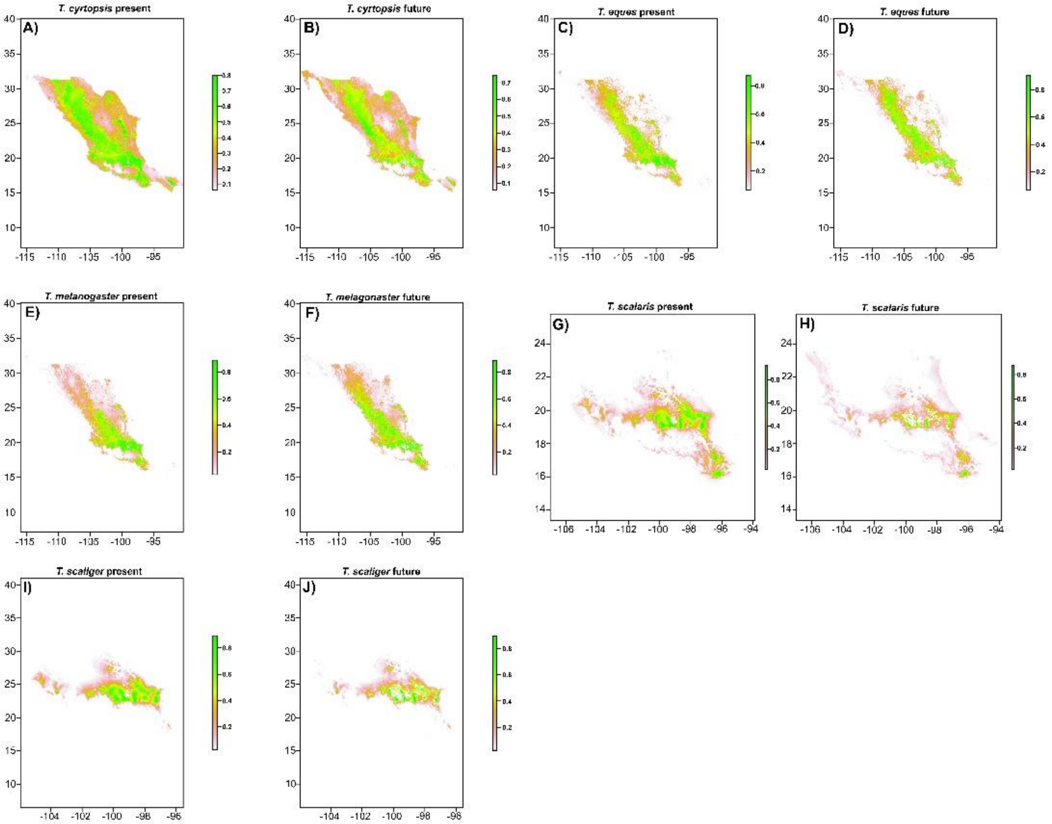


Figure 4 **Present (2011) and future (2050) potential distribution maps for each *Thamnophis* species: (A, B) *T. cyrtopsis*, (C, D) *T. eques*, (E, F) *T. melanogaster*, (G, H) *T. scalaris* and (I, J) *T. scaliger*.**

Full-size DOI: 10.7717/peerj.4618/fig-4

D'Addario, 2017) that is preferred by *Thamnophis* species (Manjarrez & Drummond, 1996). Therefore, microclimatic conditions of grasslands surrounded by forests and large areas of grasslands without forest, may be different. Grasslands surrounded by forests offer the climatic benefits of forests (moist and cold), and the food benefits of grasslands (higher availability of small prey; (Bastos, Araújo & Silva, 2005; Reinert et al., 2011; Wittenberg, 2012; Mociño Deloya, Setser & Pérez-Ramos, 2014).

Low percent of agriculture was positive for *T. eques* and *T. melanogaster*, but a high percent was negative for both species. This could be because agriculture is a tradeoff for many reptile species, especially snakes. It provides benefits for them, such as higher prey

Table 2 Present and future high suitability area (km^2) and percent of reductions in these areas for each *Thamnophis* species in Mexico and the Trans-Mexican Volcanic Belt (TMVB).

	MEXICO			TMVB		
	Present distribution (Km^2)	**Future distribution (Km^2)**	**Reduction (%)**	**Present distribution (Km^2)**	**Future distribution (Km^2)**	**Reduction (%)**
T. cyrtopsis	661,888.53	387,393.67	41.47	103,190.15	56,172.18	45.56
T. eques	583,936.04	554,336.36	5.07	102,001.64	88,928.44	12.82
T. melanogaster	255,647.78	317,411.39	−24.16	83,237.55	67,581.46	18.81
T. scalaris	110,441.63	50,474.08	54.30	54,057.65	24,825.27	54.08
T. scaliger	58,682.16	37,278.67	36.47	42,804.76	26,617.94	37.82

availability, but also exposes them to human interactions (i.e., people kill *Thamnophis* out of fear even though these species are not dangerous to humans; Sunny et al., 2015). Moreover, the persistent practice of crop burning and use of roller-chopping to prepare fields also affect their populations (*Mullin & Seigel, 2009*).

Environmental temperature is important for ectothermic species like garter snakes because they are more active when they can maintain a body temperature above approximately 22 °C (*Manjarrez & Drummond, 1996*). Environmental temperature increases may lead *Thamnophis* to physiological stress that results in reduced fitness (*Peterson, Gibson & Dorcas, 1993*). The fact that *T. cyrtopsis* was limited by low and high temperatures could be the cause of its reduced potential distribution. The maximum temperature of the warmest month was one of the most important variables explaining *T. scalaris* potential distribution; however, for all other species, the minimum temperature of the coldest month was more important. This may be because *T. scalaris* is the species occurring at the highest elevation and, consequently, is adapted to a colder climate. Therefore, while other species are more limited by lower temperatures, *T. scalaris* is more limited by higher ones, which could make this species more vulnerable to warming temperatures associated with climate change. This scenario is consistent with the future distribution model for this species as *T. scalaris* suffered the biggest reduction of the five species. The fact that its distribution already includes the existing areas with the highest altitude implies that, as climate change progresses, this species will be limited in its ability to shift its distribution upward, increasing the possibility of becoming extinct (*Sunny, González-Fernández & D'Addario, 2017*). According to the International Union for Conservation of Nature (IUCN), *T. scalaris* is considered a species of Least Concern (*Canseco-Márquez & Mendoza-Quijano, 2007*); however, our results suggest that this risk category is likely to change in the future.

Present and future potential distribution

The potential distribution of all species was located at high elevation areas (mountain ranges), which is consistent with the biology of this genus (*Rossman, Ford & Seigel, 1996*). The fact that all species had a high proportion of their potential distribution in the TMVB means that this is a very important biogeographic region for the conservation of these five *Thamnophis* species. This is especially applicable for *T. scalaris* and *T. scaliger* as their suitable habitat was mainly found in the TMVB. However, according to our models, all species will suffer large reductions in their potential distribution in the TMVB, while in other regions some species like *T. eques* and *T. melanogaster* will not. This may be because the TMVB is one of the most disturbed regions as it contains the largest extent of urban area in Mexico (*CONAPO, 2010*). Considering the entire country, all species will suffer reductions in their potential distributions in the future except *T. melanogaster*. We are surprised by this fact, as this species is the most threatened of the five, according to the IUCN (Endangered, *Vázquez Díaz & Quintero Díaz, 2007*). This species is more aquatic than the others (*Manjarrez & Drummond, 1996*) and so an approach that considers both macrohabitat and microhabitat variables (such as water source and quality) may be necessary for a better prediction of *T. melanogaster* potential distribution.

Conclusions and conservation implications

Arid vegetation has an important negative effect on habitat suitability for all species, and the minimum temperature of the coldest month is important for four of the five species. *T. cyrtopsis* has the lowest tolerance to minimum temperatures. Maximum temperature of the warmest month is important for *T. scalaris* and *T. cyrtopsis*. Low percentages of agricultural areas are positive for *T. eques* and *T. melanogaster* but at higher values agriculture has a negative effect on habitat suitability for both species. Elevation is the most important variable to explain *T. eques* and *T. melanogaster* potential distribution while distance to *Abies* forests is the most important variable to explain *T. scalaris* and *T. scaliger* potential distribution. As we predicted, all *Thamnophis* species will experience reductions in their distributions in the TMVB, however, for the entire country, *T. melanogaster* seems to increase its distribution in the future. We feel more studies should be conducted to evaluate *T. melanogaster* distribution and abundance. These studies should consider microhabitat variables such as water source and their quality. We also consider it essential to carry out studies of *T. scalaris* abundance, as this species will suffer the biggest reduction in potential distribution of the five species. Current abundance data of this species will be key to deciding if a change in its conservation status is needed. We are especially concerned about our finding that a relatively abundant species like *T. scalaris* may suffer severe reductions in its potential distribution, as this suggests that reptile species with similar distributions (such as the lizard *Barisia imbricata*; *Sunny, González-Fernández & D'Addario, 2017*) may undergo similar reductions. For less abundant species with similar distributions (such as the rattlesnake *Crotalus triseriatus*; *Sunny et al., 2015*), even larger reductions may result. Reductions in suitable area available for *Thamnophis* and other species will cause the reduction and isolation of their populations. Small populations are susceptible to demographic stochasticity (*Gibbs, 1998*; *Hicks & Pearson, 2003*) that can convert normal population fluctuations into local extinctions (*Gibbs, 1998*). Moreover, while certain isolation levels between populations may facilitate precise evolutionary adaptations to local conditions (*Tscharntke et al., 2012*), the high isolation levels affecting populations of many species in the TMVB, which are expected to increase in the future and that are limiting species distributions to the highest altitudes of the volcanoes, will lead to important losses of genetic diversity in these populations, thereby affecting their capacity to cope with environmental changes and increasing their susceptibility to extinction (*Johansson, Primmer & Merila, 2006*; *Sunny et al., 2014*).

The TMVB has the highest area of *Abies* forests (91.14%) of the country (*Sunny, González-Fernández & D'Addario, 2017*); however, it only represents 1.10% of TMBV area (*Sunny, González-Fernández & D'Addario, 2017*). Unfortunately, governmental laws have recently changed the protection status of some areas of the TMVB, like the Nevado de Toluca Volcano (*DOF, 2013*). This change could lead to logging and changes in land use (*Mastretta-Yanes et al., 2014*). The extent of *Abies* forests have held steady from 2002 to 2011 (Fig. 2) but we are afraid this could change as a consequence of this new protection status, thus affecting *Thamnophis* populations and many other species. Moreover, land use changes are expected to accelerate due to climate change (*Maclean & Wilson, 2011*; *Urban, 2015*; *Nadeau, Urban & Bridle, 2017*) so garter snakes and other species of the TMVB could suffer the synergistic effect of both factors. The process of reversing climate change involves world-wide economic systems and government decisions, so there is little we can say about this here. However, we consider the conservation of TMVB forests, especially *Abies* forests and grasslands associated with them, of great importance for the conservation of many reptile and amphibian species that live in this region (*Figueroa-Rangel, Willis & Olvera-Vargas, 2010*; *Vargas-Rodríguez et al., 2010*; *Ponce-Reyes et al., 2012*; *Bryson et al., 2014*). Moreover, in the short term, we think is essential to implement environmental education activities to teach everyone the importance of our natural environments. This in turn may lead to fewer reptiles being killed out of fear, and use of fewer wildlife-destructive agricultural practices such as roller chopping and crop burning.

ACKNOWLEDGEMENTS

AGF is grateful to the graduate program "Doctorado en Ciencias Agropecuarias y Recursos Naturales" of the Autonomous University of the State of Mexico and to the Consejo Nacional de Ciencia y Tecnología. We thank Ruthe Smith for valuable comments and English review. We thank the editor and two anonymous reviewers for their comments.

ADDITIONAL INFORMATION AND DECLARATIONS

Funding

This study was supported by a research grant from the Universidad Autónoma del Estado de México (4047/2016SF), and Andrea González-Fernández received scholarships from CONACYT and UAEMEX. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Grant Disclosures

The following grant information was disclosed by the authors:
Universidad Autónoma del Estado de México: 4047/2016SF.
CONACYT.
UAEMEX.

Competing Interests

The authors declare there are no competing interests.

Author Contributions

- Andrea González-Fernández and Armando Sunny conceived and designed the experiments, performed the experiments, analyzed the data, contributed reagents/materials/analysis tools, prepared figures and/or tables, authored or reviewed drafts of the paper, approved the final draft.
- Javier Manjarrez conceived and designed the experiments, contributed reagents/materials/analysis tools, authored or reviewed drafts of the paper, approved the final draft.
- Uri García-Vázquez contributed reagents/materials/analysis tools, prepared figures and/or tables, authored or reviewed drafts of the paper, approved the final draft.
- Maristella D'Addario conceived and designed the experiments, authored or reviewed drafts of the paper, approved the final draft.

Data Availability

The following information was supplied regarding data availability:

The raw data is provided as a Supplemental File.

Supplemental Information

Supplemental information for this article can be found online at http://dx.doi.org/10.7717/peerj.4618#supplemental-information.

REFERENCES

Anderson RP, González Jr I. 2011. Species-specific tuning increases robustness to sampling bias in models of species distributions: an implementation with Maxent. *Ecological Modelling* **222**:2796–2811 DOI 10.1016/j.ecolmodel.2011.04.011.

Anderson RP, Lew D, Peterson AT. 2003. Evaluating predictive models of species' distributions: criteria for selecting optimal models. *Ecological Modeling* **162**:211–232 DOI 10.1016/S0304-3800(02)00349-6.

Araujo MB, Guisan A. 2006. Five (or so) challenges for species distribution modelling. *Journal of Biogeography* **33**(**10**):1677–1688.

Bastos EGDM, De Araújo AFB, Silva HR. 2005. Records of the rattlesnakes *Crotalus durissus terrificus* (Laurenti) (Serpentes Viperidae) in the State of Rio de Janeiro Brazil: a possible case of invasion facilitated by deforestation. *Revista Brasileira de Zoologia* **22**:812–815 DOI 10.1590/S0101-81752005000300047.

Bivand R, Keitt T, Rowlingson B. 2017. rgdal: bindings for the geospatial data abstraction library. *Available at https://cran.r-project.org/web/packages/rgdal/index.html*.

**Böhm M, Collen B, Baillie JEM, Bowles P, Chanson J, Cox N, Hammerson G, Hoffmann M, Livingstone SR, Ram M, Rhodin AGJ, Stuart SN, Van Dijk PP, Young BE, Afuang LE, Aghasyan A, García A, Aguilar C, Ajtic R, Akarsu F, Alencar LRV, Allison A, Ananjeva N, Anderson S, Andrén C, Ariano-Sánchez D, Arredondo JC, Auliya M, Austin CC, Avci A, Baker PJ, Barreto-Lima AF, Barrio-Amorós

CL, Basu D, Bates MF, Batistella A, Bauer A, Bennett D, Böhme W, Broadley D, Brown R, Burgess J, Captain A, Carreira S, Castañeda MDR, Castro F, Catenazzi A, Cedeño-Vázquez JR, Chapple DG, Cheylan M, Cisneros-Heredia DF, Cogalniceanu D, Cogger H, Corti C, Costa GC, Couper PJ, Courtney T, Crnobrnja-Isailovic J, Crochet P-A, Crother B, Cruz F, Daltry JC, Daniels RJR, Das I, De Silva A, Diesmos AC, Dirksen L, Doan TM, Dodd CK, Doody JS, Dorcas ME, De Barros Filho JD, Egan VT, El Mouden EH, Embert D, Espinoza RE, Fallabrino A, Feng X, Feng Z-J, Fitzgerald L, Flores-Villela O, França FGR, Frost D, Gadsden H, Gamble T, Ganesh SR, Garcia MA, García-Pérez JE, Gatus J, Gaulke M, Geniez P, Georges A, Gerlach J, Goldberg S, Gonzalez J-CT, Gower DJ, Grant T, Greenbaum E, Grieco C, Guo P, Hamilton AM, Hare K, Hedges SB, Heideman N, Hilton-Taylor C, Hitchmough R, Hollingsworth B, Hutchinson M, Ineich I, Iverson J, Jaksic FM, Jenkins R, Joger U, Jose R, Kaska Y, Kaya U, Keogh JS, Köhler G, Kuchling G, Kumlutaş Y, Kwet A, La Marca E, Lamar W, Lane A, Lardner B, Latta C, Latta G, Lau M, Lavin P, Lawson D, Le Breton M, Lehr E, Limpus D, Lipczynski N, Lobo AS, López-Luna MA, Luiselli L, Lukoschek V, Lundberg M, Lymberakis P, Macey R, Magnusson WE, Mahler DL, Malhotra A, Mariaux J, Maritz B, Marques OAV, Márquez R, Martins M, Masterson G, Mateo JA, Mathew R, Mathews N, Mayer G, McCranie JR, Measey GJ, Mendoza-Quijano F, Menegon M, Métrailler S, Milton DA, Montgomery C, Morato SAA, Mott T, Muñoz-Alonso A, Murphy J, Nguyen TQ, Nilson G, Nogueira C, Núñez H, Orlov N, Ota H, Ottenwalder J, Papenfuss T, Pasachnik S, Passos P, Pauwels OSG, Pérez-Buitrago N, Pérez-Mellado V, Pianka ER, Pleguezuelos J, Pollock C, Ponce-Campos P, Powell R, Pupin F, Quintero Díaz GE, Radder R, Ramer J, Rasmussen AR, Raxworthy C, Reynolds R, Richman N, Rico EL, Riservato E, Rivas G, Da Rocha PLB, Rödel M-O, Schettino LR, Roosenburg WM, Ross JP, Sadek R. 2013. The conservation status of the world's reptiles. *Biological Conservation* **157**:372–385 DOI 10.1016/j.biocon.2012.07.015.

Bryson RW, Linkem CW, Dorcas ME, Lathrop A, Jones JM, Alvarado-Díaz J, Grünwald CI, Murphy RW, et al. 2014. Multilocus species delimitation in the *Crotalus triseriatus* species group (Serpentes: Viperidae: Crotalinae) with the description of two new species. *Zootaxa* **3**:475–496.

Canseco-Márquez L, Mendoza-Quijano F. 2007. *Thamnophis scalaris*, *Thamnophis scaliger*. The IUCN Red List of Threatened Species 2007. *Available at http://dx.doi.org/10.2305/IUCN.UK.2007.RLTS.T63992A12727613.en* (accessed on 20 March 2018).

Cardinale BJ, Duffy JE, Gonzalez A, Hooper DU, Perrings C, Venail P, Kinzig AP, Daily GC, Loreau M, Grace JB, Larigauderie A, Srivastava DS, Naeem S. 2012. Biodiversity loss and its impact on humanity. *Nature* **486(7401)**:59–67 DOI 10.1038/nature11148.

Castellano MJ, Valone TJ. 2006. Effects of livestock removal and perennial grass recovery on the lizards of a desertified arid grassland. *Journal of Arid Environments* **66**:87–95 DOI 10.1016/j.jaridenv.2005.10.016.

Chefaoui RM, Hortal J, Lobo JM. 2005. Potential distribution modelling, niche characterization and conservation status assessment using GIS tools: a case study of Iberian Copris species. *Biological Conservation* **122(2)**:327–338 DOI 10.1016/j.biocon.2004.08.005.

Clark Labs. 2012. *Idrisi 17: The Selva edition*. Worcester: Clark Photo Labs.

CONAPO. 2010. Delimitación de las zonas metropolitanas de México. *Available at http://www.conapo.gob.mx/en/CONAPO/Zonas_metropolitanas_2010*.

De Queiroz A, Lawson R, Lemos-Espinal JA. 2002. Phylogenetic relationships of North American garter snakes (Thamnophis) based on four mitochondrial genes: how much DNA sequence is enough? *Molecular Phylogenetics and Evolution* **22(2)**:315–329 DOI 10.1006/mpev.2001.1074.

Diario Oficial de la Federación Mexicana (DOF). 2013. Decreto que reforma, deroga y adiciona diversas disposiciones del diverso publicado el 25 de enero de 1936, por el que se declaró Parque Nacional la montaña denominada ''Nevado de Toluca'' que fue modificado por el diverso publicado el 19 de febrero de 1937. *Available at https://www.gob.mx/conanp/documentos/decreto-que-reforma-deroga-y-adiciona-diversas-disposiciones-del-diverso-por-el-que-se-declaro-parque-nacional-el-nevado-de-toluca*.

Drummond H, Macías García C. 1989. Limitations of a generalist: a field comparison of foraging snakes. *Behaviour* **108**:23–43 DOI 10.1163/156853989X00033.

Espinoza D, Ocegueda S. 2007. Introduccion. In: Luna-Vega I, Morrone JJ, Espinosa D, eds. *Biodiversidad de la Faja Volcánica Transmexicana*. México: Comisión Nacional para el Conocimiento y Uso de la Biodiversidad. Universidad Nacional Autónoma de México, 5–6.

Figueroa-Rangel BL, Willis KJ, Olvera-Vargas M. 2010. Cloud forest dynamics in the Mexican Neotropics during the last 1300 years. *Global Change Biology* **16**:1689–1704 DOI 10.1111/j.1365-2486.2009.02024.x.

Flores-Villela O, Canseco-Márquez L. 2007. In: Luna-Vega I, Morrone JJ, Espinosa D, eds. *Comisión Nacional para el ConocimieRiqueza de la herpetofauna*. México: Biodiversidad de la Faja Volcánica Transmexicananto y Uso de la Biodiversidad/Universidad Nacional Autónoma de México, 407–420.

Flores-Villela O, Canseco-Márquez L, Ochoa-Ochoa L. 2010. Geographic distribution and conservation of the herpetofauna of the highlands of Central Mexico. In: Wilson LD, Towsend JH, Johnson JD, eds. *Conservation of mesoamerican amphibians and reptiles*. Utah: Eagle Mountain Publishing Co., 303–321.

Food and Agriculture Organization (FAO). 1993. Forest resources assessment 1990 Tropical countries. Forestry paper No. 112. Rome, FAO.

Gibbs JP. 1998. Distribution of woodland amphibians along a forest fragmentation gradient. *Landscape Ecology* **13**:263–268 DOI 10.1023/A:1008056424692.

Guisan A, Thuiller W. 2005. Predicting species distribution: offering more than simple habitat models. *Ecology Letters* **8**:993–1009 DOI 10.1111/j.1461-0248.2005.00792.x.

Guisan A, Zimmerman NE. 2000. Predictive habitat distribution models in ecology. *Ecological Modelling* **135**:147–186 DOI 10.1016/S0304-3800(00)00354-9.

Halffter G, Morrone JJ. 2017. An analytical review of Halffter's Mexican transition zone, and its relevance for evolutionary biogeography, ecology and biogeographical regionalization. *Zootaxa* **4226**:1–46 DOI 10.11646/zootaxa.4226.1.1.

Hammerson GA. 2013. *Thamnophis cyrtopsis*. The IUCN Red List of Threatened Species 2013. *Available at http://dx.doi.org/10.2305/IUCN.UK.2013-2.RLTS.T63975A3132338.en* (accessed on 20 March 2018).

Hammerson GA, Vázquez Díaz J, Quintero Díaz GE. 2007. *Thamnophis eques*. The IUCN Red List of Threatened Species 2007. *Available at http://dx.doi.org/10.2305/IUCN.UK.2007.RLTS.T63977A12732882.en* (accessed on 20 March 2018).

Hicks NG, Pearson SM. 2003. Salamander diversity and abundance in forests with alternative land use histories in the Southern Blue Ridge Mountains. *Forest Ecology and Management* **177**:117–130 DOI 10.1016/S0378-1127(02)00319-5.

Hijmans RJ. 2016. raster: geographic data analysis and modeling. *Available at https://cran.r-project.org/package=raster* (accessed on 4 June 2017).

Hijmans RJ, Cameron SE, Parra JL, Jones PG, Jarvis A. 2005. Very high resolution interpolated climate surfaces for global land areas. *International Journal of Climatology* **25**:1965–1978.

Huey RB. 1982. Temperature, physiology, and the ecology of reptiles. In: Gans C, Pough FH, eds. *Biology of the reptilia. Physiology C. Physiological ecology*. Vol. 12. New York: Academic Press, 25–91.

Instituto Nacional de Estadística Geográfica e Informática (INEGI). 2005. *Conjunto Nacional de Uso del Suelo y Vegetación a escala 1:250,000, Serie III.* México: DGG-INEGI.

INEGI, National Institute for Statistics and Geography. 2013. *Conjunto nacional de uso de suelo y vegetación a escala 1:250,000 Serie V*. México: DGG-INEGI.

Jimenez-Valverde A, Lobo JM. 2007. Threshold criteria for conversion of probability of species presence to either—or presence–absence. *Acta Oecologica* **31**:361–369 DOI 10.1016/j.actao.2007.02.001.

Johansson M, Primmer CR, Merila J. 2006. History vs current demography: explaining the genetic population structure of the common frog (*Rana temporaria*). *Molecular Ecology* **15**:975–983 DOI 10.1111/j.1365-294X.2006.02866.x.

Jones KB. 1990. Habitat use and predatory behavior of *Thamnophis cyrtopsis* (serpenes: colubridae) in a seasonally variable aquatic environment. *The Southwestern Naturalist* **35(2)**:115–122 DOI 10.2307/3671531.

Kortsch S, Primicerio R, Fossheim M, Dolgov AV, Aschan M. 2015. Climate change alters the structure of arctic marine food webs due to poleward shifts of boreal generalists. *Proceedings of the Royal Society B: Biological Sciences* **282(1814)**:20151546 DOI 10.1098/rspb.2015.1546.

Liu C, White M, Newell G. 2013. Selecting thresholds for the prediction of species occurrence with presence-only data. *Journal of Biogeography* **40(4)**:778–789 DOI 10.1111/jbi.12058.

Lobo JM, Jiménez-Valverde A, Real R. 2008. AUC: misleading measure of the performance of predictive distribution models. *Global Ecology and Biogeography* **17**:145–151 DOI 10.1111/j.1466-8238.2007.00358.x.

Maclean IM, Wilson RJ. 2011. Recent ecological responses to climate change support predictions of high extinction risk. *Proceedings of the National Academy of Sciences of the United States of America* **108(30)**:12337–12342 DOI 10.1073/pnas.1017352108.

Manjarrez J. 1998. Ecology of the Mexican Garter Snake (*Thamnophis eques*) in Toluca, Mexico. *Journal of Herpetology* **23**:464.

Manjarrez J, Drummond H. 1996. Temperature-limited activity in the garter snake *Thamnophis melanogaster* (Colubridae). *Ethology* **102**:146–156.

Manjarrez J, Venegas-Barrera CS, García-Guadarrama T. 2007. Ecology of the Mexican alpine blotched garter snake (*Thamnophis Scalaris*). *The Southwestern Naturalist* **52(2)**:258–262 DOI 10.1894/0038-4909(2007)52[258:EOTMAB]2.0.CO;2.

Mastretta-Yanes A, Moreno-Letelier A, Piñero D, Jorgensen TH, Emerson BC. 2015. Biodiversity in the Mexican highlands and the interaction of geology, geography and climate within the Trans-Mexican Volcanic Belt. *Journal of Biogeography* **42(9)**:1586–1600 DOI 10.1111/jbi.12546.

Mastretta-Yanes A, Quadri-Barba P, Escalante T, Arredondo-Amezcua L, Piñero D. 2014. Propuesta de cambios a la zonificación y modificaciones al Programa de Manejo del APFF Nevado de Toluca tras reunión de discusión con CONANP en diciembre 2013. *Available at https://nevadodetoluca.files.wordpress.com/2014/07/propuestas_a_conanp_nevadotoluca_enero2014.pdf*.

Merow C, Smith MJ, Silander JA. 2013. A practical guide to MaxEnt for modeling species' distributions: what it does, and why inputs and settings matter. *Ecography* **36**:1058–1069 DOI 10.1111/j.1600-0587.2013.07872.x.

Mociño-Deloya E, Setser K, Pérez-Ramos E. 2014. Observations on the diet of *Crotalus triseriatus* (*Mexican dusky rattlesnake*). *Revista Mexicana de Biodiversidad* **85**:1289–1291 DOI 10.7550/rmb.43908.

Montoya JM, Pimm SL, Solé RV. 2006. Ecological networks and their fragility. *Nature* **442**:259–264 DOI 10.1038/nature04927.

Mullin SJ, Seigel RA. 2009. *Snakes: ecology and conservation*. Ithaca: Cornell University Press.

Nadeau CP, Urban MC, Bridle JR. 2017. Coarse climate change projections for species living in a fine-scaled world. *Global Change Biology* **23(1)**:12–24 DOI 10.1111/gcb.13475.

Osorio-Olvera L. 2016. NicheToolbox: a web tool for exploratory data analysis and niche modeling. *Available at http://shiny.conabio.gob.mx:3838/nichetoolb2/* (accessed on 18 May 2017).

Parmesan C, Yohe G. 2003. A globally coherent fingerprint of climate change impacts across natural systems. *Nature* **421(6918)**:37–42 DOI 10.1038/nature01286.

Pearson RG. 2007. Species distribution modelling for conservation educators and practitioners. *Bulletin of the American Museum of Natural History* **3**:54–89.

Peterson AT, Papes M, Soberón J. 2008. Rethinking receiver operating characteristic analysis applications in ecological niche modelling. *Ecological Modelling* **213**:63–72 DOI 10.1016/j.ecolmodel.2007.11.008.

Peterson CR, Gibson AR, Dorcas ME. 1993. Snake thermal ecology: the causes and consequences of body-temperature variation. In: Siegel RA, Collins JT, eds. *Snakes. Ecology & behavior*. New York: McGraw-Hill, 241–314.

Phillips SJ, Anderson RP, Schapire RE. 2006. Maximum entropy modeling of species geographic distributions. *Ecological Modelling* **190**:231–259 DOI 10.1016/j.ecolmodel.2005.03.026.

Ponce-Reyes R, Reynoso-Rosales VH, Watson JEM, VanDerWal J, Fuller RA, Pressey RL, Possingham HP. 2012. Vulnerability of cloud forest reserves in Mexico to climate change. *Nature Climate Change* **2**:448–452 DOI 10.1038/nclimate1453.

R Development Core Team. 2017. R: a language and environment for statistical computing. Vienna: R Foundation for Statistical Computing. *Available at http://www.r-project.org* (accessed on 1 May 2017).

Reinert HK, MacGregor GA, Esch M, Bushar LM, Zappalorti RT. 2011. Foraging ecology of timber rattlesnakes *Crotalus horridus*. *Copeia* **3**:430–442.

Ribeiro R, Santos X, Sillero N, Carretero MA, Llorente GA. 2009. Biodiversity and land uses at a regional scale: is agriculture the biggest threat for reptile assemblages? *Acta Oecologica* **35**:327–334 DOI 10.1016/j.actao.2008.12.003.

Rossman DA, Ford NB, Seigel RA. 1996. *The garter snakes: evolution and ecology*. Norman: University of Oklahoma Press.

Russildi G, Arroyo-Rodríguez V, Hernández-Ordóñez O, Pineda E, Reynoso VH. 2016. Species-and community-level responses to habitat spatial changes in fragmented rainforests: assessing compensatory dynamics in amphibians and reptiles. *Biodiversity and Conservation* **25(2)**:375–392 DOI 10.1007/s10531-016-1056-3.

Sáenz-Romero C, Rehfeldt GE, Duval P, Lindig-Cisneros RA. 2012. *Abies religiosa* habitat prediction in climatic change scenarios and implications for monarch butterfly conservation in Mexico. *Forest Ecology and Management* **275**:98–106 DOI 10.1016/j.foreco.2012.03.004.

Schneider-Maunoury L, Lefebvre V, Ewers RM, Medina-Rangel GF, Peres CA, Somarriba E, Pfeifer M. 2016. Abundance signals of amphibians and reptiles indicate strong edge effects in Neotropical fragmented forest landscapes. *Biological Conservation* **200**:207–215 DOI 10.1016/j.biocon.2016.06.011.

Seigel RA. 1996. Ecology and conservation of garter snakes: masters of plasticity. In: Rossman DA, Ford NB, Seigel RA, eds. *The garter snakes. Evolution and ecology*. Norman: University of Oklahoma Press, 55–89.

Suárez-Atilano M. 2015. Filogeografía de *Boa Constrictor* (Serpentes: Boidae) en México y Centro América. PhD Thesis, Universidad Nacional Autónoma de México. Posgrado en Ciencias Biológicas. Instituto de Ecología. *Available at http://bibliotecacentral.unam.mx/* (accessed on 16 June 2017).

Suárez-Atilano M, Burbrink F, Vázquez-Domínguez E. 2014. Phylogeographical structure within Boa constrictor imperator across the lowlands and mountains of Central America and Mexico. *Journal of Biogeography* **41**:2371–2384 DOI 10.1111/jbi.12372.

Suárez-Atilano M, Rojas-Soto O, Parra JL, Vázquez-Domínguez E. 2017. The role of environment on the genetic divergence between two Boa imperator lineages. *Journal of Biogeography* **44(9)**:2045–2056 DOI 10.1111/jbi.13006.

Sunny A, González-Fernández A, D'Addario M. 2017. Potential distribution of the endemic imbricate alligator lizard (*Barisia imbricata imbricata*) in highlands of central Mexico. *Amphibia-Reptilia* **38(2)**:225–231 DOI 10.1163/15685381-00003092.

Sunny A, Monroy-Vilchis O, Fajardo V, Aguilera-Reyes U. 2014. Genetic diversity and structure of an endemic and critically endangered stream river salamander (Caudata: *Ambystoma leorae*) in Mexico. *Conservation Genetics* **15**:49–59 DOI 10.1007/s10592-013-0520-9.

Sunny A, Monroy-Vilchis O, Zarco-González MM, Mendoza-Martínez GD, Martínez-Gómez D. 2015. Genetic diversity and genetic structure of an endemic Mexican Dusky Rattlesnake (*Crotalus triseriatus*) in a highly modified agricultural landscape: implications for conservation. *Genetica* **143**:705–716 DOI 10.1007/s10709-015-9868-8.

Tscharntke T, Tylianakis JM, Rand TA, Didham RK, Fahrig L, Batary P, Bengtsson J, Clough Y, Crist TO, Dormann CF, Ewers RM, Jochen F, Holt RD, Holzschuh, Klein AM, Klejin D, Kremen C, Landis DA, Laurance W, Lindenmayer D, Scherber C, Sodhi N, Steffan-Dewenter I, Thies C, Van der Putten WH, Westhpal C. 2012. Landscape moderation of biodiversity patterns and processes eight hypotheses. *Biological Reviews* **87(3)**:661–685 DOI 10.1111/j.1469-185X.2011.00216.x.

Urban MC. 2015. Accelerating extinction risk from climate change. *Science* **348(6234)**:571–573 DOI 10.1126/science.aaa4984.

Uribe-Peña AZ, Ramirez-Bautista A, Casas-Andreu G. 1999. *Anfibios y reptiles de las serranías del Distrito Federal, México.* Cuadernos del Instituto de Biología 32 Universidad Nacional Autonoma de México.

Vargas-Rodríguez YL, Platt WJ, Vázquez-García JA, Boqiun G. 2010. Selecting relict montane cloud forests for conservation priorities: the case of western Mexico. *Natural Areas Journal* **30**:156–174 DOI 10.3375/043.030.0204.

Vázquez Díaz J, Quintero Díaz GE. 2007. *Thamnophis melanogaster*. The IUCN Red List of Threatened Species 2007. *Available at http://dx.doi.org/10.2305/IUCN.UK.2007.RLTS.T63982A12725746.en* (accessed on 20 March 2018).

Venegas-Barrera CS, Manjarrez J. 2011. Spatial patterns of species richness of garter snakes *Thamnophis* in Mexico. *Revista Mexicana de Biodiversidad* **82**:179–191.

Wittenberg RD. 2012. Foraging ecology of the Timber Rattlesnake (*Crotalus horridus*) in a fragmented landscape. *Herpetological Conservation and Biology* **7**:449–461.

IV. CAPÍTULO 2 (SEGUNDO ARTÍCULO)

Este artículo fue enviado a la revista *Landscape Ecology* el 16 de Noviembre de 2018.

Local and landscape drivers of the abundance and genetic diversity of a microendemic and critically endangered salamander

Andrea González-Fernández · Víctor Arroyo-Rodríguez · Fabiola Ramírez-Corona · Javier Manjarrez · Arlene Aguilera-Hernández · Armando Sunny

Andrea González-Fernández (corresponding author) · Javier Manjarrez

Facultad de Ciencias, Universidad Autónoma del Estado de México, Instituto literario 100, Colonia Centro, 50000 Toluca, Estado de México, Mexico. E-mail: andreagofe@gmail.com; phone number: +52 1 7223032914.

Víctor Arroyo-Rodríguez

Instituto de Investigaciones en Ecosistemas y Sustentabilidad, Universidad Nacional Autónoma de Mexico. Antigua Carretera a Pátzcuaro 8701. Ex-Hacienda de San José de la Huerta, 58190 Morelia, Michoacán, Mexico.

Fabiola Ramírez-Corona

Taller de Sistemática y Biogeografía, Departamento de Biología Evolutiva, Facultad de Ciencias, Universidad Nacional Autónoma de México, Circuito Exterior s/n, Coyoacán, Cd. Universitaria, 04510 Mexico City, Mexico.

Arlene Aguilera-Hernández · Armando Sunny

Centro de Investigación en Ciencias Biológicas Aplicadas, Universidad Autónoma del Estado de México, Instituto literario 100, Colonia Centro, 50000 Toluca, Estado de México, Mexico.

Date of the manuscript draft: 11/16/2018

Manuscript word count: 6089

Acknowledgments

We thank IGECEM for the SPOT images and Francisco Reyna-Sáenz, Fabiola Judith Gandarilla-Aizpuro and Carmen Galán-Acedo for their help with SPOT images processing. A.G.-F. obtained a scholarship from CONACyT.

Abstract

Context Conversion of forest ecosystems to human-modified landscapes threatens the persistence of forest-specialist species. Yet, the local and landscape drivers of the abundance and genetic diversity of these species are largely unknown, especially for elusive and critically endangered species, such as the salamander *Pseudoeurycea robertsi* – a species microendemic to the Nevado de Toluca volcano, Mexico.

Objectives To assess the relative influence of local- and landscape-scale habitat amount and habitat spatial configuration on the abundance and genetic diversity of *P. robertsi.* Given the low vagility of the species, we expected stronger responses to local habitat amount than to landscape variables, with habitat configuration showing the weakest effects on all responses.

Methods Using multiscale and multimodel inference approaches, we tested the relative effect of local habitat amount (fallen logs volume), landscape habitat amount (forest cover) and landscape configuration (forest edge density and forest fragmentation per se) on the abundance and genetic diversity of *P. robertsi.*

Results The abundance of individuals was more strongly related to local and landscape variables than genetic diversity. As predicted, local habitat amount showed stronger positive effects on salamander abundance and number of alleles than forest cover. Yet, all response variables also increased in landscapes with higher fragmentation and lower edge density (i.e., larger core areas).

Conclusions Fallen logs volume is a major driver of this (and potentially others) forest-specialist salamanders. Yet, landscape configuration also shapes salamander populations, especially the number of individuals. Retaining fallen logs in forests and increasing forest core areas are critical for salamander conservation.

Keywords: Plethodontidae, splitting index, Trans-Mexican Volcanic Belt, genetic diversity.

Introduction

Productive activities such as agriculture, cattle ranching, timber extraction, and urbanization have transformed a large proportion of the planet's land surface (Foley et al. 2005). In fact, deforestation between 2000 and 2012 resulted in the loss of 2.3 million km^2 of tree cover (Hansen et al. 2013). These land use changes are threatening the maintenance of global biodiversity (Newbold et al. 2016), and is causing rapid changes in the composition and configuration of terrestrial landscapes (Fahrig et al. 2011). Landscape changes are also typically followed by a number of local disturbances, such as edge effects (i.e., biotic and abiotic changes at habitat edges) and resource exploitation (e.g., hunting, logging) by human populations (Laurance et al. 2002; Tuff et al. 2016; Arroyo-Rodríguez et al. 2017). These local and landscape disturbances are expected to have stronger effects on forest-specialist species (Laurance 1991; Kouki 2001; Tuff et al. 2016; Pfeifer et al. 2017), but this topic remains poorly understood, especially for elusive species, such as most amphibians.

Amphibians are among the most threatened vertebrates on Earth (Catenazzi 2015), and their populations are rapidly declining worldwide, mainly due to the loss and degradation of their natural habitats (Stuart et al. 2004; Eigenbrod et al. 2008).

Salamanders, like other low-vagility ectotherms, are highly vulnerable to local-scale disturbances, such as the loss of thermally suitable habitat (Nowakowski et al. 2018). Salamanders are important as top-down controls of many invertebrates, and can also be a source of high energy prey for other predators (Davic and Welsh 2004). They can also provide an important indirect regulatory role in the processing of detritus-litter by ingestion of detritivore prey (Davic and Welsh 2004). Although many species are narrowly distributed, salamanders can represent an important proportion of the vertebrate biomass in old-growth forests (Davic and Welsh 2004). In temperate ecosystems, terrestrial salamanders prefer areas with high relative moisture like *Abies*, *Pinus* and *Pinus-Quercus* forests, which provide important microhabitats, such as fallen logs, which are particularly used by the Plethodontidae family for shelter (Sánchez-Jasso et al. 2013). Landscape-scale disturbances can also shape amphibian communities (e.g., Skelly et al. 1999; Lowe and Bolger 2002; Van Buskirk 2005; Russildi et al. 2016), but the relative effect of local vs. landscape disturbances on salamanders is not well understood. In fact, some studies of salamanders assess the association between landscape structure and population genetics (Spear et al. 2005; Wang et al. 2009; Savage et al. 2010; Velo-Antón et al. 2013), but they are focused on genetic responses to landscape connectivity, overlooking other potential local and landscape predictors of genetic diversity, such as habitat amount. Filling this gap of information is critically needed to improve conservation strategies.

Mexico is within the five biologically richest countries in the world (Groombridge and Jenkins 2000), but it is suffering rapid and extensive land use changes (Mas et al. 2004), especially for the expansion of agricultural lands (Arredondo-León et al. 2008). Between 1976 and 2000, more than 20,000 km^2 of temperate forests were cleared in

Mexico (annual deforestation rate = 0.25%; Mas et al. 2004). Temperate forests are distributed in high elevation areas, such as the Trans-Mexican Volcanic Belt, which preserves one of the most species-rich herpetological communities in Mexico, and the most important in terms of endemic amphibian species (Flores-Villela and Canseco-Márquez 2007; Sunny et al. 2017). Unfortunately, it is one of the most disturbed regions of the country due to the expansion of big cities, such as Mexico City, Puebla and Toluca (CONAPO 2010; González-Fernández et al. 2018).

Here, we assessed the relative influence of local- and landscape-scale habitat amount and habitat spatial configuration (i.e., forest fragmentation per se and forest edge density) on the abundance and genetic diversity of *Pseudoeurycea robertsi*. This little-known salamander of the Plethodontidae family is classified as Critically Endangered, and is microendemic to the Nevado de Toluca volcano (SEMARNAT 2010; IUCN SSC Amphibian Specialist Group 2016), located in the Trans-Mexican Volcanic Belt. This species inhabits fallen logs, where it finds refuge and food (Bille 2009). Given its low vagility, we expect stronger responses to local habitat amount (i.e., fallen logs volume) than to landscape structure (Miguet et al. 2016). Yet, as this is a forest-specialist species (Davic and Welsh 2004), when assessing the effect of landscape-scale patterns, we expect stronger responses to forest cover (a proxy of landscape-scale habitat amount) than to forest spatial configuration (Fahrig 2003; Jackson and Fahrig 2016). If forest fragmentation is relevant for this species, we could expect either positive or negative effects. Positive effects are predicted because mean inter-patch isolation distance typically decreases with increasing fragmentation per se, thus favoring dispersal movements across the landscape (Fahrig 2003, 2017; Jackson and Fahrig 2016). Yet, fragmentation also increases edge-affected habitats in the landscape, and this forest-interior specialist can be negatively affected by the abiotic changes (e.g. lower relative humidity and higher temperature) that usually occur at forest edges (Arroyo-Rodríguez et al. 2017). In this sense, we predicted that forest edge density can have negative effects on the abundance and genetic diversity of salamanders.

Methods

Study area

We conducted this study in the Nevado de Toluca volcano (18°59’- 19°18’N, 99°40’- 99°59’ W), the fourth highest peak in Mexico (Bille 2009), located in the Trans-Mexican Volcanic Belt at 22 km of southwest Toluca city. The region has a temperate semi-cold climate, with precipitations during the summer and an average annual temperature ranging between 5 and 12 °C. Annual precipitation averages 1200-1800 mm (CONABIO 2000). The dominant land cover types of the volcano are old-growth and secondary forest patches of fir (*Abies religiosa*) and pine (*Pinus hartwegii* and *P. pseudostrobus*). There are also broad-leaved forests (oak and alder) in a lower extent, at the eastern part of the volcano, and alpine grasslands at the highest elevations. There is an important extension of agricultural lands in addition to induced grasslands and human settlements (Franco-Maass et al. 2006). Between 1972 and 2000, *Abies* forests experienced an important recovery, whereas *Pinus* forest cover had decreased by 40% due to timber extraction for commercial purposes (Franco-Maass et al. 2006). The Nevado de Toluca volcano is considered a priority terrestrial region for biodiversity conservation (CONABIO 2000), and was declared a National Park in 1936, although the Mexican Government has recently changed this highly restrictive protection category to a less restrictive one (DOF 2013) – a controversial decision that could lead to further degradation of the last well-preserved *Abies* forests (Mastretta-Yanes et al. 2014).

Experimental design

We sampled 14 forest sites distributed across the volcano, all located in *Abies-Pinus* forest, between 2850 and 3450 m asl (Fig. 1). We used a site-landscape approach; i.e., response variables were recorded in equal-sized sample sites, and landscape variables were measured within different-sized and concentric buffers from the geographic center of these sample sites (Fahrig 2013). We considered buffers of 10, 50, 100, 200, 300, 400 and 500 ha to assess the scale of effect, i.e., the spatial extent that yields the strongest response-landscape relationship (Fahrig 2013; Jackson and Fahrig 2015; Miguet et al. 2016). The smallest landscape size (10 ha) was selected because salamanders have low dispersion capacity (Petranka et al. 1993), which can decrease the scale of effect (Miguet et al. 2016). To avoid pseudoreplication problems, we located each study site isolated enough to avoid spatial overlap between landscapes (Eigenbrod et al. 2011; Fig. 1).

Salamander sampling and response variables

We sampled salamanders via visual encounter surveys (sensu Crump and Scott 1994). As there is no information on the timing of peak of *Preudoeurycea robertsi* abundance, we carry out a pilot study to identify the annual season with higher abundance of salamanders in *Abies* and *Pinus* forests. In particular, we surveyed three study sites from April to October 2015 and visited each site once per month. We did not record any individual in April and October. In May and September the abundance of individuals was two times lower than in June-August. We therefore recorded the abundance of individuals in all 14 sites from mid-June to mid-August 2016. In particular, during each visit 2 people looked for salamanders under the bark of fallen logs with a diameter of ≥ 5 cm and length of ≥ 30 cm. We started at 9:00 am and spent 2 effective hours of search (i.e., excluding stops), covering an area of about 10 ha in each site (the size of our smallest landscape).

The genetic survey was carried out at the same time, but we re-visited some sites in September to collect additional genetic samples, but in these extra-surveys we did not record individual abundances. We sampled 2 mm of tail tips of adult salamanders for DNA extraction (license number SGPA/DGVS/05701/16). This methodology is a low-invasive method that does not affect the survival or growth of salamanders (Arntzen et al. 1999; Polich et al. 2013). We released all individuals immediately after data collection in the same place where they were found.

Tail tissues were preserved in 90% ethanol and then frozen at -20 °C until processed. We extracted the DNA according to the commercial kit protocol "GF-1 Tissue DNA" of "Vivantis" mark. We used nine fluorescently labelled microsatellite primers developed for *Pseudoeurycea leprosa* by Velo-Antón et al. (2009). We multiplexed and run PCR microsatellite products on an ABI Prism3730xl (Applied Biosystems), with Rox-500 as an internal size standard by a commercial laboratory (Roy. J. Carver Biotechnology Center in Illinois University, USA). We obtained allele sizes with PEAKSCANNER 1.0 software (Applied Biosystems) and the fragment lengths with TANDEM 1.08 (Matschiner and Salzburger 2009). To avoid wrong data interpretation, we tested the presence of null alleles and other typing errors in the software MICROCHECKER 2.2.3 (Van Oosterhout et al. 2004). In POPPR 2.4.1 (Kamvar et al. 2014) for the R software (version 3.4.0; R

Development Core Team 2017), we made an analysis to create a genotype accumulation curve, useful to determining the minimum number of loci necessary to discriminate between individuals in a population (Kamvar et al. 2014). This function will randomly sample loci without replacement and count the number of observed multilocus genotypes (Kamvar et al. 2014). The genotype accumulation curve showed that the minimum number of loci necessary to discriminate between individuals in this population is eight (Appendix Fig. A1 in Supplementary Material). Therefore, we can assume that our study has enough microsatellite primers (N = 9).

We used up to 16 samples per site (150 samples in total) but in some sites we found less than 16 individuals. We finally excluded 3 forest sites where we found less than 4 genetic samples. To estimate the accuracy of allelic accounts we used the coverage estimator recommended by Chao and Jost (2012), which is usually used to estimate the accuracy of species inventories. Sample coverage was very high in all sites (>0.91), indicating that our sampling effort was adequate to estimate diversity metrics within each site (Chao and Jost 2012). Yet, we calculated the expected values of allele richness based on coverage extrapolations performed with the *entropart* package (Marcon and Hérault 2014) for R 3.0.1 (R Development Core Team 2014) to avoid any potential bias in our results due to differences in sample coverage among sites (Chao and Jost 2012). We calculated genetic diversity using true diversity measures (Hill 1973):

Diversity $\Delta \equiv \left(\sum_{i=1}^{k} p_i^q \right)^{1/(1-q)}$, where p_i is the population frequency of the i-th allele and the exponent q determines the measure's sensitivity to allele frequencies. When $q = 0$, the equation gives the allele number (*Na*), and when $q = 2$ it gives Kimura and Crow's (1964) effective number of alleles (*Ne*) (Jost 2008). We calculated all diversity metrics with the *entropart* package (Marcon and Hérault 2014) for R (version 3.4.0; R Development Core Team 2017). *Na* is not sensitive to allele abundances, and thus gives disproportional weight to rare alleles, while *Ne* can be interpreted as the effective number of dominant alleles in the population. These true diversity measures have the linear behavior implicit in the geneticists' concept of diversity (i.e., the ''doubling'' property sensu Hill 1973), while heterozygosity and Shannon entropy do not (Jost 2008).

Explanatory variables

Using ArcGis 10.5 software and satellite images (SPOT 6/7) of very high resolution (1.5 m) for year 2015, we made a supervised classification considering six land cover types: *Pinus* forest, *Abies* forest, grasslands, agriculture, urban areas, and water sources. Within each landscape, and at each scale, we calculated two metrics of landscape composition: (i) the percentage of remaining forest cover; and (ii) the percentage of *Abies* cover (from the total forest cover). We also evaluated two metric of landscape configuration: (iii) forest fragmentation per se, and (iv) forest edge density. We calculated forest fragmentation with the splitting index (S) proposed by Jaeger (2000): $S = A_t^2 / \sum_{i=1}^{n} A_i^2$, where A_t is the area covered by forest in the landscape and A_i is the area of the forest patch *i*. This index represents the 'effective number of forest patches' and is a measure of fragmentation per se because it is independent of the total forest cover in the landscape (Fahrig 2003). At the local scale, we also evaluated (v) fallen logs volume using the formula of the cylinder volume: $V = h\pi r^2$, where h is the sum of the length of all sampled logs in each 10-ha sample site (see above) and r is the mean radius of these logs, as fallen logs showed a similar diameter.

Data analyses

We used generalized linear models to assess the effects of local and landscape attributes on each response variable. We fixed a Gaussian error distribution for continuous response variables (expected values of Na and Ne) after verifying that model's residuals followed a Normal distribution (Shapiro-Wilk test). Abundance (count-dependent variable) was assessed by fixing a Poisson error distribution. To assess collinearity among predictor variables we estimated their variance inflation factors (VIF) using the *car* package for R version 3.0.1. A VIF > 4 suggests possible collinearity, and a VIF > 10 indicates severe collinearity (Neter et al. 1996). We found severe collinearity between forest cover (i.e., *Abies* and *Pinus* forest) and splitting index in all models. Thus, we decided to exclude forest cover from the models. We then included only *Abies* forest cover, which was not collinear with the splitting index. In fact, *Abies* forest is probably the main habitat of the species, as in our pilot survey we did not record any individual in *Pinus* forest. We used a multimodel inference approach to assess the relative effect of each predictor on each response variable (Burnham and Anderson 2002). To obtain model-averaged parameter estimates we used Akaike weights (w_i). The set of models for which $\sum w_i$ was 0.95 represents a set that has 95% probability of containing the true best model (Burnham and Anderson 2002). To be more conservative in our selection of important explanatory variables, we considered that a given predictor was important for a given response variable if accomplishing these three conditions simultaneously: (i) the predictor showed a high sum of Akaike weights (i.e., considering each candidate model in which it appeared); (ii) the model-averaged unconditional variance was lower than the model-averaged parameter estimate, and (iii) the goodness-of-fit of the model was relatively high (Russildi et al. 2016; Sánchez-de-Jesús et al. 2016). All models were built using the package *glmulti* for R version 3.0.1 (Calcagno and Mazancourt 2010).

Results

In total, we recorded 185 individuals and 33 alleles (from the 150 genotyped individuals). Both local and landscape variables showed a relatively high explanatory power, being higher for salamander abundance (75% of explained deviance) than for genetic diversity (44-56%) (Fig. 2). As predicted, local habitat amount (fallen logs volume) was relatively more important than landscape habitat amount (*Abies* forest cover), positively affecting salamander abundance (Fig. 2a) and number of alleles (Fig. 2b). Yet, contrary to our expectations, forest cover generally showed a weaker effect than forest spatial configuration, especially on salamander abundance, which responded negatively to forest edge density ($\sum w_i = 1.0$; Fig. 2a). Genetic diversity also responded negatively to increasing edge density (Fig. 2b and 2c). Although more weakly, forest fragmentation (i.e., splitting index) showed a consistently positive association with all response variables.

Discussion

This study assesses the relative effect of local- and landscape-scale habitat amount and habitat spatial configuration on the abundance and genetic diversity of *Pseudoeurycea robertsi* – a critically endangered salamander microendemic to the Nevado de Toluca volcano, Mexico. Our findings show that salamander abundance is more strongly associated with all predictor variables than genetic diversity. As expected, local habitat amount (i.e., fallen logs volume) has stronger positive effects on salamander abundance and allele number than landscape-scale habitat amount (i.e., *Abies* forest cover). Yet, contrary to our expectations, landscape-scale habitat amount shows a relatively weaker impact on salamanders than forest spatial configuration. In particular, as predicted, forest edge density is negatively related to salamander abundance, and although weaker, the effect of forest fragmentation per se tended to be positive. As discussed below, these findings have important theoretical and conservation implications.

The fact that habitat disturbance has stronger effects on salamander abundance than on genetic diversity is not surprising. The colonization or loss of a given allele in the population can require many generations, and thus, genetic diversity is expected to be regulated by a larger number of generations than abundance (Jackson and Fahrig 2014). In this sense, as most of the forested area in the study region was converted to agricultural lands in the second half of the nineteenth century (Mastretta-Yanes et al. 2014), the history of land-use change is probably not large enough to allow the full spectrum of local and landscape effects on genetic diversity be exhibited. Therefore, additional long-term monitoring studies may be necessary before we can draw stronger conclusions about the effect of habitat disturbance on the genetic diversity of this species.

As predicted, both salamander abundance and allele number were positively related to fallen logs volume. This species inhabits forest areas with high relative moisture (Sánchez-Jasso et al. 2013), especially fallen logs, where it finds refuge and food (Bille 2009). As this species has a very low vagility, we can expect that the probability that salamanders interact with spatial variables measured across larger spatial extents is relatively low (Jackson and Fahrig 2012; Miguet et al. 2016), thus explaining why species' responses to fallen logs volume were stronger than to changes in landscape structure. In fact, as fallen logs volume can be considered a proxy of habitat amount, our findings indicate that habitat loss is a major driver of population decline.

Surprisingly, and in contrast to previous studies (Fahrig 2003), landscape configuration variables (i.e., forest edge density and forest fragmentation) seems to have a higher predictive power than the percentage of *Abies* forest cover, a landscape compositional variable. We expected stronger responses to forest cover because this is a forest-specialist species (Davic and Welsh 2004). Yet, the abundance of individuals decreased mainly in forest sites surrounded by higher edge density. This can be related to the loss of 'core forest areas' in landscapes with higher edge density (Ewers and Didham 2006). This salamander species, as other ectotherms, can be negatively impacted by the abiotic changes (e.g., lower relative humidity and higher temperature) that typically occur at forest edges (Arroyo-Rodríguez et al. 2017; Nowakowski et al. 2018). Thus, as has been reported for other salamander and anuran species (deMaynadier and Hunter 1998), our findings suggest that this species can be particularly dependent on the availability of core forest areas in the landscape. In fact, changes in microclimatic conditions at forest edges, especially the loss of humidity, can have strong negative impacts on plethodontids, even stronger than on other amphibians, because salamanders from this family do not have lugs and relay on cutaneous respiration (Petranka et al. 1993; deMaynadier and Hunter 1998). Moreover, drier edge conditions also slow wood decomposition (Kapos et al. 1993), which may reduce the availability of suitable refuges for salamanders.

But, why forest fragmentation tended to have positive effects on all ecological responses? Forest fragmentation per se increases the amount of edge-affected habitats in the landscape, and thus, following our previous rationale, we can expect the opposite pattern.

However, our findings support those of Fahrig (2017), who find in a global review of fragmentation effects that this landscape variable generally shows weak effects on biodiversity, but when significant, responses to fragmentation are mostly positive. Several mechanisms can explain such positive responses, including a lower mean inter-patch isolation distance in landscapes with a higher number of forest patches, which can favor patch colonization and increase population and metapopulation persistence (Fahrig 2003, 2017; Jackson and Fahrig 2016).

Ecological and conservation implications

Our findings suggest that retaining fallen logs in the forest and increasing forest core area is critically needed to preserve *P. robertsi* populations, and potentially other forest-interior specialist species. This is consistent with previous studies that highlight the importance of leaving dead wood in the forest and avoiding clear-cutting to preserve salamanders and other forest-dwelling animals (Petranka et al. 1993; deMaynadier and Hunter 1998; Kouki 2001). The positive responses to increasing fragmentation per se also suggest that preserving all forest patches in the landscape can also be critical, as this can decrease inter-patch distances, favoring patch colonization dynamics in fragmented forests (Fahrig 2017). However, to avoid negative edge effects in these emerging landscapes, we should prevent the loss of the largest forest patches in the region, and avoid deviations from circularity in patch shapes to increase the amount of interior habitat (Ewers and Didham 2006). These management strategies need to be urgently considered given the recent change of protection level of the Nevado de Toluca volcano. Originally decreed as a National Park, this reserve was recently (2013) decreed as a Flora and Fauna Protected Area. This less restrictive category threatens the preservation of the *Abies* forest – the main habitat of *P. robertsi* –

because it allows forest harvesting practices with commercial proposes in almost all *Abies* forest extension (Mastretta-Yanes et al. 2014). Therefore, preventing the loss of *Abies* forest, especially the loss of the largest forest remnants is of paramount importance to preserve this endemic and critically endangered species.

References

Arredondo-León C, Muñoz-Jiménez J, García-Romero A (2008) Recent changes in landscape-dynamics trends in tropical highlands, central Mexico. Interciencia 33:569–577.

Arroyo-Rodriguez V, Fahrig L (2014) Why is a landscape perspective important in studies of primates? Am J Primatol 76:901–909.

Arroyo-Rodríguez V, Saldaña-Vázquez RA, Fahrig L, Santos BA (2017) Does forest fragmentation cause an increase in forest temperature? Ecol Res 32:81–88.

Arntzen JW, Smithson A, Oldham RS (1999) Marking and tissue sampling effects on body condition and survival in the newt *Triturus cristatus*. J Herpetol 33:567–576.

Bille T (2009) Field observations on the salamanders (Caudata: Ambystomatidae, Plethodontidae) of Nevado de Toluca, Mexico. Salamandra 45:155–164.

Broquet T, Ray N, Petit E, Fryxell JM, Burel F (2006) Genetic isolation by distance and landscape connectivity in the American Marten (*Martes americana*). Landscape Ecol 21:877–889.

Burnham KP Anderson DR (2002) Model selection and multimodel inference. A practical information-theoretic approach. Springer, New York.

Calcagno V, Mazancourt C (2010) glmulti: an R package for easy automated model selection with (Generalized) Linear Models. J Stat Softw 34:1–29.

Catenazzi A (2015) State of the world's amphibians. Annu Rev Environ Resour 40:91–119.

Chao A, Jost L (2012) Coverage-based rarefaction and extrapolation: standardizing samples by completeness rather than size. Ecology 93:2533–2547.

CONABIO, Comisión Nacional para el Conocimiento y Uso de la Biodiversidad (2000) Regionalización. http://www.conabio.gob.mx/conocimiento/regionalizacion/doctos/rtp_109.pdf. Accessed 5 November 2018.

CONAPO (2010) Delimitación de las zonas metropolitanas de México http://www.conapo.gob.mx/en/CONAPO/ Zonas_metropolitanas_2010. Accessed 5 November 2018.

Coulon A, Cosson JF, Angibault JM, Cargnelutti B, Galan M, Morellet N, Petit E, Aulagnier S, Hewison AJ (2004) Landscape connectivity influences gene flow in a roe deer population inhabiting a fragmented landscape: an individual-based approach. Mol Ecol 13:2841–2850.

Crump ML, Scott NJ Jr (1994) Visual encounter Surveys. In: Heyer WR, Donnelly MA, McDiarmid RW, Hayek LC, Foster MC (eds) Measuring and monitoring biological diversity: standard methods for amphibians. Smithsonian Institution Press, Washington, pp 84–92.

Cushman SA (2006) Effects of habitat loss and fragmentation on amphibians: a review and prospectus. Biol Conserv 128:231–240.

Davic RD, Welsh Jr HH (2004) On the ecological roles of salamanders. Annu Rev Ecol Evol Syst 35:405–434.

deMaynadier PG, Hunter ML (1998) Effects of silvicultural edges on the distribution and abundance of amphibians in Maine. Conserv Biol 12:340–352.

DOF, Diario Oficial de la Federación Mexicana (2013): Decreto que reforma, deroga y adiciona diversas disposiciones del diverso publicado el 25 de enero de 1936, por el que se declaró Parque Nacional la montaña denominada "Nevado de Toluca" que fue modificado por el diverso publicado el 19 de febrero de 1937. http://dof.gob.mx/nota_detalle_popup.php?codigo=5315889. Accessed 5 November 2018.

Eigenbrod F, Hecnar SJ, Fahrig L (2008) The relative effect of road traffic and forest cover on anuran populations. Biol Conserv 141:35–46.

Eigenbrod F, Hecnar SJ, Fahrig L (2011) Sub-optimal study design has major impacts on landscape-scale inference. Biol Conserv 144:298–305.

Ewers RM, Didham RK (2006) Confounding factors in the detection of species responses to habitat fragmentation. Biol Rev 81:117–142.

Fahrig L (2003) Effects of habitat fragmentation on biodiversity. Annu Rev Ecol Evol Syst 34:487–515.

Fahrig L (2013) Rethinking patch size and isolation effects: the habitat amount hypothesis. J Biogeogr 40:1649–1663.

Fahrig L (2017) Ecological responses to habitat fragmentation per se. Annu Rev Ecol Evol Syst 48:1–23.

Fahrig L, Baudry J, Brotons L, Burel FG, Crist TO, Fuller RJ, Sirami C, Siriwardena GM, Martin JL (2011) Functional landscape heterogeneity and animal biodiversity in agricultural landscapes. Ecol Lett 14:101–112.

Flores-Villela O, Canseco-Márquez L, Ochoa-Ochoa L (2010) Geographic distribution and conservation of the herpetofauna of the highlands of Central Mexico. In: Conservation of Mesoamerican Amphibians and Reptiles. Wilson LD, Towsend JH, Johnson JD. (eds.) Eagle Mountain Publishing Co. Utah, USA, pp 303–321.

Foley JA, DeFries R, Asner GP, Barford C, Bonan G, Carpenter SR, Chapin FS, Coe MT, Daily GC, Gibbs HK, Helkowski JH, Holloway T, Howard EA, Kucharik CJ, Monfreda C, Patz JA, Prentice IC, Ramankutty N, Snyder PK (2005) Global consequences of land use. Science 309:570–574.

Franco-Maass S, Regil-García HH, González-Esquivel C, Nava-Bernal G (2006) Cambio de uso del suelo y vegetación en el Parque Nacional Nevado de Toluca, México, en el periodo 1972-2000. Invest Geog 61:38–57.

Garrido-Garduño T, Vázquez-Domínguez E (2013) Métodos de análisis genéticos, espaciales y de conectividad en genética del paisaje. Rev Mex Biodivers 84:1031–1054.

González-Fernández A, Manjarrez J, García-Vázquez U, D'Addario M, Sunny A (2018) Present and future ecological niche modeling of garter snake species from the Trans-Mexican Volcanic Belt. PeerJ 6: e4618.

Groombridge B, Jenkins M (2000) Global Biodiversity. Earth's Living Resources in the 21st Century. World Conservation Monitoring Centre, Cambridge, U.K.

Hansen MC, Potapov PV, Moore R, Hancher M, Turubanova SAA, Tyukavina A, Thau D, Stehman SV, Goetz SJ, Loveland TR, Kommareddy A, Egorov A, Chini L, Justice CO, Townshend JRG (2013) High-resolution global maps of 21st-century forest cover change. Science 342: 850–853.

Helzer CJ, Jelinski DE (1999) The relative importance of patch area and perimeter-area ratio to grassland breeding birds. Ecol Appl 9:1448–1458.

Hill M (1973) Diversity and evenness: a unifying notation and its consequences. Ecology 54:427–432.

IUCN SSC Amphibian Specialist Group (2016) *Pseudoeurycea robertsi*. The IUCN Red List of Threatened Species. https://www.iucnredlist.org/species/59393/53983925. Accessed 12 February 2018.

Jackson HB, Fahrig L (2012) What size is a biologically relevant landscape? Landscape Ecol 27:929–941.

Jackson HB, Fahrig L (2015) Are ecologists conducting research at the optimal scale? Glob Ecol Biogeogr 24:52–63.

Jackson ND, Fahrig L (2014). Landscape context affects genetic diversity at a much larger spatial extent than population abundance. Ecology 95:871–881.

Jackson ND, Fahrig L (2016) Habitat amount, not habitat configuration, best predicts population genetic structure in fragmented landscapes. Landscape Ecol 31:951–968.

Jaeger JA (2000) Landscape division, splitting index, and effective mesh size: new measures of landscape fragmentation. Landscape Ecol 15:115–130.

Jost L (2008) $G_{(ST)}$ and its relatives do not measure differentiation. Mol Ecol 17:4015–4026.

Kamvar ZN, Tabima JF, Grünwald NJ (2014) Poppr: an R package for genetic analysis of populations with clonal, partially clonal, and/or sexual reproduction. PeerJ 2:e281.

Kapos V, Ganade G, Matsui E, Victoria RL (1993) ∂13C as an indicator of edge effects in tropical rainforest reserves. J Ecol 81:425–432.

Kimura M, Crow J (1964) The number of alleles that can be maintained in a finite population. Genetics 49:725–738.

Kouki J, Löfman S, Martikainen P, Rouvinen S, Uotila A (2001) Forest fragmentation in Fennoscandia: linking habitat requirements of wood-associated threatened species to landscape and habitat changes. Scand J Forest Res 16:27–37.

Laurance WF (1991) Edge effects in tropical forest fragments: application of a model for the design of nature reserves. Biol Conserv 57:205–219.

Laurance WF, Lovejoy TE, Vasconcelos HL, Bruna EM, Didham RK, Stouffer PC, Gascon C, Bierregaard RO, Laurance SG, Sampaio E (2002) Ecosystem decay of Amazonian forest fragments: a 22-year investigation. Conserv Biol 16:605–618.

Lowe WH, Bolger DT (2002) Local and landscape-scale predictors of salamander abundance in New Hampshire headwater streams. Conserv Biol 16:183–193.

Marcon E, Hérault B (2015) entropart: An R package to measure and partition diversity. http://cran.r-project.org/package=entropart.

Martin AE (2018) The spatial scale of a species' response to the landscape context depends on which biological response you measure. Curr Landscape Ecol Rep 3:23–33.

Mas J, Velázquez A, Díaz-Gallegos J, Mayorga-Saucedo R, Alcántara C, Bocco G, Castro R, Fernández T, Pérez-Vega A (2004) Assessing land use/cover changes: a nationwide multidate spatial database for Mexico. Int J Appl Earth Obs Geoinf 5:249–261.

Mastretta-Yanes A, Cao R, Nicasio-Arzeta S, Quadri P, Escalante-Espinosa T, Arredondo L, Piñero D (2014) ¿Será exitosa la estrategia del cambio de categoría para mantener la biodiversidad del Nevado de Toluca? Oikos 12: 7–17.

Matschiner M, Salzburger W (2009) TANDEM: integrating automated allele binning into genetics and genomics workflows. Bioinformatics 25:1982–1983.

Miguet P, Jackson HB, Jackson ND, Martin AE, Fahrig L (2016) What determines the spatial extent of landscape effects on species? Landscape Ecol 31:1177–1194.

Newbold T, Hudson LN, Arnell AP, Contu S, De Palma A, Ferrier S (2016) Has land use pushed terrestrial biodiversity beyond the planetary boundary? A global assessment. Science 353:288–291.

Nowakowski AJ, Watling JI, Thompson ME, Brusch GA, Catenazzi A, Whitfield SM, Kurz DJ, Suárez-Mayorga A, Aponte-Gutiérrez A, Donnelly MA, Todd BD (2018) Thermal biology mediates responses of amphibians and reptiles to habitat modification. Ecol Lett 21:345–355.

Petranka JW, Eldridge ME, Haley KE (1993) Effects of timber harvesting on southern Appalachian salamanders. Conserv Biol 7:363–377.

Pfeifer M, Lefebvre V, Peres CA, Wearn O, Marsh C, Banks-Leite C, Butchart S, Arroyo-Rodríguez V, Barlow J, Cerezo A, Cisneros L, D'Cruze N, Faria D, Hadley A, Klingbeil B, Kormann U, Lens L, Rangel GM, Morante-Filho JC, Olivier P, Peters S, Pidgeon A, Ribeiro D, Scherber C, Schneider-Maunoury L, Struebig M, Urbina-Cardona N, Watling JI, Willig M, Wood E, Ewers R (2017) Creation of forest edges has a global impact on forest vertebrates. Nature 551:187–191.

Polich RL, Searcy CA, Shaffer HE (2013) Effects of tail clipping on survivorship and growth of larval salamanders. J Wildl Manage 77:1420–1425.

R, Development Core Team (2017) R: A language and environment for statistical computing. R foundation for statistical computing, Vienna, Austria. Web Site at http://www.r-project.org).

Russildi G, Arroyo-Rodríguez V, Hernández-Ordóñez O, Pineda E, Reynoso VH (2016) Species-and community-level responses to habitat spatial changes in fragmented rainforests: assessing compensatory dynamics in amphibians and reptiles. Biodivers Conserv 25:375–392.

Sánchez-de-Jesús HA, Arroyo-Rodríguez V, Andresen E, Escobar F (2016) Forest loss and matrix composition are the major drivers shaping dung beetle assemblages in a fragmented rainforest. Landscape Ecol 31:843–854.

Sánchez-Jasso JM, Aguilar-Miguel X, Medina-Castro JP, Sierra-Domínguez G (2013) Species richness of vertebrates in a reforested woodland of the Nevado de Toluca National Park, Mexico. Rev Mex Biodivers 84:360–373.

Savage WK, Fremier AK, Bradley Shaffer H (2010) Landscape genetics of alpine Sierra Nevada salamanders reveal extreme population subdivision in space and time. Mol Ecol 19:3301–3314.

SEMARNAT (2010) Norma Oficial Mexicana NOM-059-SEMARNAT-2010, Protección ambiental-especies nativas de México de flora y fauna silvestres-Categorías de riesgo y especificaciones para su inclusión, exclusión o cambio. Lista de especies en riesgo. Diario Oficial de la Federación. http://dof.gob.mx/nota_detalle.php?codigo=5173091&fecha=30/12/2010. Accessed 5 November 2018.

Skelly DK, Werner EE, Cortwright (1999) Long-term distributional dynamics of a Michigan amphibian assemblage. Ecology 80:2326–2337.

Spear SF, Peterson CR, Matocq MD, Storfer A (2005) Landscape genetics of the blotched tiger salamander (*Ambystoma tigrinum melanostictum*). Mol Ecol 14:2553–2564.

Stuart SN, Chanson JS, Cox NA, Young BE, Rodrigues ASL, Fischman DL, Waller RW (2004) Status and trends of amphibian declines and extinctions worldwide. Science 306:1783–1786.

Sunny A, González-Fernández A, D'Addario M (2017) Potential distribution of the endemic imbricate alligator lizard (*Barisia imbricata imbricata*) in highlands of central Mexico. Amphib Reptil 38:225–231.

Temple S (1986) Predicting impacts of habitat fragmentation on forest birds: a comparison of two models. In: Verner J, Morrison M, Ralph CJ (eds) Wildlife 2000: modeling habitat relationships of terrestrial vertebrates. University of Wisconsin Press, Madison, Wisconsin, USA, pp 301–304.

Tuff KT, Tuff T, Davies KF (2016) A framework for integrating thermal biology into fragmentation research. Ecol Lett 19:361–374.

Van Buskirk J (2005) Local and landscape influence on amphibian occurrence and abundance. Ecology 86:1936–1947.

Van Oosterhout C, Hutchinson WF, Wills DPM, Shipley P (2004) MICRO-CHECKER: Software for identifying and correcting genotyping errors in microsatellite data. Mol Ecol Notes 4:535–538.

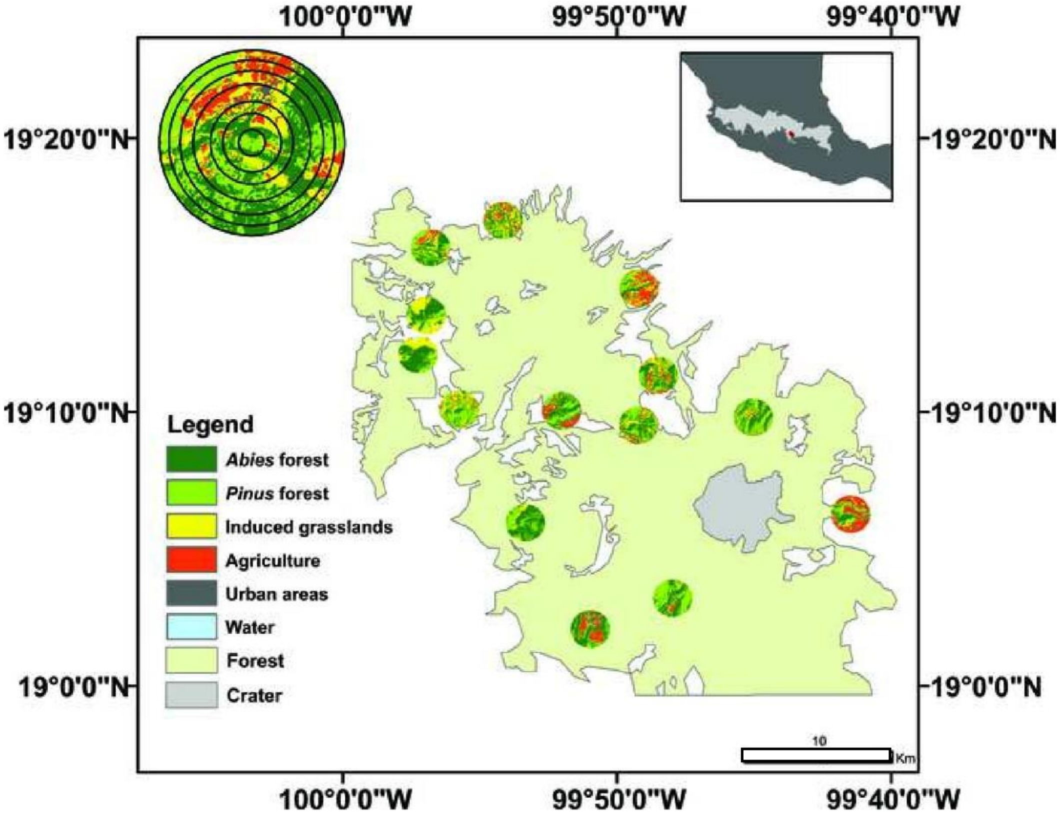
Velo-Antón G, Parra JL, Parra- Olea G, Zamudio KR (2013) Tracking climate change in a dispersal- limited species: reduced spatial and genetic connectivity in a montane salamander. Mol Ecol 22:3261–3278.

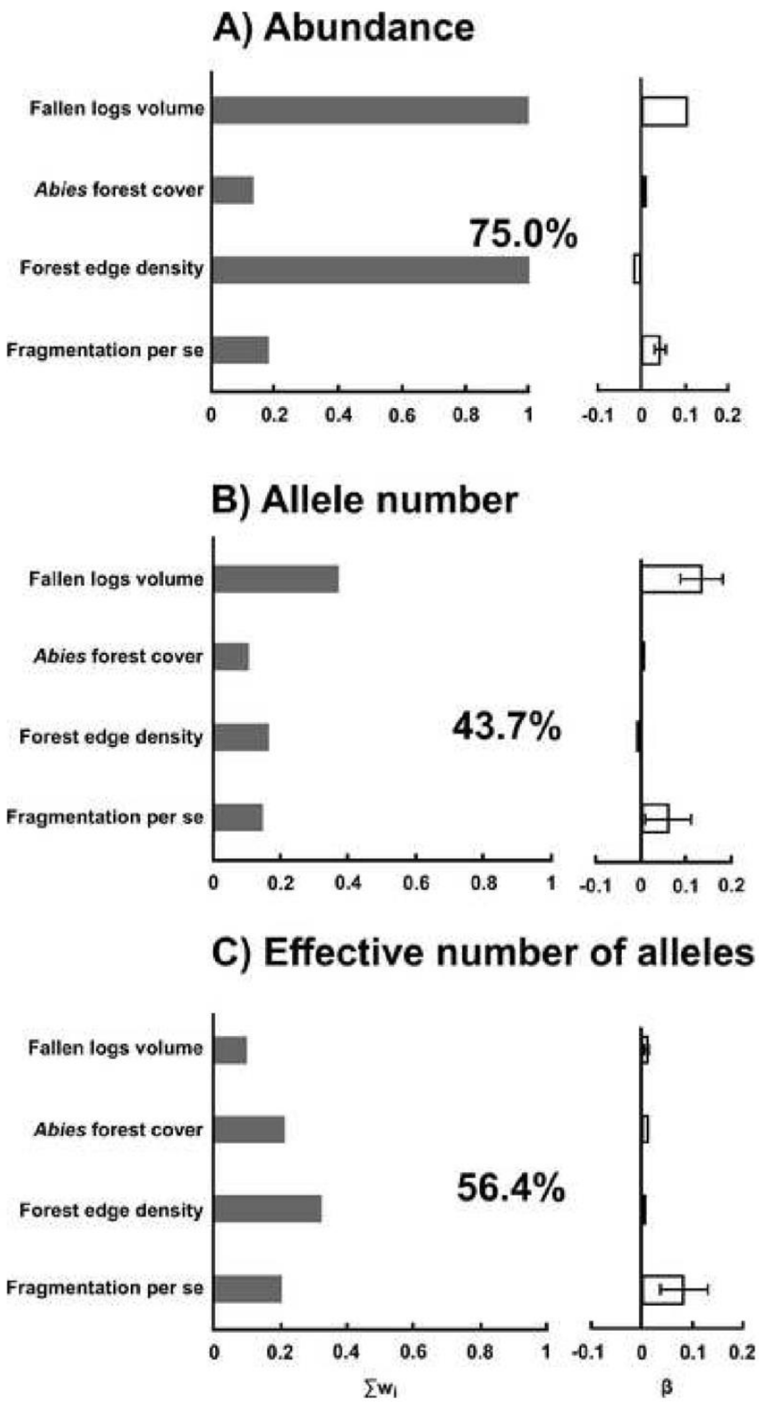
Velo-Antón G, Windfield JC, Zamudio K, Parra-Olea G (2009) Microsatellite markers for *Pseudoeurycea leprosa*, a plethodontid salamander endemic to the Transmexican Neovolcanic Belt. Conserv Genet Resour 1:5–7.

Wang IJ, Savage WK, Bradley Shaffer H (2009) Landscape genetics and least- cost path analysis reveal unexpected dispersal routes in the California tiger salamander (*Ambystoma californiense*). Mol Ecol 18:1365–1374.

Figure 1

Click here to access/download;Figure;Figure 1.tif




A) Abundance
Fallen logs volume
Abies forest cover
Forest edge density
Fragmentation per se
75.0%
0
0.2
0.4
0.6
0.8
1
-0.1
0
0.1
0.2
B) Allele number
Fallen logs volume
Abies forest cover
Forest edge density
Fragmentation per se
43.7%
0
0.2
0.4
0.6
0.8
1
-0.1
0
0.1
0.2
C) Effective number of alleles
Fallen logs volume
Abies forest cover
Forest edge density
Fragmentation per se
56.4%
0
0.2
0.4
0.6
0.8
1
-0.1
0
0.1
0.2
$\sum w_i$
β

Figures

Figure 1. Location of the study forest sites in the Nevado de Toluca volcano, Mexico. Circles within the main panel represent the 14 landscapes selected in each region, considering the largest buffer (500 ha). Yet, landscape metrics were actually measured in 6 different-sized and concentric buffers from the center of each sample site (see example in the top left corner).

Figure 2. Local and landscape predictors of the abundance, allele number and effective number of alleles of *Pseudoeurycea roberts* in the Nevado de Toluca volcano, Mexico *i*. The sum of Akaike weights ($\sum w_i$, panels in the left side) is indicated. Panels in the right side show the values of model-averaged parameter estimates (β) and unconditional variance of information-theory-based model selection and multimodel inference. The sign (+/-) of parameter estimates represents a positive or negative effect of each predictor on the response variable. The percentage of deviance explained by each complete model is also indicated in each panel as a measure of goodness-of-fit.

V. DISCUSIÓN GENERAL

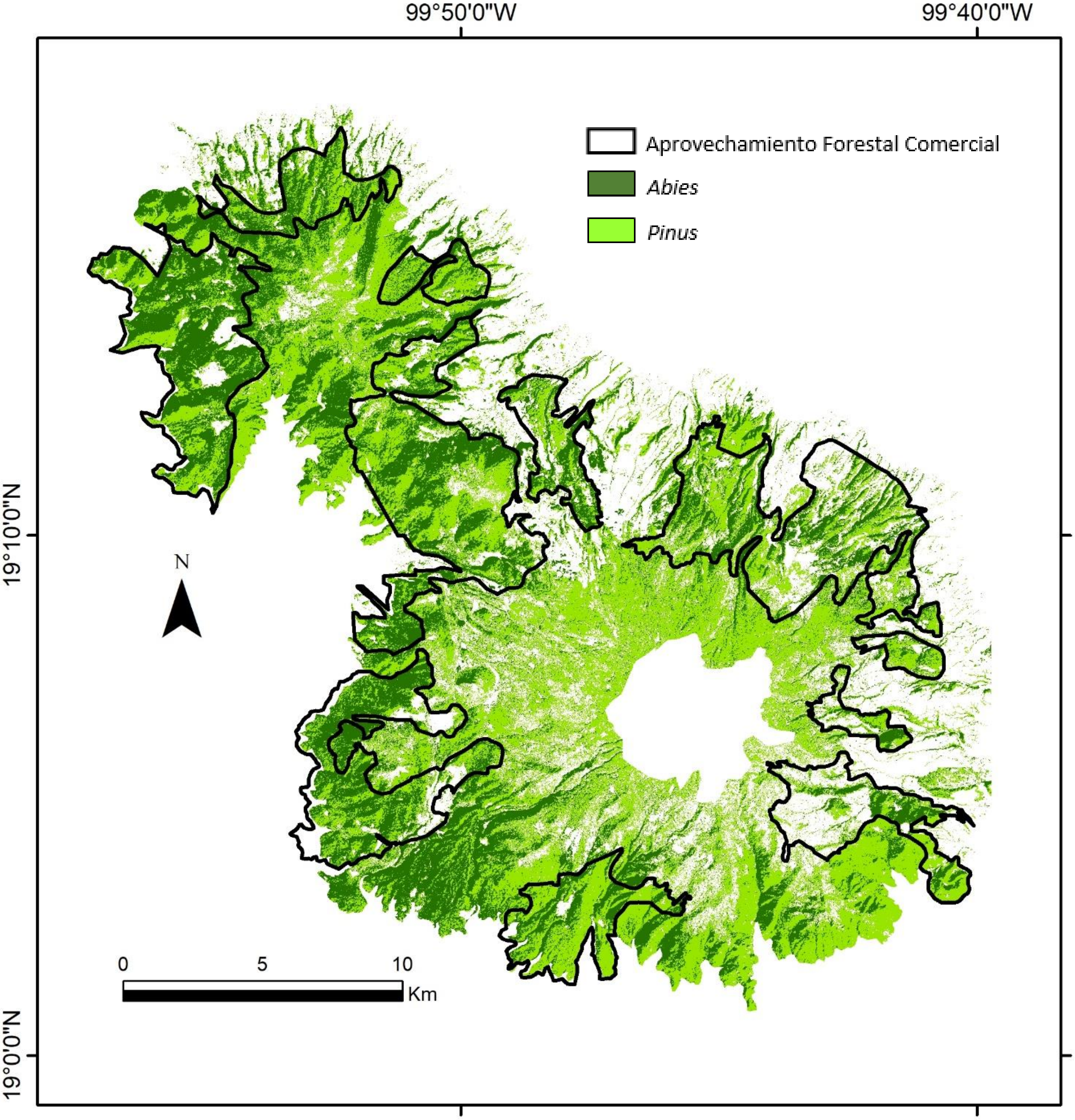
La presente tesis demuestra que la herpetofauna del Eje Neovolcánico Transversal (ENT) está fuertemente influenciada por variables ambientales medidas a diferentes escalas espaciales. En particular, la presencia de las cinco especies de *Thamnophis* en el ENT está negativamente asociada al incremento de la cobertura de vegetación árida. Las demás variables que mejor explican la distribución potencial de las especies varían en cada caso, sin embargo, cabe recalcar que la distancia al bosque de oyamel es la variable más importante para *T. scalaris* y *T. scaliger*; su probabilidad de ocurrencia disminuye en sitios más alejados a estos bosques. Todas las especies tienen una elevada proporción de su distribución potencial en el ENT, sin embargo, de acuerdo a nuestros modelos, todas las especies de *Thamnophis* sufrirán reducciones en sus distribuciones potenciales en esta región. En el segundo estudio, que evalúa la influencia del hábitat en la abundancia y diversidad genética de *P. robertsi*, se encontró que la abundancia de esta salamandra está más estrechamente relacionada con variables locales y del paisaje que la diversidad genética. De acuerdo con lo esperado, la cantidad de hábitat a nivel local (volumen de troncos caídos) tuvo un efecto positivo y fue más importante para explicar la abundancia de la salamandra y el número de alelos que la cantidad de hábitat a nivel de paisaje (porcentaje de bosque de oyamel). Sorprendentemente, la configuración tuvo un mayor efecto que la cantidad de hábitat a nivel de paisaje. En particular, la densidad de borde tuvo un efecto negativo en todas las variables de respuesta. Por lo tanto, retener troncos caídos en los bosques e incrementar las áreas núcleo es crítico para la conservación de esta especie de salamandra.

En ambos estudios se encontró que los bosques de oyamel tienen un papel fundamental; en el primero para la conservación de las culebras *T. scalaris* y *T. scaliger*, en el segundo para la conservación de la salamandra *P. robertsi*. El porcentaje de cobertura de bosque de oyamel fue menos importante para explicar la abundancia y diversidad genética de la especie que otras variables. Sin embargo, todos los individuos fueron encontrados en bosques de oyamel o de oyamel-pino, por ello, podemos afirmar que estos bosques constituyen el macrohábitat de la especie; no puede haber troncos caídos sin bosque y el efecto negativo de la densidad de borde se debe al contacto con la matriz no forestal. Lo que sugieren nuestros resultados es que una extensión mayor de estos bosques pero con poco volumen de troncos caídos y una alta densidad de borde contendrá menor abundancia de salamandras y menor diversidad genética que extensiones que, aunque sean de menor tamaño, tengan un volumen de troncos caídos elevado y baja densidad de borde. Pero si los bosques de oyamel desaparecen o sufren grandes reducciones, *P. robertsi* desaparecerá con ellos. Por lo tanto, es esencial proteger estos bosques en el Nevado de Toluca. Además de *P. robertsi*, en este volcán se encuentran otras salamandras como *P. leprosa*, *Isthmura bellii* y *Aquiloeurycea cefalica* (Bille 2009, observación personal). Estas especies también se refugian debajo de la corteza de los troncos caídos, que son abundantes en los bosques de oyamel de manera natural, ya que estos bosques se encuentran en zonas con elevada pendiente, donde muchos troncos se caen fácilmente debido al viento y las tormentas.

El ENT tiene la mayor extensión de bosques de oyamel del país (91%), sin embargo solamente representan el 1% de la superficie de esta provincia biogeográfica (Sunny et al. 2017). En este estudio se demuestra que la extensión de estos bosques se ha mantenido estable en el país entre 2002 y 2011, mientras que los bosques de pino han sufrido reducciones de alrededor de 3000 km^2 en el mismo período. Así mismo, los bosques densos de pino se redujeron en un 40% en el Nevado de Toluca entre los años 1972 y 2000 mientras que hubo una recuperación de los bosques de oyamel del volcán durante este período (Franco-Maass et al. 2006).

El gobierno mexicano recientemente modificó la categoría de protección del Nevado de Toluca de Parque Nacional a Zona de Protección de Flora y Fauna, una categoría menos restrictiva en cuanto al uso de suelo. De acuerdo al Diario Oficial de la Federación (2013) este cambio contribuirá a preservar los bosques del Nevado de Toluca de la tala ilegal mediante la regulación de las actividades productivas. La justificación para este cambio se basa en la reducción del bosque denso de pino antes mencionada. Sin embargo, esta nueva categoría permite la tala comercial en la mayor parte de la extensión del bosque de oyamel que estuvo bien preservado bajo la categoría del Parque Nacional (Mastretta-Yanes et al. 2014) por lo que creemos que este cambio de categoría está comprometiendo el hábitat de la endémica y críticamente amenazada *P. robertsi* (Figura 1). El tipo de tala que se permite en los polígonos de la Figura 1 es la tala selectiva (práctica de remover uno o dos troncos y dejar intactos los que los rodean), este tipo de tala suele afectar extensiones grandes de bosque causando daños a los árboles circundantes, a la vegetación subarbórea y a los suelos; con impactos en los procesos hidrológicos, erosión, fuego, almacenamiento de carbono y especies de plantas y animales (Asner 2005). Aunque la tala selectiva sea menos invasiva que la tala generalizada, también puede ser muy perjudicial para especies especialistas de bosque, como la salamandra *P. robertis*, especialmente si se afectan las áreas núcleo del bosque. Además de las especies de *Thamnophis* y salamandras mencionadas, los bosques de oyamel también son importantes para la conservación de otras especies de vertebrados, como la lagartija *Barisia imbricata* (Sunny et al. 2017) y de invertebrados, como las colonias migratorias de la emblemática mariposa monarca (Saunders 2018). Por lo tanto, aunque la recuperación de los bosques de pino del volcán es de especial importancia, la preservación de los bosques de oyamel, con áreas núcleo bien conservadas y con elevada disponibilidad de troncos caídos, debe ser una prioridad en el Nevado de Toluca y en todo el Eje Neovolcánico Transversal.

Figura 1: Área del antiguo Parque Nacional con los polígonos que permiten actualmente el aprovechamiento forestal comercial. El bosque de oyamel aparece en verde oscuro y el bosque de pino en verde claro.



VI. REFERENCIAS

Arredondo-León C, Muñoz-Jiménez J, García-Romero A (2008) Recent changes in landscape-dynamics trends in tropical highlands, central Mexico. Interciencia 33:569–577.

Asner GP, Knapp DE, Broadbent EN, Oliveira PJ, Keller M, Silva JN (2005) Selective logging in the Brazilian Amazon. Science 310(5747): 480-482.

Bille T (2009) Field observations on the salamanders (Caudata: Ambystomatidae, Plethodontidae) of Nevado de Toluca, Mexico. Salamandra 45:155–164.

Cardinale BJ, Duffy JE, Gonzalez A, Hooper DU, Perrings C, Venail P, Kinzig AP, Daily GC, Loreau M, Grace JB, Larigauderie A, Srivastava DS, Naeem S (2012) Biodiversity loss and its impact on humanity. Nature 486:59–67.

Catenazzi A (2015) State of the world's amphibians. Annu Rev Environ Resour 40:91–119.

CONAPO (2010) Delimitación de las zonas metropolitanas de México http://www.conapo.gob.mx/en/CONAPO/ Zonas_metropolitanas_2010. Accessed 5 November 2018.

DOF, Diario Oficial de la Federación Mexicana (2013): Decreto que reforma, deroga y adiciona diversas disposiciones del diverso publicado el 25 de enero de 1936, por el que se declaró Parque Nacional la montaña denominada "Nevado de Toluca" que fue modificado por el diverso publicado el 19 de febrero de 1937. http://dof.gob.mx/nota_detalle_popup.php?codigo=5315889. Accessed 5 November 2018.

Ehrlén J, Morris W F (2015) Predicting changes in the distribution and abundance of species under environmental change. Ecol Lett 18:303–314.

Eigenbrod F, Hecnar SJ, Fahrig L (2008) The relative effect of road traffic and forest cover on anuran populations. Biol Conserv 141:35–46.

Espinoza D, Ocegueda S. 2007. Introduccion. In: Luna-Vega I, Morrone JJ, Espinosa D, eds. Biodiversidad de la Faja Volcánica Transmexicana. México: Comisión Nacional para el Conocimiento y Uso de la Biodiversidad. Universidad Nacional Autónoma de México, 5–6.

Flores-Villela O, Canseco-Márquez L, Ochoa-Ochoa L (2010) Geographic distribution and conservation of the herpetofauna of the highlands of Central Mexico. In: Conservation of Mesoamerican Amphibians and Reptiles. Wilson LD, Towsend JH, Johnson JD. (eds.) Eagle Mountain Publishing Co. Utah, USA, pp 303–321.

Foley JA, DeFries R, Asner GP, Barford C, Bonan G, Carpenter SR, Chapin FS, Coe MT, Daily GC, Gibbs HK, Helkowski JH, Holloway T, Howard EA, Kucharik CJ, Monfreda C, Patz JA, Prentice IC, Ramankutty N, Snyder PK (2005) Global consequences of land use. Science 309:570–574.

Franco-Maass S, Regil-García HH, González-Esquivel C, Nava-Bernal G (2006) Cambio de uso del suelo y vegetación en el Parque Nacional Nevado de Toluca, México, en el periodo 1972-2000. Invest Geog 61:38–57.

Gibbons JW, Scott DE, Ryan TJ, Buhlmann KA, Tuberville TD, Metts BS, Greene JL, Mills T, Leiden Y, Poppy S, Winner CT (2000) The global decline of reptiles, déjà vu amphibians. BioScience 50:653–661.

Groombridge B, Jenkins M (2000) Global Biodiversity. Earth's Living Resources in the 21st Century. World Conservation Monitoring Centre, Cambridge, U.K.

Hoffmann AA, Willi Y (2008) Detecting genetic responses to environmental change. Nat Rev Genet 9:421–432.

Huey RB (1982) Temperature, physiology, and the ecology of reptiles. En: Gans C, Pough FH (Eds.) Biology of Reptilia. Academic Press, New York, pp. 25–74.

Mas J, Velázquez A, Díaz-Gallegos J, Mayorga-Saucedo R, Alcántara C, Bocco G, Castro R, Fernández T, Pérez-Vega A (2004) Assessing land use/cover changes: a nationwide multidate spatial database for Mexico. Int J Appl Earth Obs Geoinf 5:249–261.

Mastretta-Yanes A, Cao R, Nicasio-Arzeta S, Quadri P, Escalante-Espinosa T, Arredondo L, Piñero D (2014) ¿Será exitosa la estrategia del cambio de categoría para mantener la biodiversidad del Nevado de Toluca? Oikos 12: 7–17.

Mendelson JR III, Lips KR, Gagliardo RW, Rabb GB, Collins JP, et al. (2006) Confronting amphibian declines and extinctions. Science 313: 48.

Newbold T, Hudson LN, Arnell AP, Contu S, De Palma A, Ferrier S (2016) Has land use pushed terrestrial biodiversity beyond the planetary boundary? A global assessment. Science 353:288–291.

Nowakowski AJ, Watling JI, Thompson ME, Brusch GA, Catenazzi A, Whitfield SM, Kurz DJ, Suárez-Mayorga A, Aponte-Gutiérrez A, Donnelly MA, Todd BD (2018) Thermal biology mediates responses of amphibians and reptiles to habitat modification. Ecol Lett 21:345–355.

Parmesan C, Yohe G (2003) A globally coherent fingerprint of climate change impacts across natural systems. Nature 421:37–42.

Saunders SP, Ries L, Oberhauser KS, Thogmartin WE, Zipkin EF (2018) Local and cross-seasonal associations of climate and land use with abundance of monarch butterflies Danaus plexippus. Ecography 41(2): 278–290.

Stuart SN, Chanson JS, Cox NA, Young BE, Rodrigues ASL, Fischman DL, Waller RW (2004) Status and trends of amphibian declines and extinctions worldwide. Science 306:1783–1786.

Suárez-Atilano M. 2015. Filogeografía de Boa Constrictor (Serpentes: Boidae) en México y Centro América. PhD Thesis, Universidad Nacional Autónoma de México. Posgrado en Ciencias Biológicas. Instituto de Ecología.

Sunny A, González-Fernández A, D'Addario M (2017) Potential distribution of the endemic imbricate alligator lizard (*Barisia imbricata imbricata*) in highlands of central Mexico. Amphib Reptil 38:225–231.