Therya

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AMMAC

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La Portada

El tlacuache norteño o zarigüeya norteña (*Didelphis virginiana*) es una especie muy común en todas las regiones tropicales y en fechas recientes se ha expandido ampliamente en todo Norte América, llegan incluso a Canadá. *Didelphis virginiana* es una especie relativamente abundante y muy conspicua donde se encuentra. En los sitios en los que se distribuye es común encontrar a los ejemplares atropellados en las carreteras. La foto fue tomada en el límite entre Oaxaca y Chiapas (Fotografía de Sergio Ticul Álvarez Castañeda)

Nuestro logo "Ozomatli"

El nombre de "Ozomatli" proviene del náhuatl se refiere al símbolo astrológico del mono en el calendario azteca, así como al dios de la danza y del fuego. Se relaciona con la alegría, la danza, el canto, las habilidades. Al signo decimoprimero en la cosmogonía mexica. "Ozomatli" es una representación pictórica de los mono arañas (*Ateles geoffroyi*). La especie de primate de más amplia distribución en México. " Es habitante de los bosques, sobre todo de los que están por donde sale el sol en Anáhuac. Tiene el dorso pequeño, es barrigudo y su cola, que a veces se enrosca, es larga. Sus manos y sus pies parecen de hombre; también sus uñas. Los Ozomatin gritan y silban y hacen visajes a la gente. Arrojan piedras y palos. Su cara es casi como la de una persona, pero tienen mucho pelo."

Contenido

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DERECHOS DE AUTOR Y DERECHOS CONEXOS, año 9, No. 3, Septiembre-diciembre del 2018, es una publicación cuatrimestral editada por la Asociación Mexicana de Mastozoología A. C., Hacienda Vista Hermosa 107, Colonia Villa Quietud, Coyoacan 04960. Distrito Federal, México. Telefono (612) 123-8486, www.mastozoologiamexicana.org, therya@cibnor.mx. Editor responsable: Dr. Sergio Ticul Álvarez Castañeda. Reservas de Derechos al Uso Exclusivo No. 04-2009-112812171700-102, ISSN: 2007-3364 ambos otorgados por el Instituto Nacional de Derechos de Autor. Responsable de la última actualización de este número, Unidad de informática de la Asociación Mexicana de Mastozoología A. C. Dr. Sergio Ticul Álvarez Castañeda. Instituto Politécnico Nacional 195. La Paz, Baja California Sur, C. P. 23096. Tel, (612) 123-8486, fecha de la última modificación 30 septiembre 2018.

Therya

El objetivo y la intención de *THERYA* es ser una revista científica para la publicación de artículos sobre los mamíferos. Estudios de investigación original, editoriales, artículos de revisión y notas científicas son bienvenidas.

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Letter to the editor

Acoustic reference library of mexican insectivorous bats: Phase I.

El 15 de marzo de 2016 se firmó el Convenio Número FB1796/ME004/16 entre la Comisión Nacional para el Conocimiento y Uso de la Biodiversidad (CONABIO) y la Asociación Mexicana de Mastozoología A. C. (AMMAC), para ejecutar el proyecto "Compilación de fonoteca de referencia de los murciélagos insectívoros de México: Fase I". El objetivo principal del proyecto fue iniciar la conformación de una fonoteca de referencia de sonidos de ecolocalización de las especies de murciélagos con sonidos de alta intensidad presentes en el territorio mexicano (Sonozotz-AMMAC-CONABIO 2018). A dos años de haberse iniciado, el 28 de febrero de 2018, el proyecto técnicamente ha finalizado con resultados exitosos y la obtención de los productos comprometidos.

Desde la reunión inicial en 2014 que se efectuó en el marco del XI Congreso Nacional de Mastozoología en la Benemérita Universidad Autónoma de Puebla, la organización del proyecto supuso un gran reto para conjuntar los esfuerzos que diversos especialistas y miembros de la AMMAC venían desarrollando en la detección acústica de murciélagos. El esfuerzo tenía que ser multiinstitucional, abarcar todo México, incluir innumerables tipos de ecosistemas, considerar más de 80 especies de murciélagos que emiten pulsos de ecolocalización intensos, conformar grupos de trabajo con investigadores con diferentes formas pensar, incluir dependencias con equipo y capacidades técnicas disímiles, así como establecer procedimientos de gestión estandarizados entre instancias con manuales de administración distintos. Todo tendría que ser planeado y ejecutado con muy elevados estándares de calidad técnica y científica.

Para la AMMAC, el proyecto representó también el primero en donde la Asociación fungió como institución receptora de los recursos financieros y como responsable de la ejecución de los mismos. El aprendizaje en la gestión y administración de los fondos, marcan un precedente positivo y abren oportunidades para poder someter futuros proyectos con cobertura nacional e impacto científico internacional. De esta forma, los resultados no sólo radican en productos académicos (informes, tesis, publicaciones, formación de recursos humanos, congresos, bases de datos, metadatos, fotografías, etc.), sino también en buenas prácticas estandarizadas para la gestión y la aplicación de los resultados (procedimientos, protocolos, etc.). Por ejemplo, se desarrollaron procedimientos por parte de la AMMAC para administrar recursos financieros provenientes de Proyectos de Investigación Científica (PIC), se actualizaron los estatutos de la Asociación, se mejoró el manejo contable y se generaron convenios de colaboración con algunas de las instituciones a las cuales están adscritos los miembros participantes.

Desde un inicio, el proyecto contó con la participación de por lo menos 18 miembros de la AMMAC pertenecientes a igual número de instituciones distribuidas en todo el territorio mexicano. Se abarcó ocho regiones del país: Californiana (Baja California, Baja California Sur y Sonora); Noroeste (Durango, Sinaloa y Chihuahua); Occidente (Colima, Nayarit y Jalisco); Oriente (Puebla, Tlaxcala y Veracruz): Centro Norte (Aguascalientes, Guanajuato, San Luis Potosí, Nuevo León y Zacatecas); Centro Sur (Estado de México, Morelos, Hidalgo y Querétaro); Sureste (Campeche, Quintana Roo y Yucatán) y Suroeste (Chiapas, Oaxaca y Tabasco). Fue necesario generar acuerdos para nombrar al responsable del proyecto, a los coordinadores generales técnicos y a los coordinadores para cada región. El proceso exigió la voluntad de todos los participantes para maximizar consensos y minimizar conflictos.

El proyecto planteo siete objetivos particulares (Sonozotz-AMMAC-CONABIO 2018), que de forma sintética incluían la conformación de una red de grupos de trabajo con personal capacitado; organizar talleres de capacitación y entrenamiento; integrar grabaciones acústicas de murciélagos ya existentes; definir un diseño de muestreo adecuado; diseñar un procedimiento de validación e ingreso de datos a la fonoteca; incorporar a la fonoteca las primeras grabaciones; y determinar los procedimientos de comparación estadística para la discriminación entre especies.

Los participantes en el proyecto denominaron a esta iniciativa "Sonozotz". "Sono" hace alusión a los sonidos de ecolocalización emitidos por los murciélagos; "Zotz", significa murciélago en lengua maya y hace referencia a los importantes valores iconográficos, religiosos, simbólicos y míticos de los quirópteros vinculados con la gran diversidad cultural de México; AMMAC, además del significado de las siglas, representa los esfuerzos colegiados e institucionales de los mastozoólogos del país. En el 2016, a través de un concurso abierto, durante el XIII Congreso Nacional de Mastozoología en la Universidad de Ciencias y Artes de Chiapas, "Sonozotz" adquirió logotipo.

Como parte del proyecto, se generó un protocolo general de trabajo de campo que permitió la homogenización de la colecta de información, el procesamiento y la organización de los datos de campo, así como formatos e instructivos para la descripción de las localidades, los datos de ejemplares, los datos de grabación y el catálogo de preparación. El protocolo incluyó una guía sobre el proceso de grabación de los sonidos, la utilización de los equipos, la instalación de los programas especializados, el ensamblado y activación de los detectores, los métodos de grabación y liberación de ejemplares, la colecta de tejidos y de ejemplares para su posterior resguardo en las colecciones científicas de mamíferos autorizadas. Este protocolo representa por sí mismo un logro del proyecto, que permitirá a otros grupos interesados en acústica aplicarlo en diversos estudios.

Se realizaron cuatro talleres en diferentes sedes: 1) La estación Biológica La Mancha del Instituto de Ecología, en La Mancha, Veracruz; 2) Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional (CIIDIR) Unidad Durango del Instituto Politécnico Nacional (IPN), en Durango, Durango; CIIDIR-IPN Unidad Oaxaca, Oaxaca; Centro de Investigación en Biodiversidad y Conservación (CIByC) de la Universidad Autónoma del Estado de Morelos (UAEM), Cuernavaca, Morelos. En todos los talleres asistieron aproximadamente 80 personas que desarrollan trabajos de investigación con murciélagos en México y que participaron en sesiones prácticas para la captura, muestreo y grabación de ejemplares capturados *in situ*.

Durante las salidas de campo que se realizaron de junio de 2016 a diciembre de 2017, se aplicó el protocolo de muestreo específicamente elaborado para el proyecto. Los grupos participantes por región, tuvieron la oportunidad de contar con el apoyo de estudiantes y voluntarios. En ese sentido, tanto los talleres como el trabajo de campo y gabinete, tuvieron un importante efecto en la formación de recursos humanos de licenciatura y posgrado de diversas instituciones del país.

El proyecto logró reunir 173 colectores que participaron al menos en una salida de campo, se cubrió el 100 % de las ocho regiones propuestas con 183 localidades que cubrieron los principales ecosistemas de México. Se colectaron 1,640 individuos pertenecientes a 67 especies y se cubrió el registro de llamados de ecolocalización del 63% de las especies de murciélagos insectívoros de México con un total de 2,302 grabaciones (Sonozotz-AMMAC-CONABIO 2018).

La CONABIO desarrolló una plataforma especialmente diseñada para este proyecto en donde estarán albergadas bases de datos, mapas, metadatos, llamados y fotografías, que en el corto plazo constituirá la primera fonoteca de referencia de los murciélagos insectívoros de México y la primera a nivel mundial de un país megadiverso. Se espera que este proyecto permita plantear un programa de monitoreo a nivel nacional y consolidar la formación de la "Red Mexicana de Monitoreo Acústico de Murciélagos".

La aplicación de las metodologías de detección acústica en países como México todavía está limitada por la poca disponibilidad de equipo y, sobre todo, porque no se contaba con fonotecas de referencia adecuadas para la elaboración de criterios de identificación de las especies (Walters *et al.* 2013). En ese sentido, la compilación de una fonoteca de sonidos de ecolocalización representativa de las especies presentes en el país, así como el desarrollo de criterios de identificación acústica que pudieran estar disponibles como herramientas electrónicas de uso libre, potenciará el uso de los métodos de detección acústica.

Los murciélagos insectívoros son susceptibles a amenazas generadas por la actividad humana. Es evidente que estas especies son sensibles a la pérdida y degradación de los hábitats naturales, a los pesticidas y contaminantes (Racey y Entwistle 2003). Muchas especies son excepcionalmente sensibles a impactos con los generadores de energía eólica (Kunz *et al.* 2007) y otras son especialmente vulnerables a las perturbaciones de sus refugios (Racey y Entwistle 2003).

La disponibilidad de una herramienta para la identificación acústica y de metodologías de monitoreo estandarizadas (Zamora-Gutiérrez *et al.* 2016), facilitará la operación y la obtención de datos confiables a las empresas, agencias gubernamentales, academia y otros sectores interesados en la evaluación de riesgos e impactos a través del estudio de los murciélagos. Esto permitirá la generación de información sobre la distribución de las especies y posibilitará la aplicación de programas de monitoreo y estudios técnicos requeridos para la implementación de proyectos de desarrollo, como Manifiestos de Impacto Ambiental (MIA), Documentos Técnicos Unificados (DTU), Estudios Técnicos Justificativos (ETJ), Auditorías Ambientales (AA), Programas de Desarrollo Urbano (PDU), Programas de Manejo de Áreas Naturales Protegidas y de Unidades de Manejo de Vida Silvestre, entre otros.

La AMMAC agradece el apoyo recibido por la CONABIO, así como el trabajo realizado por todos los participantes en el proyecto y a todas las instituciones a los que están adscritos los colaboradores. Además de los productos académicos, se espera que en el corto plazo, se planteen otros proyectos relativos a la conservación y manejo de los mamíferos de México, con la participación activa de los miembros de la AMMAC.

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Food habits of puma (*Puma concolor*) in the Andean areas of Tamá National Natural Park and its buffer zone, Colombia

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Even though the puma (*Puma concolor*) is the second largest felid in America and is widely distributed in both the continent and in Colombia. Knowledge about its food habits in many areas of the Neotropics, especially in the highlands, is still limited. We surveyed nine localities in Tamá NNP (National Natural Park) and its buffer zone monthly from June 2012 to May 2015, between 2,067 and 3,500 masl. We surveyed three linear transects of 2 to 3 km in each locality searching for scats, which were located mainly on mountain ridges. We analyzed fecal contents and estimated the minimum number of samples needed to describe puma diet, as well as the absolute frequency of records for each prey species, their relative frequency of occurrence, and biomass consumed. Potential prey species were identified using photos from camera traps at the same localities. Fecal analysis ($n = 45$) suggests at least eight species of medium-sized mammals as prey. Of these, three species were the most frequent, contributing 67.87 % of the biomass consumed: *Nasua nasua*, *Mazama rufina* and *Cuniculus taczanowskii*. The latter is a newly reported food item for the puma. No large or domestic species were recorded as part of the species' diet for the study area, in spite of its availability. Considering the potential conflict with communities surrounding the protected area, by the alleged consumption of farm animals by this and other felids, it appears that the availability of wild prey in Tamá NNP is suitable for puma and apparently the species does not need to use alternative prey as livestock. Our data suggest that the minimum number of samples required to estimate the diet in our study area varies between 31 and 41, suggesting that our results ($n = 45$) are a good estimate of the species' diet. This work highlights the importance of expanding the knowledge about puma foraging ecology in order to generate better conservation strategies for a species of great importance for the dynamics and stability of the Colombian Andean ecosystems.

A pesar de que el puma (*Puma concolor*) es el segundo felino más grande en América y presenta una amplia distribución tanto en el continente como en Colombia. La información sobre los hábitos alimenticios en muchas de las áreas a nivel del Neotrópico, especialmente en zonas altas, aún es limitada. Se realizaron muestreos en nueve localidades del PNN (Parque Nacional Natural) Tamá y su zona de amortiguación mensualmente entre junio del 2012 y mayo del 2015, entre los 2,067 y 3,500 msnm. Se realizaron tres transectos lineales de entre 2 y 3 km (por cada localidad) para búsqueda de heces, las que se ubicaron principalmente sobre las crestas de las montañas. Se analizó el contenido y se estimó el número mínimo de muestras necesario para describir la dieta del Puma, la frecuencia absoluta de las presas, de ocurrencia y biomasa consumida. Las especies potenciales también se obtuvieron a través de cámaras trampa de las mismas localidades. El análisis de excretas ($n = 45$) indicó al menos ocho especies de mamíferos medianos como presas. Las tres más frecuentes y las que aportan en conjunto el 67.87 % de la biomasa consumida son: *Nasua nasua*, *Mazama rufina* y *Cuniculus taczanowskii*. Esta última especie es un nuevo registro en la dieta para el puma. Ninguna especie grande o doméstica fue registrada como parte de la dieta para el área de estudio, a pesar de su disponibilidad. Aunque el conflicto potencial con comunidades aledañas al área protegida es el supuesto consumo de animales domésticos, parece que la disponibilidad de presas silvestres en el PNN Tamá es la adecuada para los pumas y no tienen necesidad de utilizar presas alternativas como el ganado. Nuestros datos muestran que el número mínimo de muestras necesarias para estimar la dieta del puma es de 31 a 41, lo que sugiere que este trabajo ($n = 45$) es una buena aproximación. Este trabajo resalta la importancia de conocer aún más sobre estos aspectos ecológicos para así poder generar mejores estrategias de conservación para la especie, la cual es de gran importancia para la dinámica y estabilidad de los ecosistemas Andinos del país.

Keywords: Andes; biomass; dietary habits; puma.

Introduction

The puma (*Puma concolor*) is the felid with the largest distribution in America, occupying areas from southern Canada to southern Chile and Argentina (Currier 1983). In this large range, the species inhabits a wide variety of habitats (*i. e.,* savannas, humid forests, deserts) including some peri-urban landscapes surrounding large cities (Mazzolli 2012; Arias-Alzate *et al.* 2015). In the Neotropics the species is considered the main predator in mountain ecosystems, especially in areas above 2,000 masl (De Angelo *et al.* 2009, Arias-Alzate *et al*. 2015).

Despite its importance as a top predator in most ecosystems that it inhabits (*e. g.,* regulates and maintain prey populations; Hoogesteijn and Hoogesteijn 2005, Hunter and Barrett 2011), most studies about puma diet come from high latitudes or Neotropical lowlands where diet has been reported as highly diverse and strongly dependent on mammals (Chinchilla 1997; Pessino *et al.* 2001; Moreno *et al.* 2006; Hernández 2008, Skewes *et al.* 2012). For instance, diet in North American populations is mostly composed of large ungulates (*e. g., Odocoileus hemionus*; Iriarte *et al.* 1990), while in Central America and southern South America the puma shows a tendency towards feeding on medium and small species (*e. g., Dasyprocta* spp., *Cuniculus* spp., *Pecari tajacu* and *Mazama* spp.; Currier 1983; Emmons 1987; Nuñez *et al.* 2000; Pessino *et al.* 2001; Polisar *et al.* 2003; Novack *et al.* 2005; Hernández 2008; Pontes and Chivers 2007). In contrast to the work done elsewhere, studies on puma diet in other regions of South America (*e. g.,* northwest) are still scarce, especially in highlands, which have only recently received a little attention (Hernández-Guzmán *et al.* 2011).

In Colombia, little is known about the foraging ecology of this species and most information comes from opportunistic unpublished records and personal communications with only one systematic study, from Puracé National Natural Park in the central range of the Colombian Andes (departments of Cauca and Huila; Hernández-Guzmán *et al.* 2011). Here we assess puma diet in the northern Andes, in the highlands of Tamá NNP (National Natural Park; department of Norte de Santander). We emphasize that this knowledge is of great significance, since it provides greater understanding of the species' ecology and its interactions with other species, information needed for the definition of better strategies for puma management and conservation, especially in a protected area surrounded by human activities.

Materials and methods

Study area. The study was conducted in Tamá NNP, in the southeastern portion of the department Norte de Santander, in the eastern Andean range of Colombia. The park is under jurisdiction of the Toledo and Herrán municipalities between 2,067 and 3,356 masl (7° 02' and 7° 27' N; -72° 02' and -72° 28' W; Figure 1; Cáceres-Martínez *et al.* 2016). The park covers approximately 480 km^2 of the Tamá Massif on the limits with Venezuela; Tamá NNP is part of a bi-national border protected area with El Tamá National Park in Venezuela.

Selection of sampling sites. The puma is the second largest cat present in Tamá NNP with a strict carnivorous diet, after the jaguar (*Panthera onca*; Cáceres-Martínez *et al.* 2016), so there is a potential for confusing scats from these two species, mainly when there are not associated tracks or other signs that allow certain species identification. Therefore, to avoid scat misidentification, we conducted the study only at high elevations (2,067 to 3,500 masl) where the jaguar is usually absent (De la Torre *et al.* 2018); jaguar have been recorded in the park only in lowlands (between 350 and 800 masl; Cáceres-Martínez *et al.* 2016). In this altitudinal range we selected nine localities for field sampling: La Carpa, La Rochela (buffer zone), Orocué, Páramo of

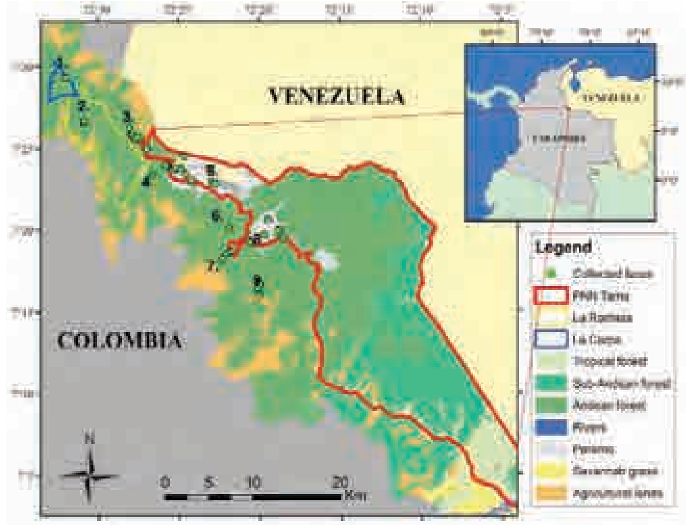

Figure 1. Cover types and locations of collected puma scats within Tamá National Natural Park and its buffer area in the high Andean region. 1. La Carpa. 2. La Rochela. 3. Orocué. 4. Páramo de la Cabrera. 5. Paramo of Tamá. 6. Belchite. 7. Asiria de Belén. 8. Páramo of Santa Isabel. 9. Santa Isabel.

Cabrera, Páramo of Tamá, Belchite, Asiria de Belén, Páramo of Santa Isabel, and Santa Isabel, covering a significant portion of the highlands of the park and buffer zone (Figure 1). Average temperature in these areas ranges from 6 to 16 °C with precipitation variating between 800 and 2,000 mm/year with a frequent mist contributing to the constant humidity of the study area (Meneses *et al.* 2004).

Collection of scat samples. Scat collection was carried out from line-transects along trails, across natural covers following mountain ridges and on streambanks. At least three transects at each location, from 2 to 3 km in length, were sampled monthly between June 2012 and May 2015. Scats were identified as from puma by the presence of tracks and specific scat characteristics (*i. e.,* diameter, length, presence of hairs), following the morphological description and biometrics of Aranda (2012). Scats were collected and stored in plastic bags for later analysis. For each sample we recorded geographical position, date and elevation.

Food habits and prey biomass analysis. To determine the puma's diet each scat was measured, weighed, and washed over wire mesh with tap water to separate the components (*i. e.,* hair, bones, claws, and teeth). Afterwards, all samples were dried at room temperature for 48 hours (Ackerman *et al.* 1984). Each item was identified to species level using a reference collection of mammal specimens from the Instituto de Ciencias Naturales (ICN) collection at the National University of Colombia (Bogotá) and through literature review. All samples were deposited at the mammal collection of the Jose Celestino Mutis museum, Universidad of Pamplona. We also estimated the minimum number of scats required for an adequate diet description through a prey-species accumulation curve (Foster *et al.* 2010; Hernández-Guzmán *et al.* 2011). We performed this analysis by randomly adding all samples and estimating average and standard deviations from 1,000 permutations (Gotelli and Colwell 2001), thus eliminating the influence of the order in which each scat was added. The analysis was performed using Vegan Package v 2. 4 - 2 with the function *specaccum* (Oksanen *et al.* 2007) implemented in R. We also estimated prey absolute frequency (AF) as the number of times a prey type was present in all scats collected (Rueda-Zozaya 2010) and then estimated the frequency of occurrence (FO) using the ratio of the absolute frequency (AF) to the total number of scats (Rueda-Zozaya 2010). We then calculated the percentage of occurrence (PO) as (PO) = (AF) / the total number of scats x 100.

Biomass consumed (*i. e.,* percentage of biomass) was calculated as the mean weight of each prey species multiplied by the frequency of each species, divided by the total weight of all species in the total scat samples, then multiplied by 100 (Medina *et al.* 2009). Body weights were based on literature reports (*e. g.,* Ackerman *et al*. 1984; Eisenberg 1989; Ramírez 2011). As large-sized prey species are more likely to occur in smaller proportion in the scats than small species, due to differential ingestion and digestion, potentially leading to overestimation of some species (Rueda-Zozaya 2010), we applied a biomass correction factor using Ackerman's (1984) equation (Y = 1.98 + 0.035X), where Y is the weight of prey consumed per scat and X is the mean weight of the prey species. Prey species identified in scats were corroborated with camera-traps records obtained in the same localities (Cáceres-Martínez *et al*. 2016).

Results

We collected a total of 45 scat samples (Appendix 1). The accumulation curve approached the asymptote, indicating that most potential prey species were detected within approximately 40 scat samples (Figure 2).

We identified eight prey species from the scats; other vertebrate items were found but their species identification was uncertain (Appendix 1) so they were excluded from analysis. The most commonly represented species were the South American coati (*Nasua nasua*), followed by the dwarf red brocket deer (*Mazama rufina*) and mountain paca (*Cuniculus taczanowskii*) with AF values of 9, 6 and 5 respectively (Table 1). Meanwhile, the western mountain coati (*Nasuella olivacea*), the Central American agouti (*Dasyprocta punctata*), the nine-banded armadillo (*Dasypus novemcinctus*) and the two-toed sloth (*Choloepus hoffmanni*) were less frequently detected (Table 1). The species with the largest FO and PO was *N. nasua* followed by *M. rufina* (Table 1).

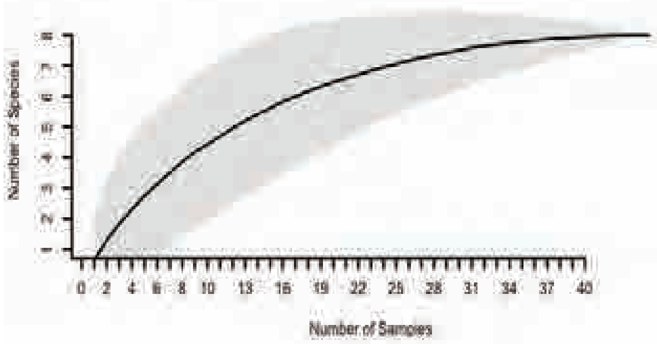

Figure 2. Prey species accumulation curves indicating the number of scats samples required to describe the food habits of puma in the study area; gray shading indicates 95% confidence interval.

In terms of biomass consumed, *C. taczanowskii* presented the greatest contribution per individual (2.435 kg; Table 1); however, *N. nasua* contributed more to the total biomass consumed (28.6 % of biomass consumed), followed by *M. rufina* (20.71 %) and *C. taczanowskii* (18.54 %), together accounting for 67.87 % of the total biomass. The other five species contributed together the remaining 32.13 % of the total biomass consumed by puma (Table 1). No large or domestic species were recorded as consumed by puma in the study area.

Table 1. Composition of puma's diet and relative biomass consumed based on 45 scats collected in Tamá NNP: Absolute frequency (AF), frequency of occurrence (FO), percentage of occurrence (PO), mean prey weight **(**MPW), biomass correction factor (BCF) and relative biomass consumed (RBC).

Species prey	FA	FO	PO (%)	MPW (kg)	BCF (kg)	RBC (kg)	Biomass (%)
Nasua nasua	9	0.20	20.00	3.07	2.08	18.79	28.61
Mazama rufina	6	0.13	13.33	8.20	2.26	13.60	20.72
Cuniculus taczanowskii	5	0.11	11.00	13.00	2.43	12.18	18.54
Nasuella olivacea	3	0.06	6.00	1.50	2.03	6.10	9.29
Choloepus hoffmanni	2	0.04	4.40	5.70	2.18	4.36	6.64
Didelphis marsupialis	2	0.04	4.40	5.50	2.17	4.35	6.62
Dasypus novemcinctus	2	0.04	4.40	3.10	2.08	4.18	6.36
Dasyprocta punctata	1	0.02	2.20	4.00	2.12	2.12	3.23

Discussion

Our work represents the first study on *P. concolor* conducted in northeastern Colombia and the second study in a protected area of the Colombian Andes. The minimum number of scats necessary to assess puma diet could vary depending on the conditions of each location, which would require different sampling efforts according to particular temporal and spatial contexts. Our data showed that the minimum number of scats necessary for estimating the species' diet in our study area is between 31 and 41 scats, suggesting that our sampling ($n = 45$) was adequate to give an accurate estimate of the species' diet. This result corresponds with previous studies in Mexico by Monroy-Vilchis *et al.* (2009) and Núñez *et al.* (2000) which suggested a minimum of 15 and 40 scats, respectively; in Colombia, a study in Puracé NNP in the Central Andes of Colombia estimated a minimum requisite of 20 scats (Hernández-Guzmán *et al.* 2011), which overlaps our estimation.

Even though it appears we had a sufficient sampling effort, it is likely some prey species were not detected given our method based on comparisons with reference material (Klare *et al.* 2011). For instance, 50 % of the samples had unidentified items due to the advanced state of decomposition. To avoid this loss of information, Foster *et al.* (2010) suggested from species accumulation curves that the minimum sample size required to fully characterize the species' diet should include at least 100 scats; however, high decomposition rates associated with warm and/or humid

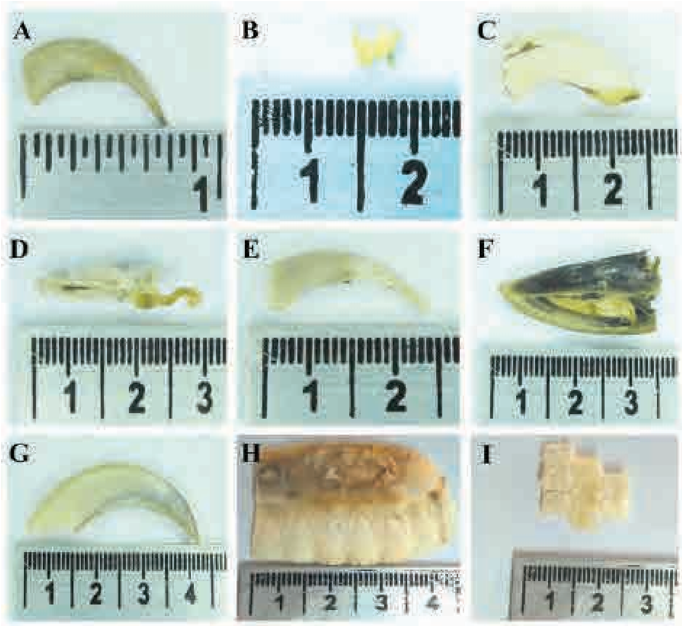

Figure 3. Prey species recorded in puma scat in Tamá NNP a) *Nasua nasua*, b) *Didelphis marsupialis*, c) *Cuniculus taczanowskii*, d) *Dasyprocta punctata*, e) *Nasuella olivacea*, f) *Mazama rufina*, g) *Choloepus hoffmanni*, h) and i). *Dasypus novemcinctus*.

conditions, such as those in tropical and cloud forests, may limit the availability and processing of scats.

Our results showed that the most frequently consumed prey species in the study area are small- to medium-sized mammals. Prey species composition in our results were very similar to those reported in other studies along different areas of the Neotropics (Nuñez *et al.* 2000, Crawshaw and Quigley 2002, Novack *et al*. 2005, Moreno *et al*. 2006, De Azevedo 2008, Foster *et al.* 2010). However, frequency of species occurrence varies considerably across studies. We found that for Tamá NNP, *N. nasua*, *M. rufina*, and *C. taczanowskii* (followed by *N. olivacea*) were the most frequently consumed species by puma, which is very similar to previous studies in Central and South America. For instance, numerous studies have found *Nasua* spp. among the most frequent prey in puma's diet in México (Nuñez *et al.* 2000), Belize (Foster *et al*. 2010), Guatemala (Novack *et al.* 2005), Costa Rica (Chinchilla 1997), and Brazil (De Azevedo 2008); *Mazama* spp. have also been among the most frequently reported prey from Belize (Foster *et al*. 2010), Guatemala (Novack *et al*. 2005), Costa Rica (Chinchilla 1997) and Brazil (Crawshaw and Quigley 2002, De Azevedo 2008). As in our study, other studies have frequently identified medium-sized rodents (*Dasyprocta* spp. and *Cuniculus* spp.) species in the diet, with examples from Panama (Moreno *et al.* 2006), Bolivia (Pacheco 2004), and Brazil (De Azevedo 2008). Overall, with slight taxonomic differences and some frequency variation, puma's diet in our study followed patterns similar to that in other regions and ecosystems in the Neotropics.

We found very similar results to those from the only other study on puma's diet in Colombia (Puracé NNP; Hernández-Guzmán *et al*. 2011), likely because of the highly similar ecosystems sampled. In Puracé NNP the most frequent prey species found were northern pudú (*Pudu mephistophiles*), *N. olivacea*, and *Mazama* sp. (likely *M. rufina*). Differences in species composition may reflect that Northern Pudú is not present in the Eastern Andean range of Colombia (Barrio and Tirira 2008) and *N. nasua* is not abundant above 3,000 masl (González-Maya *et al.* 2015); however, *Mazama* sp. was also among the most frequent prey species in Puracé NNP, and the largest contribution to biomass consumed was also *N. olivacea* and *Mazama* sp. as in our results. Previous published studies from the high Andes are scarce, so the record of *C. taczanowskii* can be considered as the first record of this species in the puma's diet across its distribution.

It is important to note that the consumption of *Coendou rufescens* by puma has been previously reported in the páramo of Belmira (2,800 to 3,000 masl, Antioquia department, north of the Central Andean range; Arias-Alzate pers. obs.); although this species is not found in Tamá NNP and it was not found on the scats collected; a similar species occurs in the area (*i. e., Coendou pruinosus*), so it could be expected in the diet. Chinchilla (1997) also highlighted a frequent consumption of *Coendou mexicanus* in the lowlands of Corcovado National Park, Costa Rica. Other potential prey species that were expected to be part of the puma's diet in Tamá NNP, and that were previously recorded by camera traps, included *Didelphis pernigra, Sciurus granatensis, Conepatus semistriatus*, and *Mustela frenata* (Cáceres-Martínez *et al.* 2016). However, and despite they have been reported previously in the diet, we did not find them in the scats collected.

It seems that variations in puma's diet are the result of opportunistic predation by this species, adapting its feeding behavior according to prey species availability both at the latitudinal and the altitudinal levels (Iriarte *et al*. 1990). Moreover, at high elevations or in regions sympatric with jaguars, pumas tend to prey more frequently on medium-sized species such as agoutis (*Dasyprocta* spp.), pacas (*C. paca* and *C. taczanowskii*), coatis (*N. narica*, *N. olivacea* and *N. nasua*), and deer (*P. mephistophiles*, *Mazama gouazoubira* and *M. rufina*) and apparently only prey on large mammals and other small species to supplement their diet (Nuñez *et al.* 2000, Novack *et al*. 2005, De Azevedo 2008, Hernández-Guzmán *et al*. 2011). However, the preferred prey size may also be limited by environmental conditions; for instance, puma typically prey more frequently on large species (frequently deer) in temperate zones (Dalrymple and Bass 1996, Cunningham *et al.* 1999, Moreno *et al*. 2006).

For some carnivore species, such as large felids (*e. g.,* puma and jaguar), the lack of suitable habitats and prey species as a consequence of anthropic pressure (*e. g.,* deforestation and hunting) may promote predation on livestock (Hoogesteijn and Hoogesteijn 2005, De Azevedo 2008). It seems that both habitat availability and the prey base at Tamá NNP are likely sufficient to avoid such behaviors, given that no livestock or other domestic species were found on the scats; pumas in Tamá may have no need to use such alternative prey. In other regions of Colombia (*e. g.,* Antioquia), generally considered more impacted by humans, puma consumption of livestock has been frequently recorded (Arias-Alzate *et al.* 2013, González-Maya *et al.* 2013), indicating that conflict prevention, and the conservation of puma, depends on the maintenance of healthy ecosystems and local prey base via sustainable forest management and poaching control.

Our results represent significant knowledge of what is likely the most important Andean predator and they provide the baseline for understanding patterns of assemblage dynamics and thus for informing conservation. Changes in puma diet dynamics will likely reflect in changes in the composition and structure of mammal assemblages, not only represented as part of puma ecology, but also in general ecosystem dynamics and function. Our data can provide the base to understand the potential responses of species assemblages to changes in threats, both inside protected areas and surrounding areas, all increasingly dominated by human activities.

We highlight the importance of continuing long-term monitoring of these ecological processes, and in general with the generation of this type of studies in other regions of Colombia, to support better conservation strategies for species such as the puma, given the important roles it plays in ecosystem dynamics and stability in both protected and unprotected areas.

Acknowledgments

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Appendix 1

Appedix1. Species consumed by puma (*Puma concolor*) in Tamá National Natural Park Andean based on remnants of prey in scats (*n* = 45).

ID	*N. nasua*	*C. taczanowskii*	*N. olivacea*	*D. marsupialis*	*D. punctata*	*M. rufina*	*C. hoffmanni*	*D. novemcinctus*	Vertebrate
1	X	X	-	-	-	-	-	-	X
2	X	-	-	-	-	-	-	-	X
3	-	-	-	-	-	-	-	-	-
4	-	-	X	X	-	-	-	-	X
5	-	-	-	-	-	-	-	-	-
6	-	-	-	-	-	X	-	-	X
7	-	-	-	-	X	-	-	-	X
8	-	-	-	-	-	X	-	-	X
9	-	X	-	-	-	-	-	-	X
10	X	X	X	-	-	-	-	-	X
11	-	-	X	-	-	-	-	-	X
12	-	-	-	-	-	-	-	-	-
13	X	-	-	-	-	-	-	-	X
14	-	-	-	X	-	-	-	-	X
15	-	-	-	-	-	-	X	-	X
16	-	-	-	-	-	-	-	-	-
17	X	-	-	-	-	-	-	-	X
18	X	-	-	-	-	-	-	-	X
19	-	X	-	-	-	-	-	-	X
20	-	-	-	-	-	-	-	-	-
21	-	-	-	-	-	-	-	-	-
22	-	-	-	-	-	-	-	-	-
23	-	-	-	-	-	-	-	-	-
24	X	-	-	-	-	-	-	-	X
25	-	-	-	-	-	-	-	-	-
26	-	-	-	-	-	-	-	-	-
27	-	-	-	-	-	-	-	-	-
28	-	-	-	-	-	-	-	-	-
29	-	-	-	-	-	-	-	-	-
30	-	-	-	-	-	X	-	-	X
31	-	-	-	-	-	-	-	-	-
32	-	-	-	-	-	-	-	-	-
33	-	-	-	-	-	-	-	-	-
34	-	-	-	-	-	-	-	-	-
35	-	-	-	-	-	-	-	-	-
36	X	-	-	-	-	X	-	-	X
37	-	-	-	-	-	-	-	-	-
38	X	-	-	-	-	X	-	-	X
39	-	-	-	-	-	-	-	X	X
40	-	-	-	-	-	-	-	-	-
41	-	X	-	-	-	-	-	-	X
42	-	-	-	-	-	-	-	-	-
43	-	-	-	-	-	-	X	-	X
44	-	-	-	-	-	X	-	X	X
45	-	-	-	-	-	-	-	-	-
TOTAL	**9**	**5**	**3**	**2**	**1**	**6**	**2**	**2**	**23**

ID	*N. nasua*	*C. taczanowskii*	*N. olivacea*	*D. marsupialis*	*D. punctata*	*M. rufina*	*C. hoffmanni*	*D. novemcinctus*	Vertebrate
1	X	X	-	-	-	-	-	-	X
2	X	-	-	-	-	-	-	-	X
3	-	-	-	-	-	-	-	-	-
4	-	-	X	X	-	-	-	-	X
5	-	-	-	-	-	-	-	-	-
6	-	-	-	-	-	X	-	-	X
7	-	-	-	-	X	-	-	-	X
8	-	-	-	-	-	X	-	-	X
9	-	X	-	-	-	-	-	-	X
10	X	X	X	-	-	-	-	-	X
11	-	-	X	-	-	-	-	-	X
12	-	-	-	-	-	-	-	-	-
13	X	-	-	-	-	-	-	-	X
14	-	-	-	X	-	-	-	-	X
15	-	-	-	-	-	-	X	-	X
16	-	-	-	-	-	-	-	-	-
17	X	-	-	-	-	-	-	-	X
18	X	-	-	-	-	-	-	-	X
19	-	X	-	-	-	-	-	-	X
20	-	-	-	-	-	-	-	-	-
21	-	-	-	-	-	-	-	-	-
22	-	-	-	-	-	-	-	-	-
23	-	-	-	-	-	-	-	-	-
24	X	-	-	-	-	-	-	-	X
25	-	-	-	-	-	-	-	-	-
26	-	-	-	-	-	-	-	-	-
27	-	-	-	-	-	-	-	-	-
28	-	-	-	-	-	-	-	-	-
29	-	-	-	-	-	-	-	-	-
30	-	-	-	-	-	X	-	-	X
31	-	-	-	-	-	-	-	-	-
32	-	-	-	-	-	-	-	-	-
33	-	-	-	-	-	-	-	-	-
34	-	-	-	-	-	-	-	-	-
35	-	-	-	-	-	-	-	-	-
36	X	-	-	-	-	X	-	-	X
37	-	-	-	-	-	-	-	-	-
38	X	-	-	-	-	X	-	-	X
39	-	-	-	-	-	-	-	X	X
40	-	-	-	-	-	-	-	-	-
41	-	X	-	-	-	-	-	-	X
42	-	-	-	-	-	-	-	-	-
43	-	-	-	-	-	-	X	-	X
44	-	-	-	-	-	X	-	X	X
45	-	-	-	-	-	-	-	-	-
TOTAL	**9**	**5**	**3**	**2**	**1**	**6**	**2**	**2**	**23**

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Presence of organochlorine pesticides and characterization of biomarkers in wild mice living in crop fields

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The use of pesticides in crops bordering conservation areas poses risks for wildlife incidentally exposed; its effects in the Yucatan Peninsula, Mexico, are still unknown. Wild mice that inhabit farming land play a key ecological role, and can also be used as bioindicators of wildlife exposure to pollutants. The objectives of this work were to determine the presence of organochlorine pesticides (OC) in liver and evaluate the seasonal response of enzymatic biomarkers (BM) such as acetylcholinesterase (AChE), glutathione-S-transferase (GST), and catalase (CAT). Wild mice (*Mus musculus*) were captured between June 2015 and April 2016 in a watermelon crop of a rural community in Quintana Roo, Mexico. Individual mice were sacrificed *in situ*, followed by tissue dissection (liver, brain and skeletal muscle). Pesticides were determined by gas chromatography; BM activity was estimated by spectrophotometry. We captured 35 individuals, with a capture success of 2.33%. The prevailing OCs detected were drines in both climatic seasons. The rainy season influenced the activity of biomarkers to a greater extent, since AChE showed a lower activity (16 % and 40 % in brain and liver, respectively). GST was activated during the same season (77 % higher), while CAT did not show significant differences between seasons. There was no significant correlation between OC concentrations and biomarker activity, except for drines and AChE in the brain. OC concentrations recorded in the present work are below (2- and 20-fold lower) those reported in other works on rodents under controlled conditions. BM activity suggests that rainfall seem to exacerbate the effects of pesticides on mice; however, it seemingly does not pose a risk for their survival. The use of wild mice as bioindicators is a valuable and practical tool to detect disturbances derived from the use of pesticides in agricultural areas. Further research is recommended using a broader BM battery to identify those pollutants with the most severe effect on the physiology of wild animals incidentally exposed to pesticides.

El uso de plaguicidas en cultivos aledaños a áreas de conservación, significa un riesgo para la fauna silvestre expuesta accidentalmente cuyos efectos son desconocidos, particularmente en la Península de Yucatán, México. Los ratones silvestres que habitan las regiones agrícolas, desempeñan un papel ecológico importante, además pueden servir como bioindicador de exposición a contaminantes. Los objetivos del presente trabajo fueron determinar la presencia de plaguicidas organoclorados (OC) en hígado y evaluar la exposición a partir de la respuesta temporal de los biomarcadores enzimáticos (BM), tales como acetilcolinesterasa (AChE), glutatión-S-transferasa (GST) y catalasa (CAT). Ratones silvestres (*Mus musculus*) fueron capturados entre junio de 2015 y abril de 2016 en un cultivo de sandía de una comunidad agrícola, en Quintana Roo, México. Los individuos fueron sacrificados in situ y se le diseccionaron los tejidos (hígado, cerebro y músculo esquelético). La presencia de plaguicidas se determinó por cromatografía de gases. La actividad de los BM se determinó por espectofotometría. Se capturaron 35 individuos, con un éxito de captura de 2.33%. Los OC más detectados fueron los drines en ambas temporadas climáticas. La temporada lluviosa influyó más en la actividad de los biomarcadores, ya que la AChE mostró menor actividad (16 y 40 % en cerebro y músculo respectivamente), la GST aumentó su actividad para la misma temporada (77 % mayor), y finalmente la CAT no mostró diferencias significativas entre temporadas. No hubo correlación significativa entre las concentraciones de OC y la actividad de BM, a excepción de los drines y la AChE en cerebro. Las concentraciones de OC encontradas en el presente trabajo, están por debajo (entre 2 y 20 veces) de las reportadas en otros trabajos con roedores en condiciones controladas. La actividad de los BM refleja que las lluvias parecen acentuar los efectos de los plaguicidas sobre los ratones, sin embargo, puede no representar un escenario de riesgo para su sobrevivencia. La utilización de ratones silvestres como bioindicadores, es una herramienta útil y práctica para vislumbrar disturbios ocasionados por el uso de plaguicidas en zonas agrícolas. Se recomiendan trabajos complementarios y ampliar la batería de BM, para poder determinar con mayor precisión cuales son los contaminantes que afectan en mayor medida la fisiología de los animales silvestres expuestos accidentalmente.

Keywords: Acetylcholineterase; catalase; glutathione-S-transferase; biomarkers; *Mus musculus*; organochlorine pesticides; wildlife

Introduction

Technology-dependent agriculture (hereinafter called tech agriculture) involving the use of high amounts of pesticides poses a risk to wildlife that comes into contact with these toxic compounds, which may cause still unknown health effects. The continued application of pesticides and other agrochemicals to crop fields causes acidification, loss of soil biodiversity, and soil toxicity (Givaudan *et al.* 2014). Pesticides not only affect the target pest species, but also impact wildlife living in and around agricultural fields (Johnston, 2000). The damage caused by pesticides on exposed organisms depend mainly on their toxicokinetics (uptake route and distribution) and toxicodynamics (effects on physiology and metabolism) and is influenced by external factors such as pesticide nature, chemical composition, persistence, dose, and time of exposure, as well as by the health status and susceptibility of the exposed organism (Ramírez and Lacasana 2001). The main characteristics of organochlorine pesticides include high persistence, mobility across great distances from their source of application, bioaccumulation, and long-term adverse effects (Mackay *et al.* 2001).

In this sense, the vertebrate group with the most severe adverse effects from incidental poisoning in the wild documented to date (Köhler and Triebskorn 2013), since they are deemed highly sensitive to pesticide exposure. In contrast, a group that has received little attention as to harm caused by pesticides is rodents, mainly because these are also considered as pests; however, this neglects the important ecological roles they play, mainly regarding seed dispersal and control of other pests such as insects and other invertebrates, besides being preyed by higher predators (Ceballos 2005).

In general, small mammals have been used as proxies to validate human exposure models by various environmental protection organizations. For example, the USEPA (United States Environmental Protection Agency) has commonly used rodents as indicators of human and environmental protection (Sheffield *et al.* 2001). Particularly, murine models have been used in the characterization of various effects of pesticides under controlled conditions. These comprise the evaluation of tolerance thresholds by determining the lethal dose 50 (LD_{50}) — the dose that kills 50% of the test population (Dell'Omo *et al.* 2003; Hirano *et al.* 2017) — and adverse effects by endocrine disruption (Bhaskar and Mohanty 2014; Ghodrat *et al.* 2014), oxidative stress (Zhao *et al.* 2016; Morales-Prieto and Abril 2017; Latchoumycandane and Mathur 2002), genotoxic effects (Peris-Sampedro *et al.* 2016), or neurotoxic effects (Chen *et al.* 2016; Dell'Omo and Shore 1996; Westlake *et al.* 1983). All these studies have involved the application of various doses of one or more (mixtures) pesticides.

However, studies that consider mice as bioindicators of environmental conditions for wildlife are scarce. Population dynamics of rodents has been used as an indicator of disturbance in polluted areas. These studies have focused mainly on accidental deaths and population declines caused by acute exposure to organophosphate pesticides (OF) and carbamates (CB) (Barrett and Darnell 1967; Giles 1970; Sheffield *et al.* 2001). For example Block *et al.* (1999) assessed the long-term population impact of the pesticide OF Counter® (terbufos), based on population parameters in two wild mice species (*Peromyscus maniculatus* and *P. leucopus*) in an agricultural field in Iowa, USA, and concluded that OF has no apparent influence on reproduction and other population parameters that would affect their long-term demography. Other studies conducted in free-living mice evaluated the accumulation of persistent organic pollutants (POPs) in tissues (Perez-Gonzalez *et al.* 2017), or the effect of various pollutants at the histological level (Gomez-Ugalde 2003), or with a biochemical approach by analyzing enzymatic biomarkers (BM; Andrade-Herrera 2011; Chi Coyoc *et al.* 2016).

In Mexico, studies reporting the influence of crop pesticides on wild rodents are scarce. Specifically in the Yucatan peninsula, these studies focus on the presence and accumulation of pesticides (Rendon-von Osten *et al.* 2005; Charruau *et al.* 2013), and relatively few studies report harmful effects to the fauna surrounding farming areas where pesticides are used (Noreña-Barroso *et al.* 2004; Cobos *et al.* 2006; Buenfil-Rojas *et al.* 2016). Accordingly, in the state of Quintana Roo, particularly in the Maya area, the main tech crops are sugar cane, Maradol papaya and watermelon (SAGARPA 2015). These involve the use of various technological packages based on the use of agrochemicals to achieve high crop yields. For example, Endosulfan 35 Le Cag® (endosulfan), Furadan® (carbofuran), Herbipol® (2.4-D Amine) and Cerillo® (paraquat), Lannate® (methomyl) or Velfosato® (glyphosate) were recorded during field trips as the main pesticides used. It was noted that these pesticides are applied in excessive amounts, and farmers apply them disregarding the safety requirements (personal protection equipment).

Therefore, the lack of studies in the region analyzing the effects of tech agriculture on wildlife health, and in particular as regards small mammals, made it necessary to generate knowledge to provide evidence about the potential risks for these animals. Thus, the aim of the present study was to analyze the influence of the pesticides used in a tech watermelon crop on wild mice (*Mus musculus*). This was done by first determining the presence of pesticides in liver tissue of individuals captured, followed by characterizing the toxic effects through enzymatic biomarkers (AChE in brain and muscle; GST and CAT in liver), and finally by exploring the relationship between the pesticides detected and the activity of biomarkers.

Materials and Methods

Field Work. The sampling site is located at coordinates 19° 35' 24.70" N, -89° 10' 51.20" W, in the Municipality of José María Morelos, Quintana Roo, in a tech watermelon crop within the Mayan community named *X noh Cruz* (Figure 1). The area is surrounded by rainfed maize fields and forest patches, and is adjacent to the *Bala'Ka'ax* Wildlife Protection Area (APFFBK).

Specimens were captured between June 2015 and April 2016 using 50 Sherman® traps placed every five meters on both sides along a 125 m transect across the crop field. Traps were baited with a mixture of oats and vanilla and left overnight. The individuals captured were identified using field guides, and standard morphometric measurements were recorded. Then, these individuals were killed *in situ* by cervical dislocation according to NOM-033-SAG/ZOO- 2014, which stipulates the humane killing of domestic and wild animals; brain, liver and skeletal muscle samples were taken

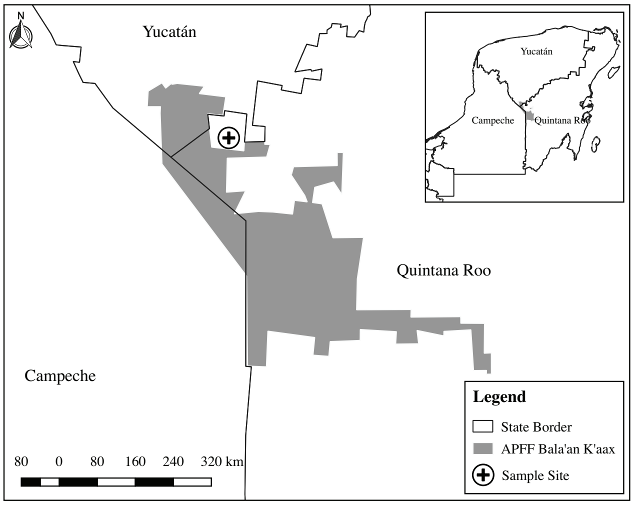

Figure 1. Sampling area, located in coordinates 19° 35' 24.70" N, -89° 10' 51.20" W, within the Mayan community of Xnoh Cruz, municipality of José María Morelos, Quintana Roo, which is adjacent to the *Bala'an 'k'aax* APFF.

for subsequent analysis. Tissues were stored in liquid nitrogen, transported to the laboratory, and stored at -80 °C.

Laboratory work. Laboratory work was divided into two phases: the first determined the presence of organochlorine pesticides (OC) in mouse liver tissue; the second characterized the effect of exposure to the various xenobiotics by assessing three biomarker enzymes: acetylcholinesterase (AChE) that reflects neurotoxicity, glutathione-S-transferase (GST) that indicates detoxification processes (phase II biotransformation), and catalase (CAT), an oxidative stress biomarker.

Determination of Pesticides in Liver Tissue. High-purity solvents were used (98 % HPLC); glass material was washed with Extran® and rinsed with distilled water, then dried for 4 hours at 200 °C and rinsed with acetone and hexane.

The analyses of pesticides in liver tissue of mice were performed according to the method described by Zhang *et al.* (2007). A fraction of liver tissue was collected, oven-dried for 24 h at 45 °C, and weighed for subsequent analysis. Any chemicals in samples were extracted with a mixture of acetone:hexane (1:1) in a sonicator for 20 min. After extraction, the amount of fat was determined to estimate the concentrations of pesticides on fat basis (µg/g of fat). Samples were purified through a glass column packed with 7 g of silica gel and 1 g of sodium sulfate. Afterwards, samples were eluted with 20 ml of hexane, 20 ml of a 1:1 mixture of dichloromethane and hexane, and 20 ml of dichloromethane; then, samples were immediately evaporated to dryness and resuspended in 50 µl of hexane for testing through gas chromatography. Pollutants were quantified using a Varian 3800 gas chromatograph with a Ni^{63} electron capture detector (GC-ECD) and equipped with a HT8 capillary column (60 m x 0.25 mm; 25 µm film thickness; SGE Analytical Science, USA). Injector and detector temperatures were 150 °C and 350 °C, respectively. Pollutant concentrations were calculated through the area under the curve with the software Star Chromatography Workstation version 6, and through the calibration of standards. Pesticides were identified and quantified using a mixture of 20 OC pesticide standards, CRM No. SUPELCO® 47426 EPA CLP Organochlorine Pesticide Mix, and the Sigma-Aldrich standards Mirex No. 45887, 2.4'-DDE No. 36663, 2.4'-DDD No. 35485 and 2.4'-DDT No. 45839, which were pooled into families for testing (Table 1).

Activity of enzymatic biomarkers (BM). The three BM were analyzed in specific tissues according to their target organ, with AChE tested in brain and skeletal muscle, while GST and CAT were analyzed in liver. For the three BM, all samples were homogenized for 20 sec. in a homogenizer (PRO Scientific 250®), using the specific buffer for each BM according to the method used. AChE activity was determined as per the method of Ellman *et al.* (1961), adapted to microplate (Guilhermino *et al.* 1996); samples were prepared with a 0.1 M potassium phosphate buffer, pH 7.2. Enzyme kinetics was recorded at 414 nm and expressed in units (U) of hydrolyzed acetylcholine per minute per mg of protein using an extinction coefficient of 13.6 x 10^3 M^{-1} cm^{-1}. GST activity was determined according to the method described by Habig *et al.* (1974). A 0.1 M solution of potassium phosphate buffer, pH 6.5, was used for sample preparation, and enzyme kinetics was read at 340 nm, expressing activity as U of S-(2.4-dinitrophenyl) glutathione per minute per milligram of protein, using an absorption coefficient of 9.6 x 10^3 M^{-1} cm^{-1}. CAT activity was measured through the decrease in H_2O_2 at 240 nm using the method by Aebi (1984) for example, methanol, ethanol, formic acid, phenols, with the con-

Table 1. Organochlorine (OC) pesticides analyzed in liver tissue of *Mus musculus*; LD = limit of detection, LR = Limit of Report

Isomer and/or metabolites analyzed	LD µg/g	Group	LR µg/g
α HCH	0.003	HCHs	0.007
β HCH	0.004		
χ HCH	0.003		
δ HCH	0.002		
Aldrin	0.004	DRINES	0.0018
Dieldrin	0.004		
Endrin	0.009		
Endrin aldehyde	0.010		
Endrin ketone	0.005		
Endosulfan I (alpha)	0.005	Endosulfans	0.007
Endosulfan II (beta)	0.006		
Endosulfan sulfate	0.005		
o,p' DDE	0.005	DIdd	0.001
p,p' DDE	0.004		
o,p' DDD	0.031		
p,p' DDD	0.025		
o,p' DDT	0.013		
p,p' DDT	0.010		
Trans Chlordane	0.004	Chlordanes	0.009
Cis Chlordane	0.004		
Methoxychlor	0.018	Methoxychlor	0.01
Mirex	0.006	Mirex	0.006
Heptachlor	0.004	Heptachlor	0.013
Heptachlor epoxide	0.004		

sumption of 1 mol of peroxide (peroxide activity, using a 50 mM solution of potassium phosphate buffer of pH 7, with activity expressed as U of hydrogen peroxide produced per minute per milligram of protein. Protein concentration was determined by the method of Bradford (1976) at 595 nm, expressed as mg/ml. In all cases, activity was determined using microplates (Corning®) in a spectrophotometer (Thermo Scientific Multiskan Spectrum®).

Statistical analyses. All analyzes were performed using the software R version 3.2.4 (R Core Team, 2016). Compliance of BM activity with the assumption of a normal distribution was tested using a Shapiro-Wilk test, and differences were evaluated by comparing BM activity between climatic seasons (raint-dry), using a Student's *t*-test when the data were normally distributed, or with a Wilcoxon test otherwise (Table 2); the significance level used was $\alpha \leq 0.05$ (Zar, 2010). Subsequently, the measures of central tendency and dispersion of pesticides were determined with the package NADA 1.6-1 (Lopaka 2013), taking into account left-censored data according to the chromatograph detection limit.

Table 2. Average activity of BMs per climatic season, with values expressed as U/min/mg protein. Also shown are the statistical *t* values (parametric), W (non-parametric); SD = standard deviation; max = maximum value; min = minimum value; DF = degrees of freedom; *p* = statistical probability; *≤ 0.05, **≤ 0.01, ***≤0.001.

Season	*n*	AChE in brain								
		Mean	SD	Max	Min	*t*	*W*	DF	*p*	α = 0.05
Rainy	17	16.81	4.55	26.86	7.43	-2.0752	-	31.99	0.046	*
Dry	17	20.07	4.63	29	1255					
		AChE in muscle								
Rainy	17	5.50	4.32	15.42	0.87	-3.1859	-	20.49	0.004	**
Dry	18	9.07	1.68	12.43	6.01					
		GST								
Rainy	17	98.36	74.80	208.60	15.75	-	253	-	0.001	***
Dry	18	30.53	9.48	52.60	10.63					
		CAT								
Rainy	17	6.15	5.76	17.99	1.19	-	127	-	0.404	-
Dry	18	6.24	3.94	15.72	1.32					

The "vegan" package (Oksanen *et al.* 2017) was used to perform correlations between the concentrations of pesticides detected (classified by chemical family) in liver and the BM analyzed. These correlations were explored using rank-transformed data (Spearman correlations), as concentration data were left-censored by the limits of detection and quantification (Helsel 2012).

Results and Discussion

This work reports the concentrations of pesticides in liver tissue (on fat, dry weight, and wet weight bases), as well as their potential effects characterized from the expression of three enzymatic BMs in three different wild-mice tissues. Adult individuals of *Mus musculus* were chosen as target species, because this was the dominant mouse species captured in sampling, as well as for its abundance in the study area, being a natural inhabitant in maize crop fields that surround watermelon crops (Alcérreca *et al.* 2009). The remaining animals were not deposited in any biological collection, given the poor condition of skin and skeleton (mainly the skull) after dissections. A total of 35 individuals were captured during field work (17 in the wet season and 18 in the dry season), over 30 nights of trapping, with a 2.33 % capture success.

Presence of OC Pesticides in Liver Tissue. Of the 24 OC pesticides analyzed, those most frequently detected in the wet season belong to the "drines" family (aldrin, endrin, endrin aldehyde, endrin ketone, and dieldrin), which were detected in 10 individuals, followed by hexachlorocyclohexanes (HCHs), detected in 7 specimens; in contrast, the least detected pesticide was methoxychlor, found in just two individuals. In the dry season, drines were also the pesticides most frequently detected in the 18 individuals analyzed, along with derivatives of dichloro-diphenyl-trichloroethane (DDT) and heptachlorine, with each detected in eight individuals; similar to the wet season, methoxychlor was the least detected pesticide, identified in only three specimens (Table 4).

On the other hand, the chemicals found in higher concentrations in both seasons were DDT isomers and methoxychlor, with peak concentrations of 702 and 524 µg/g fat, respectively, both in individuals caught in the dry season. The chemical recorded in the lowest concentration was chlordane in both seasons, with a mean concentration of 6.63 ± 11.3 µg/g fat and 15.02 ± 29.3 µg/g fat in the wet and dry seasons, respectively.

The concentrations found in this study are below the exposure-related toxicity thresholds (mainly *via* ingestion) reported in various investigations on wild rodents under controlled laboratory conditions (Jefferies *et al.* 1973; Morris 1968; Sheffield *et al.* 2001; Wolfe and Esher 1980), which are 2- to 20-fold higher versus those reported here. These works were conducted primarily to set lethal doses and tolerances to various OC including drines, HCHs and DDTs; these works report both physiological and behavioral impairment, but only some report OC content in tissues.

A fact worth considering is that bioaccumulation in wild mammals is mainly driven by ingestion, although pollutants may also be absorbed through the skin or by inhalation (Linder and Joermann 2001). In this regard, the work on wild rodents in free-living conditions focus mainly on evaluating variations in population size or recording presence/absence in areas where OC pesticides have been used (Sheffield *et al.* 2001). The few studies that quantify the presence of OC in tissues have been carried out primarily to identify DDT, dieldrin and heptachlor residues. In the case of DDT, the values found in mice in this study (between 0.001 and 1.834 µg/g dry weight) are similar to those reported by Lincer and Sherburne (1974) wet weight, ranging between 0.01 and 0.13 µg/g dry weight. In addition, Laubscher *et al.* (1971), in crop fields in Tucson, Arizona, found DDT and its metabolites at concentrations between 0.006 to 0.92 µg/g wet weight in liver of *Peromyscus eremicus* and *P. man-*

Table 4. Summary of pollutant concentrations (µg/g) in the liver of *Mus musculus*. OC= Organochlorine pesticide; (D= number of data detected; ND= number of data not detected; SD = standard deviation.

Weight Fat

	Rainy							Dry						
OC	D	ND	ND %	Min	Max	Mean	SD	D	ND	ND %	Min	Max	Mean	SD
HCHs	7	10	58.820	0.007	82.213	10.340	28.370	5	13	72.222	0.007	303.116	23.370	75.940
DRINES	10	7	41.176	0.002	135.532	21.690	43.936	8	10	55.555	0.002	279.350	22.691	66.579
ENDOSULFAN	4	13	76.470	0.007	89.165	23.127	27.844	5	13	72.222	0.007	195.783	18.027	48.257
DDTs	4	13	76.470	0.010	303.812	39.522	111.434	8	10	55.555	0.010	702.315	45.947	170.208
CHLORDANES	9	8	47.058	0.009	32.109	6.633	11.318	6	12	66.666	0.009	120.856	15.023	29.304
HEPTACHLOR	5	21	70.588	0.013	54.537	10.606	17.970	8	10	55.555	0.013	227.825	18.473	54.433
METHOXYCHLOR	2	15	88.235	0.010	232.528	232.530	0.010	3	15	83.333	0.010	524.625	60.177	137.962

Wet Weight

	Rainy							Dry						
OC	D	ND	ND %	Min	Max	Mean	SD	D	ND	ND %	Min	Max	Mean	SD
HCHs	7	10	58.820	0.005	0.023	0.009	0.007	5	13	72.222	0.004	49.930	2.770	12.780
DRINES	7	10	58.820	0.002	0.042	0.009	0.013	8	10	55.555	0.002	0.079	0.012	0.024
ENDOSULFAN	4	13	76.470	0.007	0.031	0.028	0.001	5	13	72.222	0.007	0.080	0.015	0.022
DDTs	4	13	76.470	0.001	0.094	0.019	0.031	8	10	55.555	0.001	0.200	0.021	0.057
CHLORDANES	9	8	47.058	0.004	0.357	0.028	0.087	6	12	66.666	0.003	0.064	0.010	0.016
HEPTACHLOR	5	21	70.588	0.003	0.017	0.007	0.009	8	10	55.555	0.003	0.065	0.009	0.014
METHOXYCHLOR	2	15	88.235	0.010	0.072	0.072	0.000	3	15	83.333	0.008	0.148	0.016	0.040

Dry Weight

	Rainy							Dry						
OC	D	ND	ND %	Min	Max	Mean	SD	D	ND	ND %	Min	Max	Mean	SD
HCHs	7	10	58.820	0.007	0.035	0.011	0.009	5	13	72.222	0.007	0.125	0.019	0.029
DRINES	7	10	58.820	0.002	0.057	0.012	0.018	8	10	55.555	0.002	0.943	0.066	0.229
ENDOSULFAN	4	13	76.470	0.007	0.037	0.037	0.001	5	13	72.222	0.007	1.040	0.070	0.263
DDT	4	13	76.470	0.001	0.127	0.025	0.043	8	10	55.555	0.001	1.834	0.122	0.449
CHLORDANES	9	8	47.058	0.006	0.765	0.054	0.189	6	12	66.666	0.009	0.821	0.060	0.202
HEPTACHLOR	5	21	70.588	0.004	0.023	0.008	0.010	8	10	55.555	0.004	0.094	0.013	0.021
METHOXYCHLOR	2	15	88.235	0.010	0.097	0.097	0.000	3	15	83.333	0.010	0.216	0.028	0.056

iculatus; these levels are up to five-fold higher than those recorded in this study (0.0001 to 0.200 µg/g wet weight).

However, the concentrations found here are far lower than those reported by Chi Coyoc *et al.* (2016) for DDTs, HCHs and drines (0.876, 0.546, and 0.451 µg/g dry weight basis respectively) in liver of wild rodents (*Oryzomys couesi, Peromyscus leucopus* and *Reithrodontomys gracilis*). These concentrations were up to 35 (DDT), 49 (HCHs) and 37 (Drines) times higher than those recorded in this study. These results suggest that the concentrations observed in this case may not pose a risk for the rodents exposed; however, the effects of the various pollutants are influenced by a number of factors — both environmental and physiological intrinsic to each species and even to each individual (Peakall and McBee 2001).

Finally, in general terms, there is a higher concentration of all OCs in the dry season than in the rainy season (Table 4). This could be due to the fact that rain leads to the partial leaching of toxic compounds to greater soil depths, thus reducing the exposure of organisms that live at the surface (González Valdez *et al.* 2013). An additional factor is the clayey silt soil in the sampling area (Andrade Herrera *et al.* 2018), which favors the leaching of pollutants given its low organic matter content, which adsobs OCs and makes them more persistent in upper soil layers (Cárdenas and Márquez 2015). Furthermore, factors associated with temporality may also account for the higher concentration of OCs in the dry season; for example, this coincides with the winter season, during which organisms move less due to the low temperatures (most reproductive periods occur in the rainy season), so that fat content of organisms increases (Caldas *et al.* 1999).

Activity of Enzymatic Biomarkers. As regards BMs, significant differences were found between climatic seasons in AChE and GST, while CAT did not record these differences (Table 2). AChE was determined in both brain and muscle, registering a higher mean activity (18.32 U/min/mg protein) in the brain relative to the muscle (7.35 U/min/mg protein), representing a difference slightly exceeding two orders of magnitude between the two tissues. However, both tissues show the same pattern, *i.e.* the activity recorded was higher in the dry season vs. the rainy season (Figure 2). In the brain, AChE showed an inhibition slightly above 16 % ($t_{(32)} = -2.075$, $p < 0.05$) in the wet season; in the muscle, this inhibition was about 40 % ($t_{(20)} = -3.18$, $p < 0.01$), also in the rainy season (Table 2).

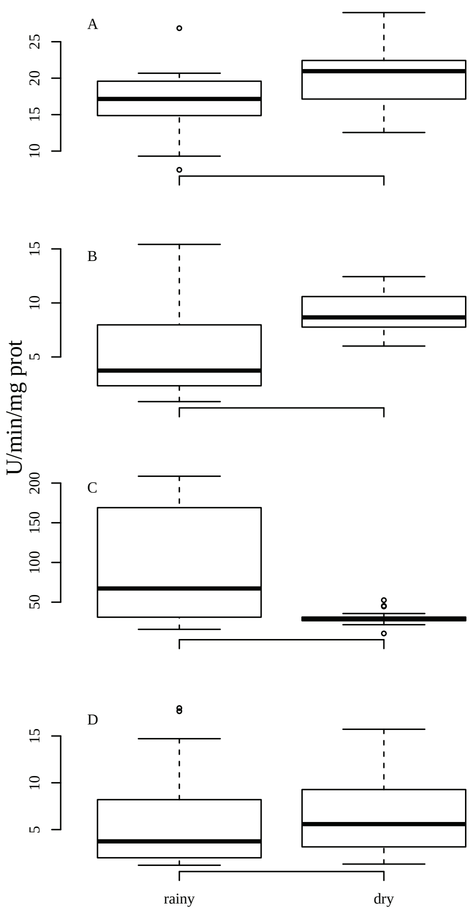

Figure 2. Activity of BMs in tissues of *Mus musculus* captured in agricultural fields. Data were compared between climatic seasons; AChE was evaluated in (A) brain and (B) muscle, while GST (C) and CAT (D) were analyzed in liver. Results are shown as median values, quartiles (box); activity is expressed as U/minute/mg protein.

AChE is considered to be the best BM for assessing exposure to organophosphate and carbamate pesticides — the main anticholinergic pesticides. However, their characteristics (poorly persistent in the environment, highly soluble in water) make it difficult to quantify them in both the environment and animal tissues (Sheffield *et al.* 2001). Rattner and Hoffman (1984) report the inhibition of AChE after the administration of sublethal oral doses of the organophosphate (OF) insecticide acephate to wild mice (*M. musculus, P. leucopus* and *M. pennsylvanicus*); inhibition rates were similar to those reported in this study (16 to 40 %) at doses between 25 mg/kg and 100 mg/kg of food for 5 days. In addition, the inhibition rates recorded in our results are seemingly harmless for the mouse populations, as in other investigations an inhibition rate of more than 40 % was found, which was not lethal in exposed mice (Dell'Omo and Shore 1996; Meyers and Wolff 1994).

Studies based on field work that characterize the inhibition of AChE in wild rodents are scarce. In this sense, our results on AChE inhibition (16 to 40 %) are consistent with some studies carried out in the wild in other rodent species such as *Microtus pennsylvanicus* (Jett 1986), *M. pinetorum* (Durda *et al.* 1989), *P. maniculatus* and *P. leucopus* (Block *et al.* 1999), *M. musculus* (Edward *et al.* 1983; Custer *et al.* 1985), and particularly in Mexico in *O. couesi* and *R. gracilis* (Chi Coyoc *et al.* 2016). In these works, it is concluded that the degree of inhibition found did not cause lethal acute poisoning, and population stability was not affected.

In the present work, AChE activity decreases during the wet season, when the main crops are maize, squash, and beans, involving the use of herbicides such as glyphosate or paraquat in soil preparation, in addition to insecticides for the protection of crops, including carbofuran, endosulfan or imidacloprid. The data recorded point to a higher exposure to anticholinergic chemicals in the rainy season, which, although not analyzed here, were reported by farmers as used for pest control.

Various xenobiotics cause oxidative stress from the production of reactive oxygen species (Limon-Pacheco and Gonsebatt 2009). To avoid oxidative stress, cells activate detoxification processes through antioxidants in order to reduce toxic effects, involving the two enzymes used as BMs, GST (phase II of xenobiotic metabolism) and CAT (antioxidant), *i. e.,* these increase their activity when exposed to toxic compounds (van der Oost *et al.* 2003). GST activity in the mice captured exhibited significant differences between climatic seasons (W = 253, $p < 0.001$), showing a higher activity in the wet vs. dry season (98.36 and 30.53 U/min/mg protein, respectively), representing a difference of nearly 77 % in BM expression between both seasons (Table 2, Figure 2).

When GST is activated, it acts on toxic chemicals by increasing their water solubility, thereby facilitating their excretion from the body (van der Oost *et al.* 2003). The influence of pesticides on GST in mice in agricultural areas is poorly documented. A study characterized its activation in agricultural workers after exposure to carbamates and γHCH (Lukaszewicz-Hussain 2010); the latter is one of the pesticides found in tissues of some mice in this work. In this regard, GST activity increased significantly in the rainy season, suggesting that rainfall leads to the release of some xenobiotics trapped in soil, leading to an increased reaction of detoxification enzymes such as GST. A similar pattern was found in wild mice (*Peromyscus* spp.) in forested areas of Mexico City where GST activity was significantly higher during the rainy season (Andrade-Herrera 2011).

Contrary to all the BMs discussed above, CAT yielded no significant differences between the dry and rainy seasons (6.15 and 6.24 U/min/mg protein, respectively). However, the average values recorded in this work are higher than those reported in other (experimental) studies, where mean activity ranges from 0.2 to 2.5 U/min/mg protein (Ayed-Boussema *et al.* 2012; Panemangalore and Baby 2000; Yilmaz *et al.* 2004). In this regard, catalase is a biomarker that can react in different ways, *i. e.*, inhibition or activation, depending on various factors, mainly the dose of the xenobiotic, time of exposure, or a combination of various compounds (van der Oost *et al.* 2003).

In laboratory experiments, dermal exposure to various pesticides leads to increased CAT activity. For example, Panemangalore and Baby (2000) recorded a 13 % increase in activity relative to its controls after 4 weeks of exposure to a mixture of OFs. Similar effects were found in other studies where rats and mice were exposed to a variety of pesticides (OF, OC, pyrethroids and neonicotinoids, among others) via ingestion, with average increases in activity of 25 % (Jin *et al.* 2011, 2013). When pesticides are administered intraperitoneally, both effects occur. For example, Ayed-Boussema *et al.* (2012), recorded increased activity levels of up to three orders of magnitude in mice after the administration of multiple doses of dimethoate for 30 days; in contrast, Yilmaz *et al.* (2004) found a significant CAT inhibition after administering a systemic hormone herbicide for three days in albino mice (*Mus musculus*).

Correlation between Pesticides and BM Activity. Spearman's correlations were performed between the OC pesticides detected (per family) and the activity of BMs recorded in mice to explore the influence of these pollutants in the activity of these BM in exposed organisms. The analyses showed a single significant correlation (*rho* = -0.489, $p <$ 0.01), between drine pesticides and AChE activity in brain tissue (Table 3). In this sense, although exposure to OC pesticides causes multiple toxic effects on organisms, AChE inhibition is caused primarily by exposure to OF and CB pesticides (Rodríguez-Castellanos and Sánchez-Hernández 2007), which were not analyzed in the present work. However, farmers mentioned the use of mixtures of various insecticides and acaricides, including organophosphates, pyrethroids, carbamates, and neonicotinoids, particularly in watermelon crops; in addition, empty containers of these pesticides were found in agricultural fields, suggesting that these pollutants (or their mixtures) may account for the results observed.

In Mexico, few works report the effect of xenobiotic agents in wild rodents. For example, Andrade-Herrera (2011) correlated the concentrations of air pollutants in Mexico City with the activity of enzymatic biomarkers, finding a greater impact in the dry season, likely due to thermal inversions, as airborne pollutants may cause adverse effects. In contrast, this study suggests that rainfall seemingly affects to a greater extent the physiology of mice, as AChE and GST were particularly affected in the wet season. Rainfall carries and deposits toxic chemicals in soil, hence increasing their availability for rodent living in the local environment.

Table 3. Spearman's correlations between concentrations of organochlorine pesticides and characterization of the activity of the BMs analyzed in mice. Figures in bold represent significance at $p \leq 0.05$.

	AChE in brain		AChE in muscle		GST		CAT	
	Rho	p	Rho	p	Rho	p	Rho	p
HCHs	0.036	0.842	-0.014	0.937	0.043	0.805	-0.260	0.131
DRINES	-0.489	0.003	-0.084	0.630	0.118	0.501	0.172	0.323
ENDOSULFAN	-0.240	0.172	-0.259	0.133	-0.036	0.839	-0.079	0.653
Dldd	-0.093	0.601	0.022	0.899	0.008	0.965	0.061	0.730
Chlordanes	-0.392	0.022	-0.260	0.131	0.062	0.724	0.010	0.954
Heptachlor	-0.186	0.293	-0.119	0.497	-0.058	0.741	0.033	0.849
Methoxychlor	-0.192	0.277	-0.068	0.697	0.099	0.573	-0.072	0.680

Conclusions

This work is the first in Mexico to show the presence of pesticides and their biochemical effects (neurotoxicity and defense response mechanisms and antioxidants) in wild mice. The study shows that mice bioaccumulate pesticides in their tissues, which poses a risk to wildlife living in agricultural fields. The use of BMs is a valuable tool for determining sublethal hazard in wildlife due to exposure to pollutants. In this work, the activity of BMs shows a higher influence of pesticides in the rainy season, likely due to the presence of various pesticides in the environment that are deposited in soil with rain. Also, the use of wild mice as bioindicators is suitable for identifying effects associated with the use of pesticides in farming areas. For future studies, it is recommended to use blood as a non-destructive matrix, besides to the fact that blood contains chemicals that were recently ingested. Other types of pesticides should also be analyzed, in addition to broadening the battery of biomarkers used, to determine other potential adverse effects and, in turn, determine in further detail which pollutants are the major disruptors of the physiology of animals exposed, as the indiscriminate use of synthetic pesticides is likely to impact the surrounding wildlife.

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Potential distribution model of *Ovis canadensis* in northern Baja California, Mexico

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The bighorn sheep is an iconic species in Baja California, being a key element for environmental conservation across its distribution range due to the huge dimensions of its habitat. In this regard, priority areas should be identified to propose feasible management practices. In this context, ecological niche models are essential because they are important methodological tools that indicate the suitability of the habitat for proper species development, based on field observations and multiple environmental variables as occupancy predictors. This investigation aims to identify the potential distribution range of the bighorn sheep in Sierra de Juarez using an ecological niche model. Indirect signs of the presence of bighorn sheep were sampled in Sierra de Juárez from January to June 2016 in order to gather evidence of the species, along with records from an aerial survey carried out in 2012. The ecological niche model was constructed applying the maximum-entropy algorithm assisted with the Maxent software. Ruggedness, orientation, slope, normalized difference vegetation index (NDVI), type of vegetation, and type of weather were used as predictive variables. In Sierra de Juarez, bighorn sheep inhabit an area of 49,844 ha with the following characteristics: climates ranging from very arid semi-warm [BWh(x')] to very arid temperate [BWk(x') and BWks]; natural vegetation comprising gallery and palm-tree patches; NDVI of 0.05 to 0.07; orientation of 0 to160°; slope of 0 to 65 %; and ruggedness of 35 to160 m (Figure 4). NDVI, vegetation type and ruggedness were the variables with the greatest contribution to the ecological niche model (Table 1). Bighorn sheep are distributed primarily in the northern and central regions of Sierra de Juárez (Figure 2). According to the niche model, these areas have environmental conditions that provide shelter and resources for this species. Therefore, it is hereby proposed to organize the local landowners to implement actions to protect the bighorn sheep habitat to warrant the conservation of this iconic species and its environment in the region studied.

El borrego cimarrón es una especie emblemática la cual representa en sus zonas de distribución un elemento clave para la conservación de su entorno, que debido a las extensas dimensiones de su hábitat requiere de la ubicación de regiones prioritarias dentro de este, con el objetivo de proponer estrategias de manejo factibles. Es en este contexto que los modelos de nicho ecológico cobran importancia al ser herramientas metodológicas que indican la idoneidad del hábitat para el establecimiento de una especie, calculada a partir de observaciones de campo y una serie de variables ambientales que actúan como predictores. El objetivo de la presente investigación es identificar la distribución potencial para el borrego cimarrón en Sierra Juárez, empleando un modelo de nicho ecológico. Se realizó un rastreo de evidencias de la presencia del borrego cimarrón en Sierra Juárez de enero a junio de 2016, para generar una base de datos de registros geográficos de la especie, la cual se complementó con los registros obtenidos de un censo aéreo efectuado en el sitio en 2012. El modelo de nicho ecologico se generó mediante el algoritmo de máxima entropía ejecutado por el programa Maxent empleando como variables predictivas la escabrosidad, la orientación, la pendiente, el índice de vegetación de diferencia normalizada (NDVI), el tipo de vegetación y el tipo de clima. El borrego cimarrón en Sierra Juárez se distribuye en un área de 49,844 ha (Figura 2), donde su presencia es más probable en sitios con: climas muy árido semicálido [BWh(x')], muy árido templado [BWk(x')] y muy árido templado [BWks]; vegetación de tipo galería y palmar natural; NDVI de 0.05 a 0.07; orientación de 0 a 160 °; pendiente de 0 a 65 %; y escabrosidad de 35 a 160 m (Figura 4). y Siendo el NDVI, el tipo de vegetación y la escabrosidad, las variables de mayor contribución para la creación del modelo de nicho ecológico (Tabla 1). Potencialmente el borrego cimarrón se distribuye en las regiones del norte y centro de Sierra Juárez (Figura 2), ya que de acuerdo con el modelo de nicho estas zonas presentan condiciones ambientales que le brindan cobertura y recursos a la especie. Por lo que se propone organizar a los propietarios de estas regiones prioritarias para llevar a cabo acciones de protección y mantenimiento del hábitat, que aseguren la conservación de esta emblemática especie y su entorno.

Key words: desert bighorn sheep; ecological niche model; potential distribution model; Sierra de Juárez; wild sheep.

Introduction

In Mexico, the bighorn sheep is an iconic species naturally distributed across the Baja California peninsula and the state of Sonora; populations maintained under semi-captivity exist in the states of Chihuahua and Coahuila (Sandoval and Espinoza 2001; Sánchez 2005; Uranga and Valdez 2011). In these areas, this species contributes to the ecosystem production chain by consuming plant resources and as a source of energy for large predators and scavengers (Monson 1980). In addition, it represents a key element for the conservation of its habitat, mainly due to its importance as a game species, since the prices of permits for legal hunting are high, ranging between $45,000 and $60,000 USD (Lee 2011; Ruiz 2014).

The sustainable use of the species requires the development of a comprehensive and adaptive management plan, designed based on scientific information addressing the needs and interests of the managers of this resource (Walters 1986; Mandujano 1994). However, the dimensions of the bighorn sheep habitat are far too extensive to propose feasible management strategies, thus making it necessary to identify the sites of higher probability of occurrence of individuals of this species to focus conservation efforts on them (Lara *et al.* 2001; Escobar *et al.* 2015).

Studies on habitat preferences of bighorn sheep in the Baja California peninsula and Sonora, using use-availability methods, methodologies based on generalized linear models, and satellite remote sensing techniques, have shown that the species does not use habitat resources according to their abundance, as some are preferred while others are clearly overlooked. Elevation, slope, ruggedness and plant cover are the characteristics of the habitat that determine the sites preferred by the species (López *et al.* 1999; Lara *et al.* 2001; Guerrero *et al.* 2003; Escobar *et al.* 2015). Hence, the potential distribution of the species in a particular locality can be determined using an ecological niche model, as it is a representation of the environmental suitability for the establishment of a particular species, calculated from field observations (presence/absence) and the availability of a series of environmental variables (climate, topography, vegetation, among others) that act as predictors (Soberón and Peterson 2005; Ferrier and Guisan 2006; Naoki *et al.* 2006).

Although several methods to develop niche models are available, the one based on a statistical approximation named maximum entropy and run by the Maxent software (Phillips 2013) offers some unique advantages relative to others. It works with presence data only, while other models require presence and absence data; it shows a consistently good performance *versus* other similar methods, in differentiating between sites that are either suitable or unsuitable for the establishment of a particular species, especially with a small number of samples (Elith *et al.* 2006; Phillips *et al.* 2006; Hernández *et al.* 2008); also, it is freely available.

In the Baja California peninsula, the bighorn sheep is distributed from Sierra de Juarez on the border with the United States, to the Sierra de San Juan de la Costa adjacent to Bahía de La Paz (Sánchez 2005; Ruiz 2014). Sierra de Juarez is listed as a high-priority terrestrial region for biodiversity conservation in Mexico due to its physical and biotic characteristics; for being a key area for the functional connectivity for wildlife populations across the landscape, given its location in an international border; and because this mountain range is associated with six hydrological basins that flow to the main cities in the state of Baja California (CONABIO 1998; Arriaga *et al.* 2000; Buchalski *et al.* 2015).

The objective of this work was to construct a model of potential distribution for the bighorn sheep in Sierra de Juarez, to facilitate the identification of the areas of greatest importance for the species across its range. This information is key to design a management plan aimed at ensure the conservation of the species and its habitat.

Materials and Methods

Study Area. The habitat available for bighorn sheep in Sierra de Juarez is located in the area of ravines and canyons of the mountain range, which starts in the border with the United States of America in La Rumorosa and stretches southward to Valle de la Trinidad (31.300° N, -115.370° W and 32.630° N, -116.035° W), covering an area of approximately 145,105 ha. Land tenure is shared by the ejidos Emiliano Zapata, Cordillera Molina, Mission Santa Catarina, Jamau and 16 de Septiembre (RAN 2016; Figure 1).

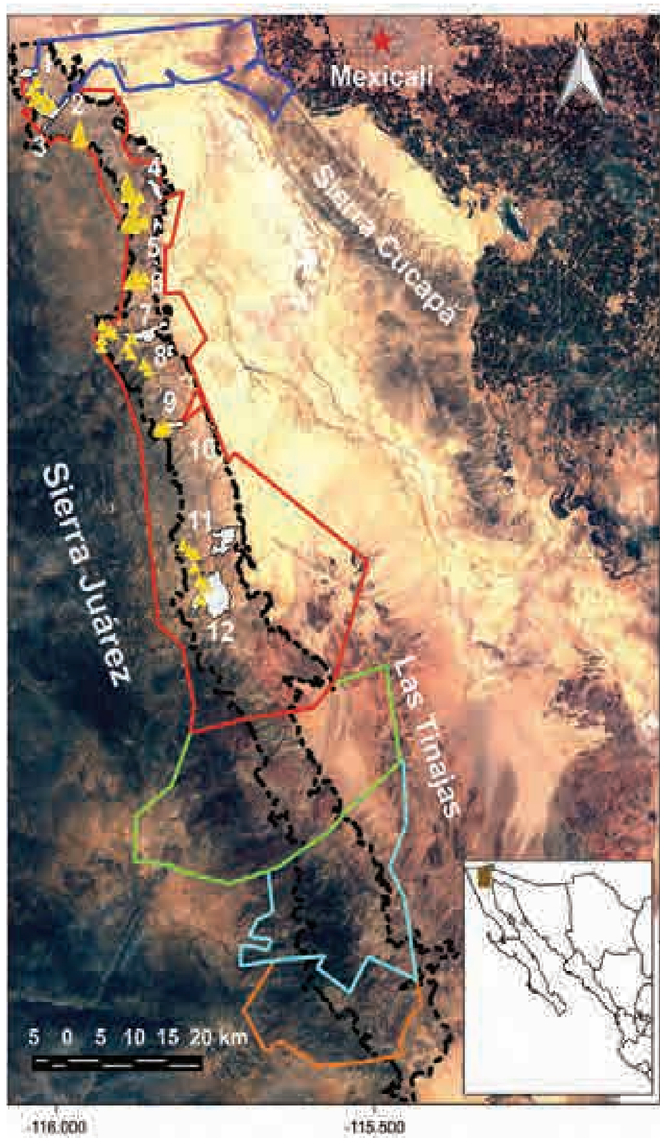

Figure 1. Study area (black dots) showing 2012 aerial census records (yellow) and sampling sites (white): 1) Cañón de Los Álamos; 2) Cañón de los Llanos; 3) Cañón del Ranchito; 4) Cerros Boludos; 5) Cerros de la Ponderosa; 6) La Rosa; 7) Cañón del Tajo; 8) Cerros de los Laureles del desierto; 9) Cañón de Guadalupe; 10) Cerros del Ejido Manatou; 11) Cañón del Alamar; 12) Cañón del Palomar. Ejidos located in the study area are: Emiliano Zapata (dark blue), Cordillera Molina (red), Misión de Santa Catarina (green), Jamau (light blue), 16 de septiembre (orange).

Physiographically, the area is formed by an asymmetric mountain range that is steep-sloped to the east and with gentle slopes to the west. The predominant orientation in the area is to the east, with slopes between 0 % and 72 %, dominated by those lower than 20 %, and most ruggedness values range between 0 and 50 m, with a maximum of 228 m (INEGI 2013).

Six vegetation types cover the study area: pine forest in the highest peaks, chaparral scrubs growing along the western slope, microphyll desert scrub to the east, rosette-shaped desert scrub patches, natural palm-tree patches in the bottom of canyons with water availability all year round, and gallery vegetation along riverbanks (INEGI 2013).

Sierra de Juárez shows five types of climates in ravines: very arid, semi-warm [BWh(x')]; very arid, temperate with rainfall throughout the year [BWk(x')]; very arid, temperate with winter rainfall [BWks]; temperate [Cs]; and sub-humid semi-cold [CB's] (García and CONABIO 1998).

Construction of the database on bighorn sheep evidence. Twelve sites were selected for being recognized by Sierra de Juárez local inhabitants as bighorn sheep distribution areas between La Rumorosa and Cañón del Palomar, which were traveled from January to June 2016 (Figure 1). In each site, one transect was established, the length of which varied according to accessibility, ranging between 2 km and 24 km long. The presence of bighorn sheep was recorded and geo-referenced in each individual transect through indirect evidence (excreta and footprints), discarding those records separated from each other by less than 30 m, to avoid over-fitting.

The database was supplemented with records of the aerial census conducted by the San Diego Zoo in 2012, where four persons flew over Sierra de Juárez for 10 hours on board a Hughes 500™ helicopter with doors removed, from La Rumorosa to Valle de la Trinidad (García 2014; Figure 1).

Environmental Characteristics of the Study Area. We used the free software QGIS 2.8.6 (QGIS Development Team 2016) for geospatial data handling and processing. This software processed the information of a digital terrain model corresponding to the study area, with a spatial resolution of 30 x 30 m pixel (INEGI 2013). We calculated the area covered by particular orientation, slope and ruggedness index data, the latter estimated with the method by Riley *et al.* (1999). The vector information regarding the land-use and vegetation layer of the study area (INEGI 2013) was rasterized to a 30 x 30 m pixel resolution, preserving the vegetation type field.

Plant cover was estimated from Landsat 8 OLI/TIRS satellite images (039 Path, 038 Row) for 21 April 2016, with a 30 x 30 m pixel resolution (USGS 2016), using the calculated value of a Normalized Difference Vegetation Index (NDVI), where NDVI = [(Band 5 - Band 4) / (Band 5 + Band 4)] (Purevdorj *et al* 1998; Pettoreli 2013). With this index, a value between -1 to 1 is assigned to each pixel in the matrix of the study area, where figures below 0.1 correspond to rocky outcrops with sparse vegetation, those between 0.2 and 0.5 to scrubs and grasslands, and those from 0.5 to 1 to forests and cropland (USGS 2015).

The types of climates in the study area were updated using high-resolution climate areas for Mexico (Cuervo *et al.* 2014). These were interpolated to a 30 x 30 m pixel resolution with the thin-plate interpolation technique using the software ANUSPLIN 4.3 (Hutchinson 2006). The parameters of climate surface areas were combined to produced updated types of climates, by entering the respective data into the PATN.EXE 3.1.2.0 program (PATN Development Team 2013), and conducting a cluster analysis with mixed variables using the Gower's similarity coefficient. The resulting climate types were classified according to the nomenclature used by García and CONABIO (1998).

Potential Distribution Modelling. An ecological niche model was constructed using the Maximum Entropy algorithm with the software Maxent version 3.3.3k (Phillips 2013). The database of bighorn sheep evidence records were entered, along with the GIS information layers for orientation, slope, ruggedness index, climate types, vegetation types and NDVI.

The algorithm implementation was used to construct a model of average values, performing 20 replicates with 1,000 iterations each, where 80 % of occurrence records were used for constructing the model and 20 % for the test analysis, by activating the *Random seed* option in order to use different occurrence localities in each replicate for the construction of the models. A regularization factor β of 1 was used, disabling the *Extrapolate* and *Do clamping* options to avoid data overfitting. The selected output was *Logistic*, which classifies pixel values according to a zero-to-one scale, and interprets the scale as the probability of occurrence of the species (Phillips *et al*. 2006).

The predictive accuracy of the model was determined by calculating the area under the curve (AUC) of the receiver operating characteristic (ROC). The fraction of sites misclassified as absences (omission errors) was determined by calculating the omission rate and the average predicted area (Phillips *et al*. 2006). The model obtained was reclassified to produce a binary (presence-absence) map, where a cut-off threshold was applied for presence of training data points (Pearson *et al.* 2007). Model variables were assessed through a Jacknife test, which yielded the information and percentage contributed by each variable. In addition, response curves were plotted to relate bighorn sheep presence data with each environmental variable used to elaborate the maximum-entropy model (Phillips *et al*. 2006).

Results

A total of 196 bighorn sheep records of were obtained, with 116 corresponding to indirect evidence of its presence found during transect walks carried out from January to June 2016, and 80 to direct observations of individuals during the 2012 aerial census (Figure 1). The potential distribution range of the species covers 49,844 ha (34.3 %) of Sierra de Juárez, an area that is concentrated to the northern

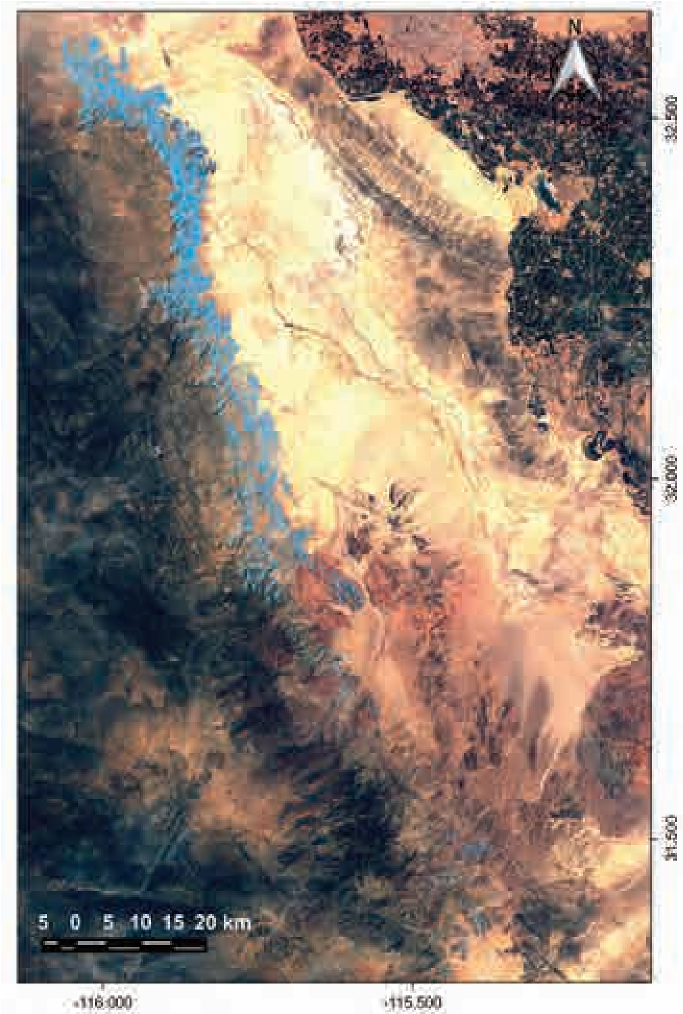

Figure 2. Potential distribution of bighorn sheep in Sierra de Juárez (blue).

region of the cordillera and drops steadily toward the south of the mountain range, being finally scattered in patches in areas near Sierra de Las Tinajas (Figure 2).

The omission rate and predicted area showed that omission in test points did not fully match the predicted omission rate (Figure 3a). For its part, the area under the curve

Table 1. Relative contribution (in percentage) of topographical and environmental variables in the construction of the bighorn sheep potential distribution model in Sierra de Juárez.

Variable	Contribution (%)
Plant cover	26.7
Vegetation type	24.1
Ruggedness	22.2
Climate	11.8
Orientation	10.3
Slope	4.9

(AUC) of the receiver operating characteristic (ROC) yielded a model with a value of 0.877 and a standard deviation of 0.011 (Figure 3b). Taken together, the variables plant cover (26.7 %), vegetation type (24.1 %), and ruggedness (22.2 %) contributed with 73 % of the information necessary for the construct of the model (Table 1).

The response curves generated by the potential distribution model show that the highest probability of bighorn sheep encounter occurs in sites with the following characteristics: very dry semi-warm [BWh(x')], very arid temperate [BWk(x')] and very arid temperate [BWks] climates; gallery and natural palm-tree vegetation; NDVI from 0.05 to 0.07; orientation from 0 to 160°; slope from 0 to 65 %; and ruggedness from 35 to 160 m (Figure 4).

Discussion

The potential distribution range of the bighorn sheep covers regions of northern and central Sierra de Juarez, as according to the niche model these have environmental conditions that provide shelter and resources to the species (Soberón and Peterson 2005). However, the modeling reported here should be regarded as a partial instrument, as the closeness between the localities where the species was recorded led to an overfitting of the data caused by the dependency between data points (Figure 3a). The end result is that the distribution area identified for the bighorn sheep was smaller than the true area. However, the model

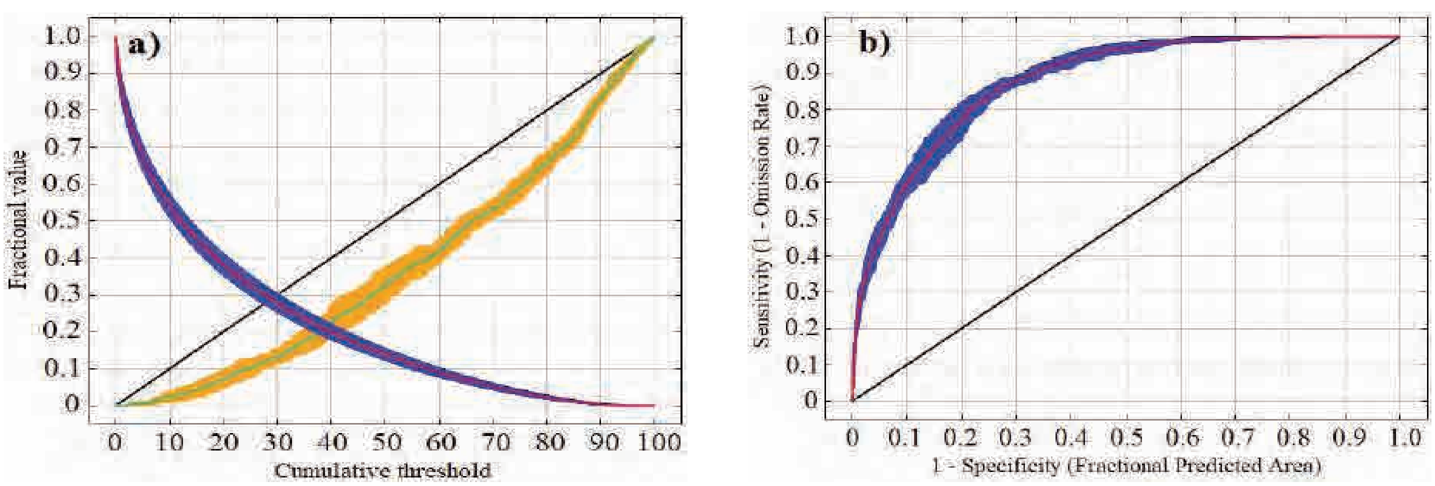

Figure 3. Plots of average omission rate and predicted area (a) and average receiver operating characteristic sensitivity (b), obtained from the modeling carried out for *Ovis canadensis* at Sierra de Juárez. In Figure a, the red line marks the average area; the blue strip, the standard deviation of the average area; the light blue line, the average omission rate; the yellow strip, the standard deviation of the omission rate; and the diagonal black line, the predicted omission rate. In Figure b, the red line marks the average; the blue strip, the standard deviation; and the black diagonal, the expected line if the model is not better than a random one.

functions properly to differentiate between sites with high vs. low probability of bighorn sheep occurrence (Figure 3b; Phillips *et al.* 2006).

In the model, vegetation was the primary component of the habitat that explained most of the bighorn sheep distribution in Sierra de Juarez (Table 1). It provides food resources for the species, is an indicator of water availability, and is closely related to its anti-predatory strategy (Monson 1980; Wilson *et al.* 1980; Holl 1982; Álvarez *et al.* 2009; Escobar *et al.* 2015). The type of vegetation that predominates in regions identified as distribution areas of bighorn sheep is microphyll desert scrub (INEGI 2013). This includes shrubs and herbs of importance as food sources for the bighorn sheep, such as *Ephedra nevadensis*, *Eriodyction angustifolium*, *Senecio spartioides*, *Ceanothus gregii*, *Eriogonum fasciculatum*, *Simmondsia chinensis*, *Ditaxis lanceolata*, *Galium wigginsii*, *Cardiospermum corindum*, *Bromus ciliatus*, *Erioneuron pulchellum*, *Ferocactus* sp. and *Agave* sp. (Reyes, 1976; Sánchez, 1978). In addition, this plant community has a low vegetation index (NDVI = 0.05 to 0.07), providing more visibility or lesser visual obstruction for the detection of predators (Monson 1980; Wilson *et al.* 1980; Holl 1982; Álvarez *et al.* 2009; Escobar *et al.* 2015).

The response of the distribution of organisms to the slope turned out to be indifferent (Figure 4), in contrast with the findings reported by López *et al.* (1999), and Guerrero *et al.* (2003), who concluded that the desert sheep evades sites with slopes below than 60 %. This situation is

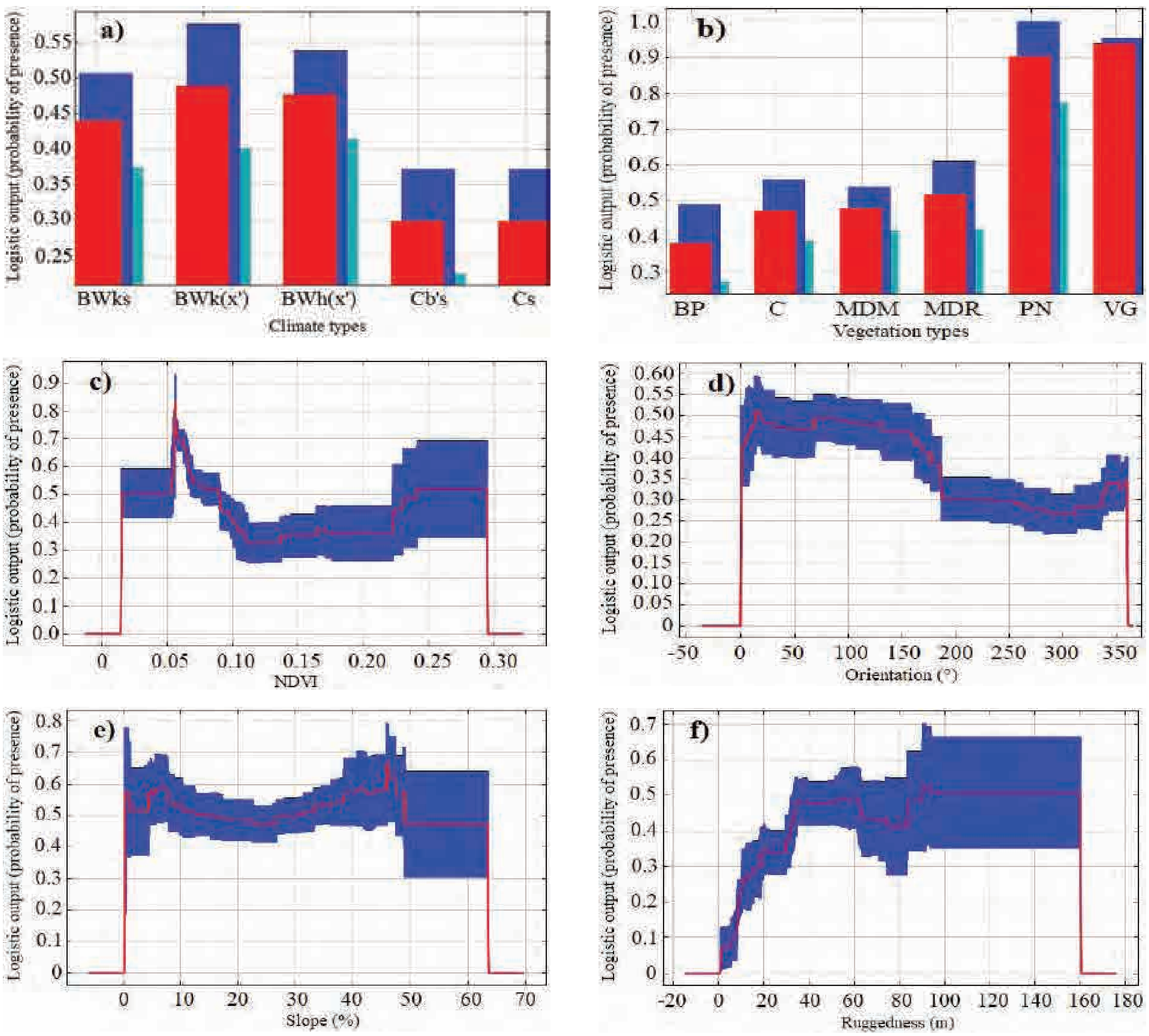

Figure 4. Response curves of bighorn sheep presence data vs each environmental variable used to elaborate the maximum-entropy model. The red line indicates the average, and the blue strip, the standard deviation. The vegetation types are: BP= pine forest; C= chaparral scrub; MDM= microphyll desert scrub; MDR= rosette-shaped desert scrub; PN= natural palm-tree forest; VG= gallery vegetation.

attributed to the fact that the suitability values of topography vary according to the scale used (Divine *et al.* 1996). Differences may stem from the fact in the studies just mentioned, the slope was measured in the field, so that the true value was recorded on site. By contrast, in our study slope was calculated from a digital terrain model with a 30 x 30 m resolution; this approach involves intrinsic inaccuracies related to the original source of the data used to elaborate it, and to the interpolation of elevations that do not represent the true altitude of the terrain for a particular location. In addition, the accuracy of the elevation models is higher in flat versus rugged areas (Riley *et al.* 1999). It has been recorded that slope is a structural element of minor importance in the elaboration of models of the potential distribution of wild ungulates that inhabit mountainous areas; in contrast, vegetation, climate and the anthropic component are the variables with the greatest influence on their distribution (Keya *et al.* 2016; Khan *et al.* 2016).

Ruggedness was one of the variables with the highest contribution to the potential distribution model (Table 1). This variable is related to the proportion of land that facilitates the escape of the species, resulting from the calculation of relief heterogeneity (Riley *et al.* 1999; Escobar *et al.* 2015). Relatively smooth land (35 to 160 m) was associated with the peninsular distribution of the bighorn sheep in Sierra de Juárez (Figure 4). These results are consistent with those recorded in the Santa Isabel and Mechudo mountain ranges by Álvarez (2009) and Escobar *et al.* (2015). In this areas, the bighorn sheep is associated with ruggedness values between 21 m and 267 m, typical of mid- and high-altitude sierras with ravines that offer protection to sheep herds.

The distribution of the bighorn sheep was found to be related to natural palm-tree and gallery patches (Figure 4), both of which are associated with surface water bodies. Likewise, it was found that bighorn sheep individuals thrive along slopes facing east (Figure 4), consistent with the findings reported for Sierra del Mechudo by Guerrero *et al.* (2003), as this is the prevailing slope orientation in both mountain ranges. As regard climate, the very arid semi-warm [BWh(x')], very arid temperate [BWk(x')], and very arid temperate [BWks] types were also associated with the distribution of the species (Figure 4).

The data obtained can be used as a baseline to elaborate an integrated management plan of the bighorn sheep habitat in Sierra de Juárez with the strategic aim to organize the resource owners in a monitoring group. Regular inspections to the bighorn sheep distribution area should be conducted in order to prevent poaching and habitat disruption. In addition, its populations should be monitored, and water bodies used by them should be kept in good condition. Another objective should be to promote the productive diversification of sites with tourism potential, equipping ranches adjacent to bighorn sheep areas to increase their capabilities to conduct nature-tourism activities in sites that are key for the bighorn sheep and that are currently used as rangeland. The above are measures that will contribute to the conservation of this iconic species and its environment.

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Interannual and daily activity patterns of mid-sized mammals in Maracaibo Lake Basin, Venezuela

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Despite its biogeographic importance, the mammals of Maracaibo lake basin have been poorly studied. The objectives of this study were to: 1) provide a list of the mammal species detected by combining information from camera traps and other sources, and 2) describe diurnal and annual activity patterns for some of the species detected. Camera-trapping was carried out for one year in five localities within the Burro Negro Protection Zone (Spanish acronym ZPBN). Records of terrestrial mammals (excluding Chiroptera) were compiled from: 1) direct and indirect opportunistic records during field visits, 2) informal interviews with local inhabitants, 3) three national natural history collections, and 4) scientific literature. The complementarity between sources, similarity with other localities within the region, and temporal changes in composition were evaluated with the Sørensen Similarity Index (RS), and annual differences in the number of detections per sampling effort were evaluated using a X^2 test. Sampling effort was 1,799 camera days, resulting in 569 events of mammal detection recorded and 20 species from 17 families in 9 orders identified. Four species (*Dasypus novemcinctus, Dasyprocta leporina, Cerdocyon thous* and *Leopardus pardalis*) were captured all year round. Camera traps detected half of the non-flying mammal species in the area, including five that had not been previously reported by other sources (*Cuniculus paca, Coendou prehensilis, Sylvilagus floridanus, Procyon cancrivorus* and *Puma yagouaroundi*). At least three species (*Panthera onca*, *Odocoileus virginianus*, and *Hydrochoerus hydrochaeris*) had been reported as historically abundant, but were not detected during the year of camera trapping, which could be due to recent declines in their populations. Camera trapping provides reliable records on the presence of four species with data gaps in their distribution ranges (*Myrmecophaga tetradactyla, Coendou prehensilis, Cerdocyon thous* and *Procyon cancrivorus*), as well as baseline data for evaluating the distribution overlap between pairs of species, such as *Tamandua mexicana* and *T. tetradactyla*, and *Dasyprocta leporina* and *D. punctata*. A higher species richness was observed between June and August, in the rainy season.

A pesar de su importancia biogeográfica, la mastofauna de la cuenca del Maracaibo sólo se conoce parcialmente. Los objetivos del presente trabajo eran: 1) elaborar una lista de especies de mamíferos terrestres combinando el fototrampeo con otras fuentes de información, y 2) describir patrones de actividad diarios y anuales para algunas de las especies detectadas. Se realizó fototrampeo durante un año en cinco localidades dentro de la Zona Protectora de Burro Negro (ZPBN). Se compilaron registros de presencia de mamíferos terrestres (excluyendo quirópteros) de: 1) muestreos no sistemáticos de indicios indirectos y directos, 2) entrevistas informales a habitantes locales, 3) tres colecciones zoológicas nacionales, y 4) literatura científica. Se evaluó la complementariedad entre fuentes, similitud entre localidades o cambios temporales en composición con el índice de Sørensen (RS) y los cambios en el número de detecciones a lo largo del año según el esfuerzo de muestreo mensual con una prueba χ^2. El esfuerzo de muestreo fue de 1,799 días-cámara, se registraron 569 eventos de presencia de mamíferos. Se identificaron 20 especies de 17 familias y 9 órdenes. Cuatro especies (*Dasypus novemcinctus, Dasyprocta leporina, Cerdocyon thous, Procyon cancrivorus* y *Leopardus pardalis*) fueron detectadas constantemente durante el año. El fototrampeo detectó la mitad de las especies no voladoras conocidas en la zona, incluyendo cinco no reportadas por otras fuentes (*Cuniculus paca, Coendou prehensilis, Sylvilagus floridanus* y *Puma yagouaroundi*). Especies como *Panthera onca*, *Odocoileus virginianus* y *Hydrochoerus hydrochaeris* eran históricamente abundantes, pero no pudieron ser detectadas, lo que podría sugerir disminución en las poblaciones. Se aportan registros de presencia para cuatro especies con información vacíos de (*Myrmecophaga tetradactyla, Coendou prehensilis, Cerdocyon thous* y *Procyon cancrivorus*), aún se debe evaluar el solapamiento en la distribución de varias especies, como por ejemplo las dos especies de *Tamandua* y las dos especies de *Dasyprocta*. La mayor riqueza se observó entre junio y agosto que correspondió a la fase inicial del periodo de lluvias.

Key words: Camera trapping; diurnal activity; inventory; monitoring; seasonality.

Introduction

From the biogeographical point of view, the Maracaibo basin is a center of biotic complexity and phylogenetic diversity within the Neotropics, influenced by two centers of maximum convergence of endemism areas: the province of Maracaibo in the Colombian-Venezuelan La Guajira, and the North-Andean province (Ruggiero *et al.* 1998; Noguera-Urbano and Escalante 2015). However, the floristic and faunistic aspects on the eastern coast of this basin have been little studied (Smith and Field 2001; González-Fernández and Nieves 2011; Pietrangeli *et al.* 2011).

One of the few remnants of natural forest vegetation in the Maracaibo lake eastern coast spreads around the Pueblo Viejo dam, also known as Burro Negro, in the southwestern slope of Serrania del Empalado. Despite its location within the Pueblo Viejo Hydraulic National Reserve and the Burro Negro Protected Zone (Bevilacqua *et al.* 2006), this forest area is facing serious pressure from deforestation, forest fires, and indiscriminate wildlife exploitation (Consejo Zuliano de Planificación 1975; Rodríguez *et al.* 2010; Pietrangeli *et al.* 2011; Portillo-Quintero *et al.* 2012; Ferrer-Paris 2016a). On the northern slopes of the Burro Negro Protected Zone (Socopó, Río Socopito), where forest fragmentation is greatest, at least 37 mammal species have been reported, excluding bats. This list is based primarily on field sampling (González-Fernández and Nieves 2011) and literature reviews (Handley 1976; Linares 1998). On the Burro Negro southern slope, where the best preserved forest patch is located, no inventories of fauna have been published to date.

The use of automated photographic capture systems, also referred to as camera trapping, allows a non-invasive recording of the presence of terrestrial non-arboreal vertebrates over extended periods of time and in hard-to-access places, thus improving the power to detect nocturnal or cryptic species, and is also an effective alternative to traditional methods (Barea-Azcon *et al.* 2007; Balme *et al.* 2009; Roberts 2011).

In the present work, we contribute to the knowledge of the fauna in Venezuela by building an inventory of medium- and large-sized terrestrial mammals inhabiting an area of interest for conservation and biogeographical studies that currently shows biological information gaps. We applied the camera-trapping method to monitor for a year the terrestrial fauna of a sector of the Pueblo Viejo Hydraulic National Reserve and the Burro Negro Protected Zone. We developed a list of the species observed by combining camera-trapping with other data sources (zoological collections, bibliography, direct and indirect sightings, and interviews), and used photographic records to describe daily and annual activity patterns for some species.

Methods

Study Area. The study area is located within the Burro Negro Recreation Park and the Pueblo Viejo Hydraulic National Reserve that comprises an extension of 75,000 ha, and overlaps with the Burro Negro Protected Zone between coordinates 10.17445º N, -71.04636° W and 10.22054° N, -71.02438º W, in the municipalities of Lagunillas (Parroquia Campo Lara) and Valmore Rodríguez (Parroquia Raúl Cuenca; Figure 1a). Mean annual temperature is 27.4 °C. Mean annual precipitation ranges from 450 to 1,500 mm, characterized by a marked seasonality, with 60 % to 80 % of annual precipitation from May to October (Plan Bonito Station Series No. 1083, 1956-1983; data from INAMEH; http://www.inameh.gob.ve).

The natural vegetation comprises seasonal deciduous and semi-evergreen forests, with a floristic richness of over 250 species of plants, dominated by species of the families Leguminosae, Bignoniaceae, Sapindaceae, and Malvaceae (Pietrangeli *et al.* 2011). The western bank of the dam is surrounded by large deforested areas, cleared for livestock grazing activities and, to a lesser extent, for agriculture (Pietrangeli *et al.* 2011). This zone is also subjected to periodic burning by farmers as a weed-control measure and illegal wildlife poaching method (pers. obs. Lisandro Morán; Consejo Zuliano de Planificación 1975; Ferrer-Paris 2016a). The main human settlements are located to the west of the dam and consist of scattered hamlets with low population densities (10 to 20 inhabitants/km^2; Fuenmayor 2005; Ferrer-Paris 2016b).

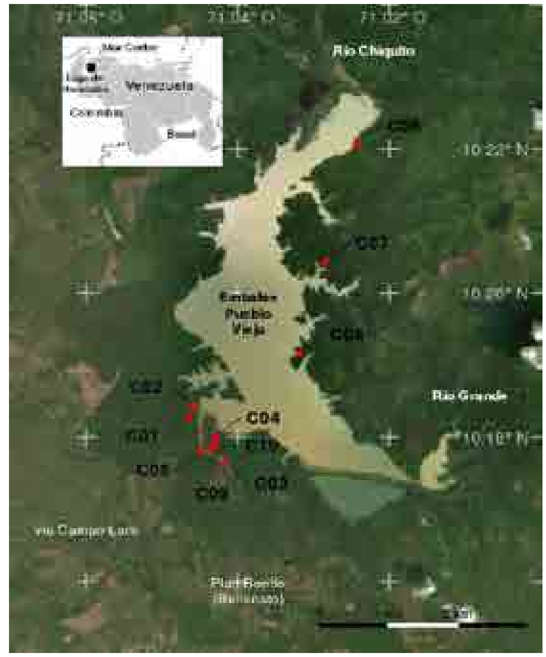

Figure 1. Study area and location of camera traps. Geographic location of the Pueblo Viejo dam in the municipality of Lagunillas, Zulia, Venezuela, and detail of the study area showing sampling localities (C01 to C10). Source of the satellite image: Microsoft Bing Maps Imagery Service API (reviewed on 10 December 2017; Burro Negro, Venezuela, between coordinates 10.17445' 00" N, -71.04636 11" W and 10 24" N, 71° 00' 00" W 10.22054 N, -71.02438 W).

Camera-trapping. Five sampling localities were selected for the monitoring of the Pueblo Viejo dam western bank, which were separated by an average distance of 328 m (61 m minimum; 440 m maximum; Figure 1). These were spread across an area of 83 ha calculated by superimposing operating ranges of 20 ha around each individual camera and taking into account the overlap between these. This first group of five cameras was in operation for a year and, given the proximity between them, are more likely to have captured the same individual on several occasions. We identified three localities on the eastern bank (C06, C07 and C08) separated by an average distance of 1,690 m (1,480 to 1,910 m), which were sampled during the first period only (Table 1). Because of their isolation, these captured relatively independent samples that covered a total area of approximately 60 ha. Localities C01 and C03 were relocated to C09 and C10 for the second sampling period to ensure a better protection of the equipment.

The criteria for selecting sampling localities were: 1) accessibility by water, road or trails throughout the year; 2) indirect evidence of the presence of fauna (*e. g.,* feces, footprints etc.); 3) forest coverage and composition; 4) proximity to water sources; 5) proximity to potential shelter areas for wildlife.

In each sampling locality, one trap camera (Bushnell, Ltl Acorn, Moultrie and Cuddeback) was set on a tree at ~40 cm from the ground, facing the potential route of displacement of animals, and approximately at 3 m from the intended photo capture site. Cameras were in operation during six periods of variable duration (Table 1). At the end of each period, the memory and batteries of cameras were replaced and the information was backed up.

In each sampling station, we recorded the latitude, longitude, elevation, forest cover, understorey density, topography, soil type, and disturbance. Images captured by each camera were downloaded and organized in a relational database, including image metadata, notes and subsequent identifications. The database was used to elaborate tables that summarize the information for each camera according to the effective number of days of operation, and the number of photos and events in each period (Table 1). Photographs were grouped into detection events, each defined as a sequence of consecutive photos separated by less than 1 minute between them. For each event, we identified the species detected using the morphological description and the distribution information outlined in Linares (1998). Percentages of certainty of identification were set based on: 1) image quality, as a percentage established by the identifier according to the degree of sharpness (0 % unfocused – 100 % very sharp), contrast (0 % very dark – 100 % optimal contrast), and frame (0 % partial image of the animal – 100 % complete image); 2) percentage of identification of distinctive features of the species, such as spot patterns, number of rings, pelage color, etc. (Reyes *et al.* 2017). The degree of certainty assigned to a photo is the average of the percent sharpness, contrast, frame, and degree of identification of distinctive features. In addition, we noted descriptions of evidence of reproductive events such as mating, presence of pairs of adults or adults with offspring (Ouboter and Kadosoe 2016).

Table 1. Camera-trap Sampling Effort. The coordinates of each locality, start and end dates of each sampling period, and the number of images/events per camera captured in each sampling period. *Periods with no images due to camera operation issues.

		Period 1	Period 2	Period 3	Period 4	Period 5	Period 6
		05/23/16	07/09/16	08/10/16	09/14/16	11/10/16	03/04/17
Camera	**Coordinates**	07/09/16	08/10/16	09/14/16	11/10/16	03/04/17	06/25/17
C01	10.18282, -71.04636	139/17					
C02	10.18439, -71.04589	455/92	33/9	466/60	216/42	495/74	5319/356
C03	10.17628, -71.04179	*					
C04	10.17999, -71.04295	105/15	*	*	*	386/63	24/6
C05	10.17817, -71.04486	160/35	91/27	299/70	76/19	227/47	4245/589
C06	10.19164, -71.03184	19/4					
C07	10.20433, -71.02850	180/39					
C08	10.22054, -71.02438	71/13					
C09	10.17445, -71.04340		160/30	144/26	907/182	495/93	961/1358
C10	10.17884 -71.04315		*	36/10	69/12	30/6	5068/1358

Additional Sources of Presence Records. Records on the presence of species of non-flying terrestrial mammals in Burro Negro were compiled from the following sources: 1) non-systematic sampling of indirect evidence (footprints and traces) and direct observations; 2) semi-structured interviews with local inhabitants and staff of Instituto Nacional de Parques (National Institute of Parks; INPARQUES); 3) national zoological collections of La Salle Museum of Natural History (MHNLA), Museum of the Rancho Grande Biological Field Station (EBRG), Collection of Vertebrates at University of Los Andes (CVULA), Museum of Biology at University of Zulia (MBLUZ); 4) scientific literature.

Field trips for revision of camera traps included non-systematic samplings when trails adjacent to sampling localities were walked. All samplings were conducted during the day and consisted of recording and identification of prints and traces based on size and shape (Navarro and Muñoz 2000). In direct sightings, specimens were identified in the field by two persons based on the description of Linares (1998). In all cases, the geographical coordinates, date and time of sighting were recorded. During that same period of time, semi-structured interviews were held with 40 persons, including INPARQUES staff and local inhabitants of nearby towns (Plan Bonito and Campo Mara) that use the dam frequently, who were questioned about the mammals observed by them during their regular visits to the area. During interviews, species were identified using illustrations and photographs of the species potentially present in the area (Linares 1998).

Of the four biological collections reviewed, only three (MHNLA, EBRG and MBLUZ) had records of terrestrial mammals sampled in "Burro Negro", "Embalse Pueblo Viejo" or other equivalent locality names. The only literature sources found for the area were the work of Osgood (1912) that refers to El Panorama and El Empalado, located at 50-70 km away from the study area before the construction of the dam, and a review of the fauna of Venezuela by Linares (1998), a work with distribution maps that include records of observations and collections for the study area, albeit of low cartographic accuracy. The list of species compiled for the study area was compared with two preliminary lists of mammals for the hills of the northern slope of Serranía de Ziruma (González-Fernández and Nieves 2011) and for Serrania de Baragua (Sanchez *et al.* 1995), located 90 km to the east-northeast. For all taxonomic lists and comparisons, we used the nomenclature of Wilson and Reeder (2005), except for species of the genus *Mazama* (Gutiérrez *et al.* 2017).

Data Analysis. The frequency of daily and monthly occurrence of each species detected was estimated as the number of photographic events by species divided by the sampling effort conducted at each time of the day or month. A list of adult+offspring or mating events was elaborated. For species with more than 40 detections, we conducted a χ^2 test of observed vs. expected values according to the monthly sampling effort. The complementarity between sources, similarity between localities, or temporary changes in composition, was assessed using the Sørensen index (Legendre and Legendre 1998).

Results

The total effective sampling effort of camera trapping was 1,799 camera days (Table 1), representing 87.7 % of the initial estimated effort (2,051 camera days). This difference was due to operational errors of cameras (*i. e.,* memory, mechanical and configuration issues), and the theft of one camera during the sampling period.

A total of 3,356 photographic events were obtained, divided into 2,506 (74.7 %) false positives, 101 (3.0 %) events involving the detection of other animals (birds, reptiles, amphibians), 132 (3.9 %) partial images that did not allow an unambiguous identification, and 569 (17.0 %) records of mammals suitable for identification to species. We identified 20 species of 17 families and 9 orders (Table 2). Of the total number of mammal recording events, 74.7 % were identified with high certainty (> 80 %), 22.3 % with intermediate certainty (50 % to 80 %), and 2.9 % with low certainty (< 50 %). The species that were most difficult to identify were the two species of *Mazama,* given their similar characteristics, while *Didelphis marsupialis*, *Cebus albifrons*, *Dasyprocta leporina*, *Cerdocyon thous*, and *Leopardus pardalis* had events of both high and low certainty, mainly due to differences in illumination or framing (Figure 2).

In addition to the 20 species detected by camera traps, direct or indirect evidence of the presence of other species (*Panthera onca*, *Hydrochoerus hydrochaeris*, *Pecari tajacu* and *Conepatus semistriatus*) was also observed, for a total of 27 species detected in the field (Table 2). The three collections reviewed and the publication of Osgood (1912) yielded eight and 18 species, respectively, although the localities include a broader and inaccurate region. The work of Linares (1998) contributed the largest number of species (29), but is inaccurate as to the original sources of records; maps involve poor resolution that also restrains an accurate origin. The review of museum specimens and bibliography yielded a total of 35 species recorded. For their part, interviews recorded seven species. From all information sources related to the Burro Negro area, the preliminary list reaches 40 species of non-flying mammals belonging to 24 families and nine orders (Table 2). Sierra de Ziruma has 37 species and shares 33 with Burro Negro ($I_S = 0.857$), while the highlands of Baragua have 32 species and shares 28 ($I_S = 0.778$).

The months with the greatest number of species detected were June and July (13 and 14, respectively), coinciding with the months when the sampling area was extended by installing three cameras on the eastern side of the dam (Figure 3). Even after excluding these additional cameras, a high number of species (11) remained, with a high similarity between these two months ($I_S = 0.833$). The months with the lowest number of species recorded were April and May, with six species; the absence of primates (*Cebus albifrons* and *Allouatta seniculus*), marsupials (*Didelphis marsupialis),* and mapurite (Amazonian hog-nosed skunk, *Conepatus semistriatus;* Figure 3) is worth noting. The composition of species remained more stable between September and February (inter-month $I_S > 0.7$) and was slightly more variable the rest of the year ($I_S < 0.7$, except for June and July).

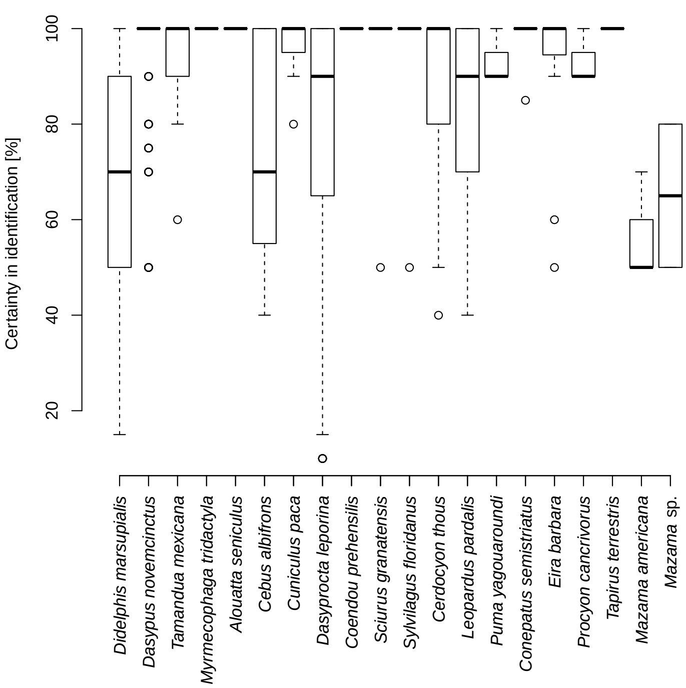

Figure 2. Certainty in the identification of species captured with camera traps. Box Plot indicating the distribution of percent certainty of images for each species. Species are arranged according taxonomically.

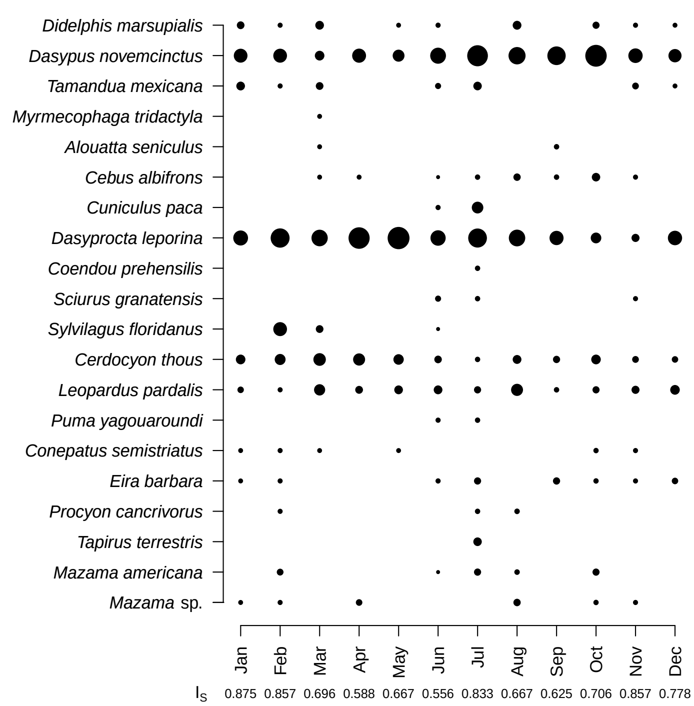

Figure 3. Frequency of monthly activity, expressed as the number of events adjusted for sampling effort (*i. e.*, active camera-trap days) for each species detected throughout a year of sampling. Circle size is proportional to frequency of activity. For each month, the value of the Sørensen index (I_S) is indicated with respect to the previous month.

Table 2. Preliminary list of mammals recorded in Burro Negro (excluding the order Chiroptera). FT: camera-trapping; A: direct sightings during field work; R: traces, footprints, skins, bones; E: Interviews; EBRG: Collection of the Rancho Grande Biological Field Station; MBLUZ: Museum of Biology at University of Zulia; MHNLS: La Salle Museum of Natural History. * *M. gouazoubira* (Fischer, 1814) or *M. nemorivaga* (Cuvier, 1817), see comments in Gutiérrez *et al.* (2017).

Order	Family	Species	Evidence
Didelphimorphia	Didelphidae	*Didelphis marsupialis* (Linnaeus, 1758)	FT, A, Osgood (1912), Linares (1998)
		Marmosa robinsoni (Bangs, 1898)	EBRG, Linares (1998)
Cingulata	Dasypodidae	*Dasypus novemcinctus* (Linnaeus, 1758)	FT, A, R, E, Linares (1998)
Pilosa	Megalochynidae	*Choloepus hoffmanni* (Peters, 1858)	MHNLS
	Myrmecophagidae	*Tamandua mexicana* (Saussure, 1860)	FT, A, R, MHNLS, Linares (1998)
		Tamandua tetradactyla (Linnaeus, 1758)	Osgood (1912)
		Myrmecophaga tridactyla (Linnaeus, 1758)	FT, E, Osgood (1912)
Primates	Aotidae	*Aotus griseimembra* (Eliot, 1912)	E, MHNLS
	Atelidae	*Alouatta seniculus* (Linnaeus, 1766)	FT, A, E, MBLUZ, Osgood (1912), Linares (1998)
		Ateles hybridus (Geoffroy, 1829)	A, E, Linares (1998)
	Cebidae	*Cebus albifrons* (Humboldt, 1812)	FT, E, MHNLS, Osgood (1912), Linares (1998)
Rodentia	Caviidae	*Hydrochoerus hydrochaeris* (Linnaeus, 1766)	E, Osgood (1912), Linares (1998)
	Cuniculidae	*Cuniculus paca* (Linnaeus, 1766)	FT, E
	Cricetidae	*Sigmodon alstoni* (Thomas, 1880)	Osgood (1912), Linares (1998)
		Rhipidomys venezuelae (Thomas, 1896)	Linares (1998)
		Oligoryzomys fulvescens (Saussure, 1860)	Linares (1998)
		Neacomys tenuipes (Thomas, 1900)	Linares (1998)
		Necromys urichi (Allen & Chapman, 1897)	Linares (1998)
		Oecomys speciosus (Allen & Chapman, 1893)	Linares (1998)
		Zygodontomys brevicauda (Allen & Chapman, 1893)	Linares (1998)
	Dasyproctidae	*Dasyprocta leporina* (Linnaeus, 1758)	FT, A, E, Osgood (1912), Linares (1998)
	Echimyidae	Proechimys guairae (Thomas, 1901)	Osgood (1912)
	Erethizontidae	*Coendou prehensilis* (Linnaeus, 1758)	FT, A
	Heteromyidae	*Heteromys anomalus* (Thompson, 1815)	Osgood (1912), Linares (1998)
	Sciuridae	*Sciurus granatensis* (Humboldt, 1811)	FT, A, MBLUZ, MHNLS, Osgood (1912), Linares (1998)
Lagomorpha	Leporidae	*Sylvilagus floridanus* (Allen, 1890)	FT, A, E
Carnivora	Canidae	*Cerdocyon thous* (Linnaeus, 1766)	FT, A, E, Osgood (1912), Linares (1998)
	Felidae	*Leopardus pardalis* (Linnaeus, 1758)	FT, R, E, Osgood (1912), Linares (1998)
		Panthera onca (Linnaeus, 1758)	R, E, Linares (1998)
		Puma yagouaroundi (Goeffroy, 1803)	FT, A, E
		Puma concolor (Linnaeus, 1771)	Linares (1998)
	Mephitidae	*Conepatus semistriatus* (Boddaert, 1785)	FT, Osgood (1912), Linares (1998)
	Mustelidae	*Eira barbara* (Linnaeus, 1758)	FT, A, E, Osgood (1912), Linares (1998)
	Procyonidae	*Procyon cancrivorus* (Cuvier, 1798)	FT
		Potos flavus (Schreber, 1774)	E, Linares (1998)
Perissodactyla	Tapiridae	*Tapirus terrestris* (Linnaeus, 1758)	FT, E, Osgood (1912), Linares (1998)
Artiodactyla	Cervidae	*Mazama americana* (Erxleben, 1777)	FT, Osgood (1912), Linares (1998)
		Mazama sp.*	FT, Linares (1998)
		Odocoileus virginianus (Zimmermann, 1780)	E, Osgood (1912), Linares (1998)
	Tayassuidae	*Pecari tajacu* (Linnaeus, 1758)	E, MHNLS

For the species with sufficient records, which were detected in all months of the year (*Dasyprocta leporina, Dasypus novemcinctus, Leopardus pardalis* and *Cerdocyon thous*), it was possible to analyze the statistical differences throughout the year. For *D. leporina* and *D. novemcinctus*, detections were unevenly distributed throughout the year, even after adjusting for variations in sampling effort. *D. leporina* was observed significantly more frequently in the first half of the year (χ^2 =40.786; $P < 0.001$, *d. f.* = 11). *D. novemcinctus* was significantly more common in the second half of the year (χ^2 =35.560; $P < 0.001$, *d. f.* = 11). In contrast, *L. pardalis* and *C. thous* showed no significant differences (χ^2 =11.773; $P = 0.381$; *d. f.* = 11; χ^2 = 14.777; $P = 0.193$; *d. f.* = 11, respectively). Sixteen events associated with reproductive behavior were recorded for nine species throughout the year of sampling; in the case of *D. leporina* these events did not occur within the period of higher frequency of detection. In the case of *C. thous*, a single mating event

Period	Camera	Date	Species	Activity
P01	C01	06/10/16	*Sciurus granatensis*	Pair of adults
P01	C07	07/03/16	*Tapirus terrestris*	Adult with offspring
P02	C02	07/12/16	*Mazama americana*	Offspring
P02	C09	07/22/16	*Dasyprocta leporina*	Adult with offspring
P03	C02	08/17/16	*Mazama gouazoubira*	Pair of adults
P03	C09	08/20/16	*Leopardus pardalis*	Adult with offspring
P03	C10	08/28/16	*Dasyprocta leporina*	Offspring
P03	C05	09/08/16	*Cerdocyon thous*	Pair of adults
P04	C10	09/15/16	*Dasyprocta leporina*	Offspring
P04	C09	09/16/16	*Eira barbara*	Couple (adult with offspring?)
P04	C09	11/01/16	*Tamandua mexicana*	Adult with offspring
P05	C05	02/06/17	*Mazama americana*	Adult with offspring
P05	C05	02/06/17	*Mazama americana*	Adult with offspring
P05	C02	02/11/17	*Cerdocyon thous*	Copula
P06	C09	04/23/17	*Cerdocyon thous*	Adult with offspring
P06	C09	06/06/17	*Cerdocyon thous*	Two adults with offspring

Table 3. Events associated with reproductive behavior for the species recorded by camera trapping in Burro Negro, in chronological order.

was observed in February, and the presence of offspring was recorded in April and June 2017 (Table 3).

The analysis of the daily activity indicated that seven species were detected almost exclusively during daylight hours (between 06:00 h and 18:30 h), particularly primates, *D. leporina*, squirrel (*Sciurus granatensis*), ferret (*Eira barbara*), gray deer (*Mazama* sp.), and jaguarundi (*Puma yagouaroundi*; Figure 4). Eight were detected exclusively at night (19:00 h to 05:30 h). *L. pardalis* was spotted at night, but also in twilight hours and at dawn. Four species — red brocket deer (*Mazama americana*), northern tamandua (*Tamandua mexicana*), *D. novemcinctus*, and *C. thous* — showed signs of activity in both periods. The first hours of nighttime (between 19 h and 21 h) is the time of the day when more active species were observed (Figure 4).

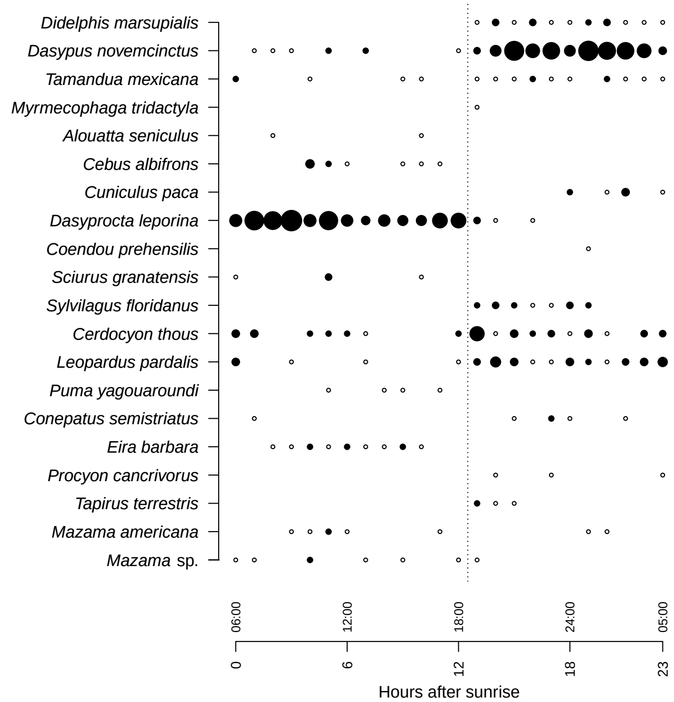

Figure 4. Frequency of daily activity, expressed as the number of events recorded for each species by time of day. Open circles indicate a single event; closed circles indicate more than one event. Circle size is proportional to frequency of activity.

Discussion

Camera trapping is a useful method for the study of terrestrial vertebrates involving low-cost equipment and short field work, thus offering a good cost/effectiveness that produces robust and reliable results on the richness and temporal patterns of species in an area (Balme *et al.* 2009). However, it requires proper planning to ensure high effectiveness. Failures in the operation of cameras and the number of non-informative photographic events (false positives or partial images) consume field and laboratory work time, which should be considered when planning studies at broad geographic scales. In our case, about 13 % of field effort was lost due to equipment failures, and about 83 % of laboratory work time was invested in the processing of non-informative photographic events.

Another aspect that influences the effectiveness of this method is the approach based on visual identification, which may be affected by taxonomic similarities, illumination and framing, camera sensitivity, and expertise of identifiers (Barea-Azcon *et al.* 2007; Reyes *et al.* 2017). Therefore, it is advisable to include protocols that improve the certainty of identifications by persons with different training background (students, volunteers, or experts), or that are unaffected by differences in image quality (Meek *et al.* 2014). During the year of sampling, most (*ca.* 75 %) events were identified with high certainty; most identification issues were due to poor image quality, and only two species involved identification issues by morphological similarity.

The number of species detected was similar to the one derived from an inventory based on camera trapping performed in the Caura river, in southern Venezuela (Perera-Romero *et al.* 2015), and was much larger than the eight species of mammals reported for Parcelación Lomalarga in the southwest of the Department of Valle del Cauca, Colombia (Mosquera-Munoz *et al.* 2015), an area with environmental characteristics that are similar to those in our study area. However, this only represents 50 % of terrestrial mammals according to the sources consulted. Most of the differences between camera trapping and other sources relate to the list of small and medium-sized rodents that are difficult to capture, as these activate the camera less frequently and generally require being physically captured and identified by experts (Chaval *et al.* 2010; De Bondi *et al.* 2010; García *et al.* 2012; Herbreteau *et al.* 2011).

The jaguar (*Panthera onca*) is an iconic and highly valued species in conservation programs in the Neotropics (García *et al.* 2012). ZPBN is an important refuge area within the Maracaibo lake basin, given the high probability of occurrence and the low probability of extinction estimated (Jędrzejewski *et al.* 2017). However, jaguars were not recorded in this study. The available records are indirect (skins and interviews), and are probably two years old. The absence or low abundance of this species can only be determined with a more intensive and prolonged sampling effort (Hernández-Pérez *et al.* 2015).

Osgood (1912) mentions that the white tailed deer (*Odocoileus virginianus*) and the capybara (*Hydrochoerus hydrochaeris*) were common in savannas and wetlands of this region in the early twentieth century. Most likely, their populations have been seriously reduced due to overexploitation by hunting, thus being difficult to spot today (Correa-Viana 1991; Vaughan and Rodriguez 1997; Ojasti 2011). The rescue and protection of a remnant population of *H. hydrochaeris* could set the basis for a breeding program in a sector within the reservoir area, following the rational management model that has been implemented in the Western Orinoco Plains (Ojasti 2011). The photographic record of the tapir (*Tapirus terrestris*) confirms that this species still thrives in the study area, in spite of the negative trends reported for other zones of its range (Naveda-Rodríguez *et al.* 2012).

The phylogenetic analysis of deer has revealed a huge information gap restraining a precise definition of the distribution of *M. gouazoubira* and *M. nemorivaga.* so that the photographic record of gray deer (Table 2) cannot be identified accurately without a physical sample or additional evidence (Gutiérrez *et al.* 2017). *Tamandua mexicana* and *T. tetradactyla* have disjunct distributions, the former being present in Sierra de Ziruma (part of Serranía del Empalado; González-Fernández and Nieves 2011) and *T. tetradactyla* in Serrania de Baragua (Sanchez *et al.* 1995). The evidence of the presence of both species in the study area suggests an overlap in their distribution range. The population of *Dasyprocta leporina* inhabiting the Maracaibo lake eastern bank and the western portion of the States of Falcón and Lara appears to be isolated from the general distribution in the rest of the country, while *D. punctata* appears in the lake basin (Ojasti 1972). The records of *D. leporina* in Serrania de Baragua and Ojo de Agua El Cardon, to the northwest of our study area, are ambiguous, as pointed out by the authors themselves (Sanchez *et al.* 1995; Ferrer-Paris *et al.* 2015). Some of the photographic records assigned here as *D. leporina* with poor certainty could in reality correspond to *D. punctata.* In addition, it is the southernmost limit of the distribution of a disjunct population of *Myrmecophaga tridactyla* in the Maracaibo lake eastern banks (Linares 1998). Separately, no records are available for *Cohendu prehensilis* within 200 km of the study area (Linares 1998; GBIF Secretariat 2017).

One of the most valuable contributions of camera trapping is the continued and detailed recording — which cannot be achieved using other methods of active search or conventional traps — including the exact date and time of each capture event, hence providing useful data about the behavior of species (Ouboter Kadosoe and 2016). During 12 months of sampling, fluctuations in both the number of species detected and the detections by species were recorded (Figures 3 and 4), likely due to changes in activity or behavior patterns. The highest species richness was observed at the beginning of the sampling period, between June and August 2016, which corresponded to the early rainy season after a dry period with a high incidence of fires (Ferrer-Paris 2016a). The composition of species remained stable between September 2016 and February 2017 despite the changes in precipitation at the end of the rainy season and the beginning of the drought. The number of species detected declined between April and May 2017, at the beginning of another rainy season. This pattern is probably due to a sampling artifact. As cameras were located near water bodies, the detectability of animals was likely higher during the dry season, as these visited waterholes more frequently. In contrast, animals were probably more scattered during the rainy season as they were able to encounter water sources elsewhere, thus decreasing the probability of detection of cameras.

The sampling effort allowed visualizing a whole annual cycle and gather evidence of reproductive events for at least eight species (Table 3). The species that were detected throughout the year include *D. leporina,* which showed an increase in the number of detections during the driest semester of the year, a pattern that contrasts with the one observed for species of the same genus in other localities (Rocha *et al.* 2006; Jax *et al.* 2015*;* Ouboter Kadosoe and 2016). One possible explanation is that this species extends its period of activity in search of food, thus offsetting the temporary shortages of resources (Smythen 1978). It may also reflect a larger population size after the breeding season between July and September. *Dasypus novemcinctus* was detected more frequently in the wettest semester of the year, in line with reports for a different area (Rocha *et al.* 2006). It is likely that carnivores such as *Cerdocyon thous* and *Leopardus pardalis* show minor seasonal fluctuations, given their ability to adjust their diet to the resources available in different seasons of the year (Sunquist 1992).

Daily activity patterns often reflect the effect of the energy balance (food demand and supply) and the relationship between predators and prey (Schaik and Griffiths 1996), and can be altered by hunting pressure and modifications of the habitat that affect resource availability. The activity pattern of *Leopardus pardalis* showed a most intense activity at night, plus records in twilight hours, and isolated events during the day; this is consistent with the activity pattern reported by several authors (Ayala *et al.* 2010; Hernández-Pérez *et al.* 2015; Mosquera-Munoz *et al.* 2015; De la Torre *et al.* 2016). This pattern is probably influenced by the nocturnal activity of its potential preys, such as *Didelphis marsupialis, Dasypus novenmcinctus, Tamandua mexicana* and *Sylvilagus floridanus* (Martinez 2013). The few records of *Puma yagouaroundi* show a diurnal activity pattern that matches the activity of non-flying birds, which are reported as its most common prey (Mosquera-Munoz *et al.* 2015; Briones-Salas *et al.* 2016).

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Predictive species distribution model of two endemic kangaroo rats from Mexico: *Dipodomys ornatus* and *D. phillipsii* (Rodentia: Heteromyidae)

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Predictive species distribution models (SDMs) allow identifying suitable areas for the conservation of populations of endangered species. *Dipodomys ornatus* and *D. phillipsii* are two Nearctic rodents endemic to Mexico that inhabit arid and semiarid environments. Endemic species have a restricted distribution, making it difficult to monitor their distribution and conservation status. Therefore, it is important to understand the interactions between these species and the environmental/ecological variables in their local habitat. In this study, we constructed SDMs for these two rodents using MaxEnt (Maximum Entropy model). Also, we defined the areas with the suitable ecological characteristics for the preservation of the species. In order to construct an SDM for each species, we used MaxEnt at a national scale (Mexico), with a resolution of 30 arc-sec (1 km^2). To this end, 91 presence data (63 for *D. ornatus* and 28 for *D. phillipsii*) reported in the literature and data bases were used, along with 27 climatic and ecological variables. Maps with a high predictive capability (Area under a Receiver Operating Characteristic [ROC] Curve = AUC > 0.9) were obtained for both species. Principal Component Analyses were carried out, resulting in 11 PCs that accounted for 95% of variability of the original environmental variables. The *D. ornatus* SDM is defined mainly by PC2, PC11 and PC1. PC4, PC2 and PC6 are the most influential variables in predicting the SDM of *D. phillipsii*. Sites with suitable environmental conditions for *D. ornatus* are located in the States of Durango, Zacatecas, Aguascalientes, San Luis Potosí, and Guanajuato, whereas the best environmental conditions for *D. phillipsii* were found in Puebla, Tlaxcala, Veracruz, and Hidalgo. As these species have a restricted distribution and field sampling is difficult, determining their conservation status is a complex issue; however, SDMs are a useful tool to identify areas with ideal characteristics to conduct surveys aimed at determining their conservation status or that could be used as future natural protected areas.

Los modelos predictivos de distribución de especies (MDE), permiten identificar áreas con condiciones adecuadas para la conservación de especies en peligro de extinción. *Dipodomys ornatus* y *D. phillipsii*, son dos roedores neárticos, endémicos de México, que habitan principalmente en regiones áridas y semiáridas. Las especies endémicas poseen una distribución restringida, lo que dificulta monitorear su distribución y estado de conservación. Por lo tanto, es importante comprender las interacciones que existen entre las especies y las variables ambientales/ecológicas de los sitios donde estas habitan. En este estudio, se realizó un MDE de estos dos roedores utilizando MaxEnt (Modelo de máxima entropía). También se definieron áreas con las características ecológicas necesarias para la sobrevivencia de las especies que pueden ser utilizadas en programas futuros de conservación. Para construir el MDE, se utilizó el modelo MaxEnt a una escala nacional (México), con una resolución de 30 arcos – segundos (1 km^2). Para este estudio, se utilizaron 91 datos de presencia (63 para *D. ornatus* y 28 para *D. phillipsii*) previamente reportados en la literatura y bases de datos, que fueron usados con 27 variables climáticas y ecológicas. Se obtuvieron mapas con un valor alto de capacidad de predicción (Área bajo una curva Característica Operativa del Receptor [ROC] = AUC > 0.9) para ambas especies. Se realizó un Análisis de componentes principales (ACP), y 11 CPs contienen el 95% de la variabilidad de las variables ambientales originales. El MDE para *D. ornatus* se define principalmente por los CP2, CP11 y CP1. CP4, CP2 y CP6 fueron las variables que más influyeron en la predicción del MDE de *D. phillipsii*. Los lugares con las condiciones ambientales favorables para *D. ornatus* se encuentran en los estados de Durango, Zacatecas, Aguascalientes, San Luis Potosí y Guanajuato; mientras que para *D. phillipsii* están en Puebla, Tlaxcala, Veracruz e Hidalgo. Para especies con distribución restringida, en las que la recolecta en campo es difícil, y que por ello es complicado determinar su estatus de conservación, los MDE, son una herramienta útil para identificar áreas con las propiedades o características ideales, donde se les debe buscar para determinar un estatus de conservación, o que incluso pueden ser utilizados para futuras áreas naturales a conservar.

Key words: conservation; desert; endemic; species distribution model; habitat; rodents.

Introduction

Kangaroo rats *Dipodomys ornatus* and *D. phillipsii* are two Nearctic rodents endemic to Mexico, adapted to semi-arid and arid environments. Both taxa are currently distributed across a narrow strip of arid and semi-arid habitats in central-southern Mexico, stretching from southeast Durango (Mexican Plateau, MP) toward southern Puebla and northern Oaxaca (Trans-Mexican Volcanic Belt, TMVB; McMahon 1979; Schmidly *et al.* 1993). The current taxonomy recognizes *D. ornatus* at species level (previously *D. p. ornatus*), and *D. phillipsii* comprising three subspecies: *D. p. oaxacae*, *D. p. perotensis* and *D. p. phillipsii* (Fernández *et al.* 2012; Ramírez-Pulido *et al.* 2014).

Although both species are morphologically similar (Genoways and Jones 1971; Jones and Genoways 1975; Hall 1981; Genoways and Brown 1993), these differ in size, pelage color, and cranial shape (Merriam 1894); also, there are genetic differences and a marked geographical variation between their populations. *D. ornatus* and *D. phillipsii* are mid-sized kangaroo rats (total length 245 to 280 mm). Comparing both taxa, northern populations (*D. ornatus*) are medium to large in size, with pale fur and broad cranium. In contrast, *D. p. phillipsii* inhabits the Valley of Mexico and adjacent areas, being mid-sized, darker in color, and with a broad interorbital region. The populations of *D. perotensis* are distributed in Tlaxcala, Puebla, and Veracruz, characterized by larger individuals with fur color intermediate between *D. ornatus* and *D. phillipsii*; for its part, *D. p. oaxacae*, which has been reported in southern Puebla and northern Oaxaca, is the smallest of all subspecies (Jones and Genoways 1975).

These rodents are nocturnal, feeding mainly on seeds, leaves, and small plants. They get metabolic water from the food they eat, living in mound-shaped, gently-sloping burrows with various entrances built in open areas. Both species prefer semi-arid and arid zones with sandy soils and xerophytic vegetation with large grasses (Davis 1944; Hall and Dalquest 1963; Genoways and Jones 1971; Jones and Genoways 1975). Their altitudinal distribution ranges from 950 m a.s.l. in Oaxaca to 2,850 m a.s.l. in Veracruz (Jones and Genoways 1975).

The Mexican authorities in conservation issues (Mexican Official Standard 059) consider *D. phillipsii* and *D. ornatus* as a single species (*D. phillipsii*), listed under the endangered category, and hence subject to protection (SEMARNAT 2010). The first stage in management and conservation plans for threatened or endangered species is to determine their distribution and ecological niche. Grinnell (1917) defined ecological niche as the habitat characterized by environmental conditions that are suitable for population to survive and reproduce. These environmental conditions determine the distribution of a given species.

Predictive species distribution models (SDMs) facilitate the identification of suitable habitats for the conservation of populations, aimed at preventing extinctions. SDMs are based on the correlation between geographic records of species and the respective environmental variables; these kind of data have been used for modeling the potential distribution of species (Elith *et al.* 2006; Peterson 2006; Kumar and Stohlgren 2009) like a threatened and endangered tree species in New Caledonia, using small number of occurrence records (11).

To date, few studies focus on predicting potential distribution areas for species endemic to Mexico with a distribution restricted to MP and TMVB (Jones and Genoways 1975). Therefore, the objective of this study was to model the ecological niche of two endemic rodents, *D. ornatus* and *D. phillipsii*, using MaxEnt to infer potential areas for conservation.

Materials and Methods

Study Area. Presence data were used for *Dipodomys phillipsii* and *D. ornatus* in 11 States of Mexico: Aguascalientes, Durango, State of Mexico, Jalisco, Oaxaca, Puebla, Queretaro, San Luis Potosí, Tlaxcala, Veracruz, and Zacatecas (Figure 1), all of which include arid and semi-arid zones (Fernández *et al.* 2012; Ramírez-Albores *et al.* 2014). According to presence data for both species, these are distributed in xeric vegetation covering semi-arid and arid zones (Rzedowski 2006); this vegetation includes rosetophilous desert scrub, succulent scrub vegetation, natural grasslands and

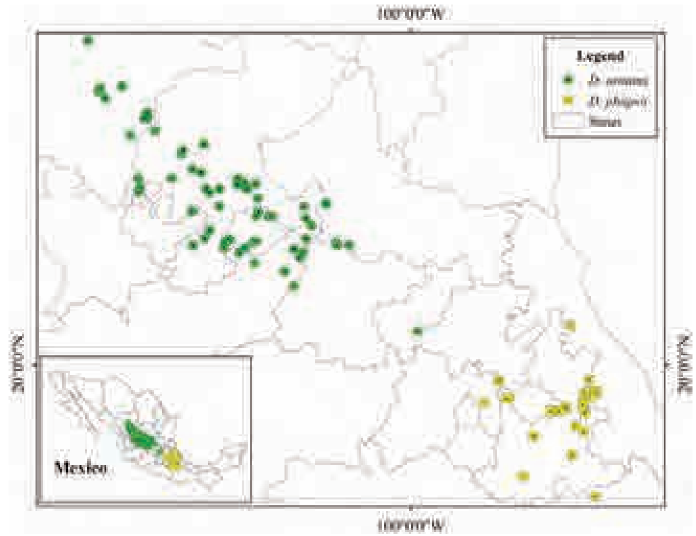

Figure 1. Location map of collection records of kangaroo rats *Dipodomys ornatus* and *D. phillipsii*.

low deciduous forests, in addition to annual rainfed agricultural crops (INEGI 2016).

The region stretching from Durango to northern Oaxaca comprises various topographic conditions and soil types; those related to kangaroo rat presence data are Kastanozem, Leptosol, Chernosem, Durisol, Phaeozem, Cambisol, Andosol, Arenosol, Regosol, and Vertisol (INEGI 2016). Climate ranges from arid and dry (central-northern Mexico) to temperate and humid (center-south). Mean annual temperature ranges between 12 ºC and 26 °C. The northern part of the country is a desert area, where climate is usually more extreme and with little annual precipitation, which increases southwards (INEGI 2016).

Data sources. A total of 91 presence data were gathered, 63 for *D. ornatus* and 28 for *D. phillipsii*, as reported by Fernández *et al.* (2012), Ramírez-Albores *et al.* (2014) and obtained from the VertNet database (http://www.vertnet.org/index.html, Esselstyn 2015; Garner 2015; Gegick 2015; Orrell 2015; Prestridge 2015; Revelez 2015; Abraczinskas 2016; Conroy 2016; Cook 2016; Flannery 2016; Opitz 2016; Braun 2017; Gall 2017; Grant 2017; Slade 2017; Feeney 2018, and Millen 2018, Figure 1). Table 1 lists the 27 environmental variables used in the analysis, with a resolution of 30 arc-seconds (pixel size approx. 1 km^2), including 19 climatic variables obtained from the WorldClim database (www.worldclim.org; Hijmans *et al.* 2005), three variables on soil properties, three topographical variables, one climate variable, and one variable regarding land use and vegetation; all were obtained from the National Institute of Statistics and Geography of Mexico (INEGI 2015). The latter were selected a *priori* taking into consideration previous work describing the

Table 1. Predictive environmental variables used in the distribution model of kangaroo rats *Dipodomys ornatus* and *D. phillipsii*.

Variables
1) WorldClim Climate Variables
BIO1 = Mean annual temperature
BIO2 = Mean diurnal range (maximum temperature - minimum temperature; monthly average)
BIO3 = Isothermality (BIO1/BIO7) * 100
BIO4 = Temperature seasonality (coefficient of variation)
BIO5 = Maximum temperature of the warmest period
BIO6 = Minimum temperature of the coldest period
BIO7 = Annual temperature range (BIO5-BIO6)
BIO8 = Mean temperature of the wettest quarter
BIO9 = Mean temperature of the driest quarter
BIO10 = Mean temperature of the warmest quarter
BIO11 = Mean temperature of the coldest quarter
BIO12 = Annual precipitation
BIO13 = Mean precipitation of the wettest period
BIO14 = Mean precipitation of the driest period
BIO15 = Precipitation seasonality (coefficient of variation)
BIO16 = Mean precipitation of the wettest quarter
BIO17 = Mean precipitation of the driest quarter
BIO18 = Mean precipitation of the warmest quarter
BIO19 = Mean precipitation of the coldest quarter
2) Soil Properties
HUM = Soil moisture
SUELOTY = Soil type
TEXT = Soil texture
3) Topographic Variables
SLOPE = slope
TOPO = Topoforms
CEM = Mexican elevation continuum
4) Type of Climate
CLIM = Climate Units
5) Land Use and Vegetation
USES = Land use and vegetation

ecology of the species studied (Merriam 1894; Genoways and Jones 1971; Genoways and Brown 1993).

Modeling. First, a randomness test was run with the georeferenced records of each species (*D. ornatus* and *D. phillipsii*) using the R statistical package (R Development Core Team 2018). The randomness test determines whether records are spatially distributed at random or clustered (Bivand *et al.* 2008). If data are not clustered, 75 % of records are used as training data, and the remaining 25 %, as testing data. For clustered data, a pattern analysis is conducted to estimate the probability of finding one record at a certain distance, expecting no spatial autocorrelation between data (Hengl 2007). The pattern analysis is run with the ILWIS 3.3 software (https://52north.org/software/software-projects/ilwis/). If the pattern analysis confirms clustered data, the software ArcGis v9.2 (ESRI 2006) randomly selects 50 % of the total number of records, which are used for model training, and the rest are used for model validation.

From the 27 climate variables in raster data files, a Principal Component Analysis (PCA) was run to avoid multicollinearity — a statistical issue defined as a high degree of correlation between variables. When variables are highly correlated, small changes in data or variables may lead to considerable changes in the distribution of species; consequently, estimates from the resulting models are unreliable (Quinn and Keough 2002). Using the R statistical package, these data were described as a group of new variables (components) that were not correlated with each other. Eleven principal components (PC, raster data files) were used as new environmental variables, which accounted for over 95% of the variance of the original variables.

SDMs of the rodents studied were constructed using the MaxEnt software (https://biodiversityinformatics.amnh.org/open_source/maxent/), which requires geographic presence records (which is an advantage for the modeling of endemic species with small populations and scarce records; Phillips *et al.* 2006), and a set of climatic and/or ecological variables that are part of the information about the known distribution of species. This software works with a limited number of samples and climatic variables (topography, climate, soil type, Papeş and Gaubert 2007; Pearson 2007; Hernandez *et al.* 2008; Phillips and Dudik 2008; Wisz *et al.* 2008), as well as categorical and continuous variables, requiring knowledge about the biology of the species for the correct interpretation of results (Elith *et al.* 2006). In addition, it provides a continuous result and analyses are repeatable (Phillips *et al.* 2006; Phillips and Dudik 2008).

The parameters used for modeling were those displayed by default by MaxEnt version 3.3.3k, except for the "Extrapolate" and "Do clamping" parameters, which were disabled; the data output was logistic. The models obtained were validated using a cut-off threshold value equal to the maximum test sensitivity plus specificity (Liu *et al.* 2005), which maximizes the cases where the model erroneously assigns an unsuitable habitat (true negative) and ignores the suitable habitat (false positive); this approach is very common when using MaxEnt (Ferraz *et al.* 2012; Jorge *et al.* 2013; Kebede *et al.* 2014). In addition, we conducted a preliminary validation by calculating the area under the curve (AUC = Area under a Receiver Operating Characteristic [ROC] Curve). The AUC ranges from 0 to 1; an AUC of 0.5 indicates that this model is not better than one constructed at random, while a value of 1 indicates a perfect fit of the model (Pearce and Ferrier 2000; Newbold *et al.* 2009). Subsequently, a binomial test was performed, which evaluates whether the model obtained is better than one derived at random ($p > 0.5$) based on omission rates (i.e., proportion of test records that fall outside of the predicted area, considering cut-off threshold). Successful records are those with logistical values above the selected cut-off threshold (Elith *et al.* 2011; Cruz-Cárdenas *et al.* 2014). Finally, the models obtained with the MaxEnt algorithm were reclassified into Boolean layers (presence/absence), using the cut-off threshold. MaxEnt produces logistic data, i.e., continuous values ranging from 0 to 1 that represent the probability of occurrence of the species in the area, and which are represented on a map (Bean *et al.* 2014). Data were reclassified using the software ArcGis v9.2 (ESRI 2006; Pliscoff and Fuentes-Castillo 2011). Some previous studies of threatened species (Aguilar-Soto *et al.* 2015; Rovzar *et al.* 2013) established ranges of probability of occurrence based on the distribution of species. The 0.7 to 1 range indicates high probability of occurrence, where most geographic records are located; 0.5 to 0.7 indicates medium probability, including the remaining records; and finally threshold to 0.5 represents low probability of occurrence. These probability ranges have been used in ecological studies for various species (Bailey *et al.* 2002; Woolf *et al.* 2002; Liu *et al.* 2005; Gil and Lobo 2012; Martínez-Calderas *et al.* 2015). To calculate the area (km^2) predicted by models, the number of pixels belonging to each classification was counted and multiplied by 30 arches-seconds (pixel resolution) for each probability of occurrence (low, medium, and high).

Results

The randomness analyses of geographic records of both species reveals a clustered pattern; Figure 2 shows that values (solid line, G^obs[r]) lie outside the confidence interval (gray area G^hi[r] and G^o[r]) estimated by distance (r). If the solid line (G^obs[r]) lies within the upper and lower lines (G^hi[r] and G^o[r]), the data are statistically distributed at random, with no cluster pattern. If G^obs[r] is located outside of these confidence lines, it indicates a spatial pattern, *i.e.*, data are clustered. The records for *D. ornatus* (Figure 2A) show a clustering pattern, where the probability of finding another record is G(r) = 0.9 at a distance of 33 km (r = 0.3 degrees). On the other hand, the records for *D. phillipsii* (Figure 2B) show a lower clustering pattern, with a probability of finding another record G(r) = 0.8, although these involve a distance of 22 km (r = 0.2 degrees).

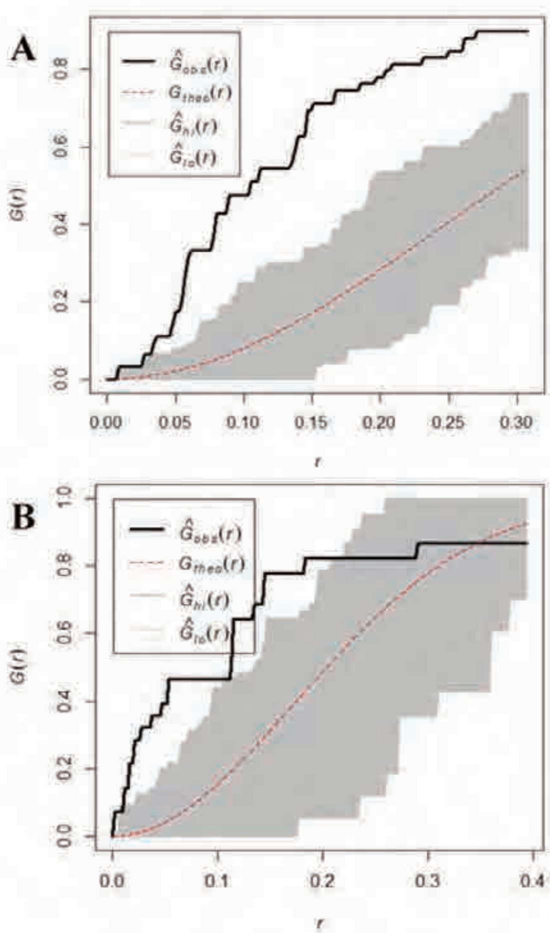

Figure 2. Randomness analysis of geographic records. A) *Dipodomys ornatus,* and B) *Dipodomys phillipsii.* The dotted line corresponds to calculated theoretical values, the gray area corresponds to the confidence interval, and the solid line corresponds to observed values in our data.

The pattern analysis of geographical records of both species with the software ILWIS 3.3 resulted in the selection of 8 of 63 records for *D. ornatus* for model training, and 4 of 28 records for *D. phillipsii*. To validate the model, 12 test records

Table 2. Contribution of environmental variables used in the Principal Component Analysis (Loading factors) for kangaroo rats *Dipodomys ornatus* and *D. phillipsii*.

Original variables	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9	PC10	PC11
bio1	-0.219	0.290	-0.098	0.008	-0.053	0.047	-0.021	-0.007	0.030	0.017	-0.046
bio2	**0.242**	-0.032	-0.110	0.175	0.063	-0.242	**0.131**	-0.016	0.134	-0.535	-0.154
bio3	-0.156	-0.196	-0.344	-0.182	-0.071	-0.070	-0.011	0.002	0.157	-0.385	-0.044
bio4	**0.222**	0.187	0.271	0.214	0.067	-0.029	0.064	-0.018	-0.083	0.094	-0.021
bio5	-0.024	**0.393**	0.005	0.190	0.005	-0.067	0.040	-0.029	0.026	-0.192	-0.139
bio6	-0.281	0.139	-0.131	-0.127	-0.071	**0.097**	-0.087	-0.010	0.024	0.025	0.000
bio7	**0.268**	0.104	0.135	0.246	0.074	-0.140	0.112	-0.008	-0.008	-0.144	-0.086
bio8	-0.107	**0.334**	0.011	0.209	-0.020	-0.010	0.099	0.035	0.023	0.183	-0.025
bio9	-0.213	0.231	-0.157	0.018	-0.003	0.020	-0.059	-0.050	0.057	-0.339	-0.101
bio10	-0.089	**0.396**	0.042	0.124	-0.011	0.041	0.005	-0.021	-0.017	0.029	-0.078
bio11	-0.269	0.146	-0.205	-0.091	-0.067	0.055	-0.051	-0.003	0.060	-0.058	-0.035
bio12	-0.281	-0.133	0.090	0.156	-0.008	-0.036	0.066	0.012	0.008	-0.065	0.065
bio13	-0.264	-0.142	-0.013	0.275	0.028	-0.001	0.114	0.026	-0.021	-0.005	0.097
bio14	-0.228	-0.059	**0.369**	-0.031	-0.013	-0.155	-0.034	-0.035	0.025	-0.102	0.041
bio15	0.046	-0.054	-0.409	**0.429**	**0.135**	0.018	**0.241**	0.111	0.006	0.037	0.060
bio16	-0.262	-0.153	-0.029	0.262	0.017	-0.002	0.121	0.030	-0.007	-0.021	**0.085**
bio17	-0.227	-0.064	**0.380**	-0.022	-0.010	-0.146	-0.039	-0.054	0.016	-0.104	0.068
bio18	-0.211	-0.145	0.016	**0.344**	**0.095**	-0.028	**0.164**	-0.007	-0.076	0.133	**0.217**
bio19	-0.206	-0.061	**0.357**	0.064	0.041	-0.080	-0.067	-0.144	0.002	-0.326	0.071
CEM	0.156	-0.339	-0.048	0.013	-0.002	-0.110	0.059	0.044	0.052	-0.020	0.029
CLIM	-0.242	-0.139	-0.106	0.020	-0.041	-0.024	0.054	**0.140**	0.043	**0.212**	-0.080
HUM	-0.113	-0.198	0.202	0.058	0.071	**0.178**	0.102	**0.243**	**0.270**	0.120	-0.812
SLOPE	-0.012	-0.188	-0.095	0.264	-0.074	**0.244**	-0.409	-0.300	-0.653	-0.104	-0.309
SUELOTY	-0.044	-0.001	-0.099	0.071	**0.756**	-0.142	-0.515	-0.149	**0.279**	0.139	0.012
TEXT	0.084	-0.086	-0.039	0.245	-0.453	-0.017	-0.118	-0.632	**0.511**	**0.199**	-0.030
TOPO	0.070	0.004	0.010	**0.281**	-0.380	-0.213	-0.590	**0.590**	0.109	-0.007	**0.093**
USES	-0.111	0.024	-0.153	-0.126	-0.072	-0.819	0.073	-0.124	-0.280	**0.253**	-0.267

PC = PCA principal component. The loading factors of original variables are highlighted in black.

were selected at random for *D. ornatus,* and 6 for *D. phillipsii.* When the potential distribution model was constructed for both species, these did not pass the binomial test validation; hence, the models obtained are not better than one obtained at random ($P > 0.5$). Consequently, all records were splitted in 50 % for model training plus 50 % for model validation for both species. The PCA of the 27 climatic variables produced 11 PCs that altogether account for 95 % of the variability of the original variables. The environmental

Table 3. Percent contribution of the main components resulting from MaxEnt models for kangaroo rats *Dipodomys ornatus* and *D. phillipsii*. The most important principal components for each species are highlighted in bold.

	D. ornatus	*D. phillipsii*
PC1	**17.7**	0
PC2	**30.6**	**21.0**
PC3	11.5	3.3
PC4	5.8	**58.2**
PC5	0.7	4.9
PC6	4.5	**9.4**
PC7	2.2	0
PC8	2.7	0
PC9	0.8	1.9
PC10	3.5	1.9
PC11	**19.9**	0

variables with the highest contribution in the 11 PCs used are listed in Table 2, taking into account the loading factors.

At this scale level, for the distribution model of *D. ornatus*, PC2, PC11, and PC1 had the highest contribution to the probability of occurrence of the species (68.2 %; Table 3). For the distribution model of *D. phillipsii*, PC4, PC2, and PC6 made the highest contribution to the probability of occurrence of the species (80.14%; Table 3). The variables maximum temperature of the warmest period (BIO5), mean temperature of the wettest quarter (BIO8), and mean temperature in the warmest quarter (BIO10) are elements of the PC with the highest contribution to the predictive distribution model for *D. ornatus*. For their part, seasonality of precipitation (BIO15), precipitation in the warmest quarter (BIO18), and topography (TOPO) are the variables with the greatest influence in the distribution model of *D. phillipsii*.

Figure 3 shows the omission rates and predicted area for both species. The calculated omission rates are expected to be close to the predicted omission rates (black line). Figure 3A shows that the omission rate of the calculated test records are close and above the predicted omission data; thus, it can be considered that the test records used for the model of *D. ornatus* are not spatially autocorrelated, and can be considered as an appropriate model. By contrast, Figure 3B, for the model of *D. phillipsii*, shows that the omission rate of the calculated test records is close to but below the predicted omission values, indicating that the training and test records are not independent and are spatially autocorrelated, due to the number of records used for training and testing, in addition to the clustering pattern of these records (Phillips *et al.* 2006).

Figure 4 shows the predicted distribution model for *D. ornatus* and the areas of occurrence predicted by this model. The cut-off threshold value selected was 0.173 (maximum test sensitivity plus specificity; Liu 2005). Predicted distribution areas were classified into low (0.173 to 0.5), medium (0.5 to 0.7), and high (0.7 to 0.951) probability of occurrence. The model shows an AUC = 0.963 for training data and an AUC = 0.954 for test data, with a standard deviation of 0.0068. This cut-off threshold was associated to an omission rate of test records equal to 0. The model shows that the predicted distribution area for this species is concentrated in the central region of Mexico in mostly dry climates, distributed across the States of Durango, San Luis Potosí, Zacatecas, Aguascalientes, Guanajuato, Querétaro, and Hidalgo. The predicted low, medium and high probability areas (green, yellow and red, respectively) were approximately 197,488 km^2, 30,894 km^{2,} and 20,927 km^2 (Figure 4).

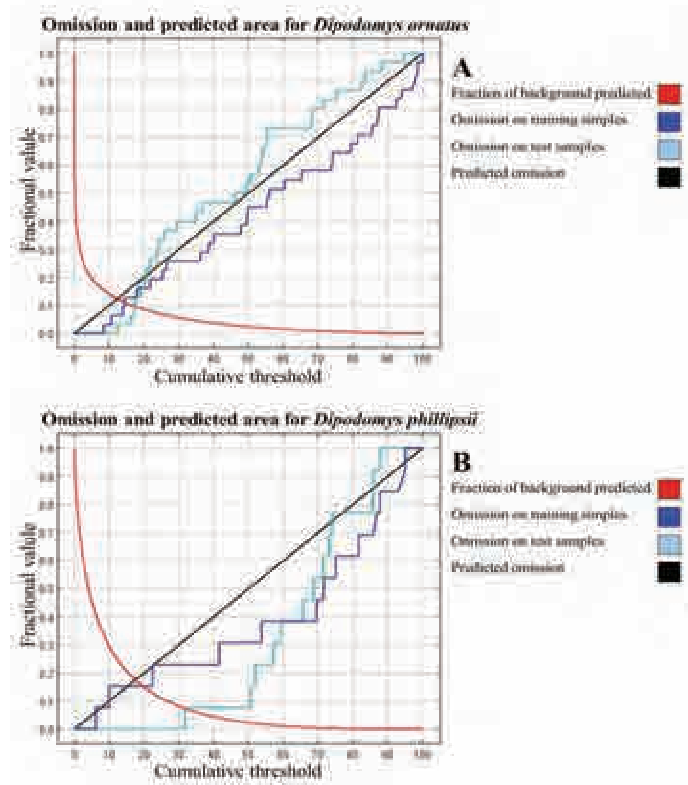

Figure 3. Omission rates and predicted area as a function of cumulative threshold.

Figure 5 shows the predicted distribution model for *D. phillipsii* and the areas of occurrence predicted by this model. The cut-off threshold value of the maximum test sensitivity plus specificity was 0.248. Similar to the model for *D. ornatus*, the predicted distribution areas were classified into low (0.173 to 0.5), medium (0.5 to 0.7), and high (0.7 to 0.987) probability of occurrence. The model shows an AUC = 0.930 for training data and an AUC = 0.987 for test data, with a standard deviation of 0.0053. This cut-off threshold was associated to an omission rate of test records equal to 0. The model shows that the predicted distribution area for this species is concentrated in the southern region of the country, particularly in the States of Puebla, Veracruz, Tlaxcala, Hidalgo, and the State of Mexico. Other areas with high a probability of occurrence are the southwestern part of the State of Nuevo León, center-south of Oaxaca, Chiapas, and center-north of Yucatan. The predicted low, medium and high probability areas (green, yellow and red, respectively) were approximately 115,203 km^2, 30,894 km^{2,} and 15,944 km^2 (Figure 5). Finally, the binomial test determined that the predictive distribution models for both species were better than random models ($P > 0.5$).

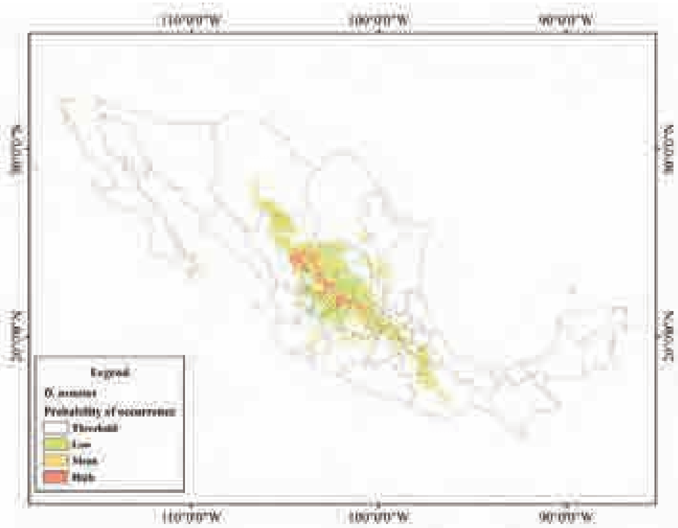

Figure 4. Map of the predictive distribution model for *Dipodomys ornatus* in Mexico.

Figure 5. Map of the predictive distribution model for *Dipodomys phillipsii* in Mexico

Discussion and Conclusions

Species distribution is determined by the availability of habitat with suitable environmental conditions, in addition to physical barriers such as rivers, mountains and topography that restrain their dispersal, as well as stochastic and anthropological processes (Wiser *et al.* 1998). Currently, mammals and other taxa are threatened due to multiple factors, including the loss of habitats associated to changes of land use from natural vegetation to cropland and human settlements, resource overexploitation, and climate change (Stuart *et al.* 2004; Thomas *et al.* 2004).

It is broadly acknowledged that endemic taxa are particularly susceptible to extinction, as a result of their restricted distribution, specificity of habitat and habits, and high vulnerability to environmental changes due to their low genetic variability, which reduces their ability to respond to selective pressures (Isik 2011). The geographical distribution of a species derives from three factors: biotic (preys, competitors, and predators), geographical space available for a species, and environmental conditions where a population can survive (fundamental niche, Maguire 1973). However, some of these conditions may also be found in adjacent areas, representing suboptimal sites where individuals of a species can spread to (Hutchinson, 1957; 1978, Soberón and Peterson 2005). For this reason, predictive species distribution models are an important tool in the analysis of potential habitats; however, this is a limited tool for species with restricted ranges, unique environmental requirements, and limited geographic records, thus restraining their monitoring and conservation (Rhoden *et al.* 2017), as is the case of some rodents. It should be noted that predicting the distribution of a suitable habitat of a species requires understanding its interactions in addition to the environmental variables in areas of occurrence (Baldwin 2009; Adhikari *et al.* 2012). For improving the conservation status of the species, potential area and habitat for reintroduction were predicted using Maximum Entropy (MaxEnt).

The MaxEnt models at national level showed an adequate performance in estimating the potential distribution of *D. ornatus* and *D. phillipsii*, yielding AUC values of 0.963 and 0.987, respectively; also, both models were validated by a binomial test and an analysis of omission rates. Although the records selected for both species showed a clustering pattern, as these failed the randomness test, these met the statistical requirements for final validation.

A key aspect is to determine that the environmental variables used for modeling are not spatially correlated; otherwise, the models obtained will tend to predict areas with a smaller surface, *i.e.*, the likely area of occurrence of a species is underestimated (Cruz-Cárdenas *et al.* 2014). For this reason, the environmental variables used should not be spatially correlated; alternatively, the dimensionality of variables should be reduced through statistical analyses such as PCA, which yields PCs that are projections with no correlation with the original variables. This type of statistical analysis should be used on climatic variables such as temperature and precipitation, as is the case of the WorldClim climate variables (Phillips *et al.* 2006).

The area with a high probability of occurrence, defined by environmental conditions that are favorable for *D. ornatus*, is situated mainly in the central region of Mexico, where the MP and the Chihuahuan Desert are located, both being areas with dry climate and extreme temperatures. By contrast, the potential distribution areas modeled for *D. phillipsii* are located mainly at TMVB and SMO, where environmental conditions are wetter with more stable temperate temperatures (INEGI 2016). This indicates habitat preferences of each species; however, it should be stressed that the areas predicted by SDMs are highly correlated with the geographical records used.

The predictive model for *D. phillipsii* yielded a high probability of occurrence in the south-west of the State of Nuevo León, in an area near the SMO covered by coniferous forest where this species has not been reported. In general, to validate and define the actual distribution of both species, field surveys should be conducted in areas with a probability of occurrence greater than or equal to 0.7. Suboptimal areas showing a probability of occurrence of 0.5 or above may be used as potential shelters in conservation strategies. These rodents are hard to capture given their restricted distribution, and SDMs can contribute to identify potential sampling areas.

Although these species are distributed in several States across the country, only some collection sites have been described in detail; consequently, the ecology of these species and their differences in habitat preference at the local level are poorly known. In general, both species live at altitudes between 950 and 2,850 m a.s.l., in sandy soils covered by low grasslands in association with different cactus species, including prickly-pear cacti, and low thorny shrubs; however, both have specific distribution requirements at a local level.

Dipodomys ornatus inhabits mainly flat deserts with dry climate covered by grasslands and shrubs; the areas of low and medium probability of occurrence obtained with the SDM match these habitats. The areas with a high probability of occurrence correspond to a semidry climate, mainly in natural grasslands of the Chihuahuan Desert and areas with low precipitation and soil moisture, corresponding to MP States (Durango, Zacatecas, San Luis Potosí, and Querétaro; INEGI 2016; CONAGUA 2016). More specifically and according to the literature, southwest to the city of San Luis Potosi, *D. ornatus* lives near low hills adjacent to oak forests (Dalquest 1953); in Durango, *D. ornatus* inhabits intermontane valleys covered with low grasslands, thorny shrubs (*Mimosa monancistra*), and cacti (*Opuntia*; Jones and Genoways 1975); in Zacatecas, specimens have been collected in areas with volcanic soils, in plains near hills with volcanic rocks, which during the past decade have been used for agricultural crops, forcing *D. ornatus* to survive in the edges of corn fields (change of land use and vegetation).

By contrast, *D. phillipsii* was captured in flat, desert habitats with temperate climate and sandy soils covered with shrubs and cacti; sites where this species has been recorded usually have either temperate-humid or warm-humid climate (INEGI 2016). In the States of Hidalgo, Puebla, Tlaxcala, State of Mexico, and Veracruz, the climate is humid due to proximity to the Gulf of Mexico and high precipitation (800-3,000 mm; CONAGUA 2016). Areas of high probability of occurrence, such as the Oriental Basin, where the natural vegetation has been replaced by maize crops (INEGI 2016), match collection sites where Genoways and Jones (1971) collected some specimens. In Veracruz, *D. phillipsii* is located in open, semi-arid areas covered by shrubs, cacti and agave, with wet climate (Hall and Dalquest 1963); in the State of Mexico and Hidalgo, *D. phillipsii* inhabits mainly semi-arid valleys covered by grasslands and surrounded by pine-oak forest; in Tlaxcala, specimens of *D. phillipsii* have been collected within maize crop fields (pers. obs. J. A. Fernandez).

Since the geographical records of the rodents studied are concentrated in MP, the Chihuahuan Desert and TMVB, the variables with the greatest influence in PCs were those related to temperature and precipitation. As observed from SDMs, *D. ornatus* and *D. phillipsii* show particular habitat preferences. These models, influenced by the geographical records and environmental variables used, in addition to the genetic evolution and the appearance of physical barriers such as the TMVB, and the multiple physiographic regions of Mexico that act as barriers that isolate geographically and genetically populations of these species, result in the differentiation of two groups defined by their geographical distribution and genetic identity, supporting the taxonomic proposal of Fernández *et al.* (2012). It is proposed that the altitude of the MP produces conditions that differ from those in high intermontane valleys of TMVB, resulting in two different potential distribution models influenced by different PCs and environmental variables.

Understanding how extrinsic factors influence the distribution of species and the selection of future habitats is key for researchers (Baldwin 2009). These questions can be solved with the assistance of SDMs, as these can provide useful information for the survey and prediction of areas with suitable conditions for the dispersal of species with a restricted distribution (Elith *et al.* 2011). In addition, SDMs allow the *a priori* selection of potential sampling areas, thus reducing costs and effort (Fois *et al.* 2015). It is concluded that MaxEnt is a valuable tool to describe and locate favorable areas for the expansion of populations of endemic species, or to describe more clearly the actual distribution of species in future research.

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First Record of *Myotis occultus* (Vespertilionidae) in the State of Guanajuato, Mexico

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The bat, *Myotis occultus* (Vespertilionidae) is a rare and small-sized bat. The known distribution of *M. occultus* extends from the semi-arid areas of the southwestern United States down as far as the middle half of Mexico, not having beeing previously registered in the state of Guanajuato in Mexico. In August 2008 and February 2013, we collected bats using mist nets in the locality of Llanos de Santa Ana located within the municipality of Guanajuato, Gto., Mexico. We collected twelve specimens of *M. occultus* at the entrance of an abandoned mine in the municipality of Guanajuato, which represents the first documented record of the species in Guanajuato and confirms its presence for the state as predicted by potential distribution maps of the species. It is a small-sized bat, with few records in Mexico and, according to previous studies, its distribution is confirmed for the states of Chihuahua, Zacatecas, Mexico State, Mexico City, and now Guanajuato.

Myotis occultus, es un murciélago vespertiliónido raro y de tamaño pequeño. La distribución conocida de *M. occultus* incluye las áreas semiáridas del suroeste de Estados Unidos, hasta la mitad central de México, sin haberse registrado previamente en el estado de Guanajuato, México. En agosto del 2008 y febrero de 2013, se realizó colecta de quirópteros utilizando redes de niebla en la localidad de Llanos de Santa Ana, Municipio de Guanajuato, Gto., México. Se recolectaron doce ejemplares de *Myotis occultus* en la entrada de una mina abandonada, lo cual constituye el primer registro documentado de la especie y se confirma su presencia para el estado de Guanajuato, México, de acuerdo con la distribución potencial para la especie. Es un murciélago de tamaño pequeño, con pocos registros en México, que de acuerdo con estudios previos se distribuye en los estados de Chihuahua, Zacatecas, Estado de México, Ciudad de México y ahora Guanajuato.

Key words: geographical distribution; Guanajuato; protected natural area; small sized bat.

Introduction

Thirty species of bats have been documented in Guanajuato (Sánchez *et al.* 2009; Sánchez *et al.* 2012; Sánchez 2014; Sánchez *et al.* 2016), a state located in central Mexico. However, no complete list showing the full diversity of bats in the state of Guanajuato yet exists; hence this number will undoubtedly increase as new studies are carried out in the area.

The bat, *Myotis occultus* (Vespertilionidae), is a small-sized vesper bat. This species differs from other species of *Myotis* from Guanajuato, *M. thysanodes* and *M. velifer*, for its medium size and different dorsal pelage coloration. *M. thysanodes* have big ears, and fringed uropatagium, whereas *M. velifer* has a small patch with few hairs (sometimes appearing as an almost-naked areas) in the middle part of the back, and both species have a well developed sagittal crest. *M. occultus,* is more similar in size to *M. yumanensis*, and both species lack a sagittal crest. *M. occultus,* has long, thick, and glossy fur on its back, which is black at the base and bright yellow at the tip (except for the ones on the base of the neck); it has large, dark brown or blackish ears with a long spear-shaped tragus, and *M. yumanensis* has dorsal hairs that are shorter with paler tips, pelage duller and not glossy, paler ears, and larger tragus having a semicircular border. In *M. occultus*, the braincase does not rise abruptly, and the middle and occipital regions are almost at the same level. In *M. yumanensis* the braincase rises at an abrupt angle from the rostrum, the mastoid breadth is smaller, and the calcaneum is not keeled. Dental formula of *M. occultus* is I 2/3, C 1/1, PM 3/3, M 3/3 = 38 (Ortega and Arita 2005; Medellín *et al.* 2008; Álvarez-Castañeda and Gónzalez-Ruiz 2015; Braun *et al.* 2015).

The species feeds on insects and migrates in winter to temperate regions (Humphrey 1982). The known distribution of *M. occultus* extends from the semi-arid areas of the southwestern United States down as far as the middle half of Mexico (Arroyo *et al.* 2008). It is a rare species with few records in Mexico, most of which have been documented in the states of Chihuahua (Anderson 1972), Zacatecas and Mexico State, as well as in Mexico City (Hall and Kelson 1959; Álvarez and Ramírez 1972; Urbano *et al.* 1987; Sánchez *et al.* 1989; Ortega and Arita 2005; Hortelano *et al.* 2016; Ríos-Muñoz *et al.* 2017).

Other studies report the presence of *M. occultus* in Aguascalientes (Chávez-Andrade *et al.* 2015) and Hidalgo (Mejenes-López *et al.* 2010, Ramírez-Pulido *et al.* 2017), but cite for this purpose other studies that are either unpublished (Aguascalientes) or report only a potential distribution within the borders of the state (Ortega and Arita 2005). In addition, a more recent checklist for Hidalgo, based on published papers, specimens in biological collections, and databases, does not include *M. occultus* (Sánchez-Rojas *et al.* 2016). Therefore, in the absence of verifiable, well-documented records, these states are not considered as part of the confirmed distribution of *M. occultus*.

This paper describes the first documented record in Guanajuato of the bat (*Myotis occultus*, Hollister 1909), based on 12 specimens caught in the locality of Llanos de Santa Ana in the municipality of Guanajuato, Mexico.

Materials and methods

Study Area. The locality of Llanos de Santa Ana is situated within the municipality of Guanajuato, Mexico, in the state of the same name. It is also located within the Protected Natural Area (PNA) of Cuenca de la Soledad, a state-level PNA classified as one requiring special attention in terms of ecological restoration, for being an important aquifer recharge area that feeds La Soledad reservoir, one of the main sources of water for the city of Guanajuato. The local vegetation corresponds to that of deserts and xeric shrublands, with a high rate of deforestation caused by the intense mining activity in the region.

On August 6, 2008, as part of a study of the bats in the area, we collected twelve specimens of *M. occultus* at the entrance of an abandoned mine (21° 03′ 25″ N, -101° 16′ 27″ W, 2,240 m.), located 4.25 km NW of Guanajuato (Figure 1), by using a 12 m x 2.5 m mist net that remained open from 18:00 h to 23:00 h. We identified individuals using keys for Mexican mammals (Medellín *et al.* 2008; Alvarez *et al.* 2015) and prepared the specimens for scientific study following the recommendations of Gannon *et al.* (2007). All specimens were deposited in the National Collection of Mammals (CNMA) at the Institute of Biology of the National Autonomous University of Mexico (UNAM) under catalog numbers CNMA 44523 to 44532 and 47305-47306. Prior to collecting the specimens, we first obtained the necessary permit (number FAUT-0070) from the Ministry of Environment and Natural Resources (SEMARNAT). Somatic and cranial measurements were registered following Álvarez-Castañeda and Gónzalez-Ruiz (2015).

We obtained Cytochrome Oxidase I (COI) gene sequences for five of the specimens collected, CNMA 44523- 44527 (GU686195, GU686214, GU686212, GU686213, GU686193, respectively), which are available on the BOLD Systems website and GenBank for future studies but they were not used to corroborate the identity of the specimens.

Results

The twelve specimens collected were reproductively inactive males. The range and average external measurements (in mm) of these individuals were: total length (TL): 71 - 84, = 79.95; length of tail (LT): 30 - 38, = 34.5; length of hind foot (LHF): 6 - 14, = 9.58; length of ear (LE): 11 - 14, = 13.4; and length of forearm (LF: 31 - 36, = 33.5; Figure 2). The average cranial and mandibular measurements were: length of skull (LS): 13.70 - 14.34, = 13.98; cranial width (CW): 6.86 - 7.60, = 7.12; cranial height (CH): 5.96 - 6.58, = 6.17; zygomatic breadth (ZB): 7.72 - 8.28, = 8.06; mastoid breadth (MB): 6.08 - 7.06, = 6.94; interorbital breadth (IOB): 3.62 - 3.96, = 3.79; breadth across upper molars (BAM): 5.20 - 5.24, =

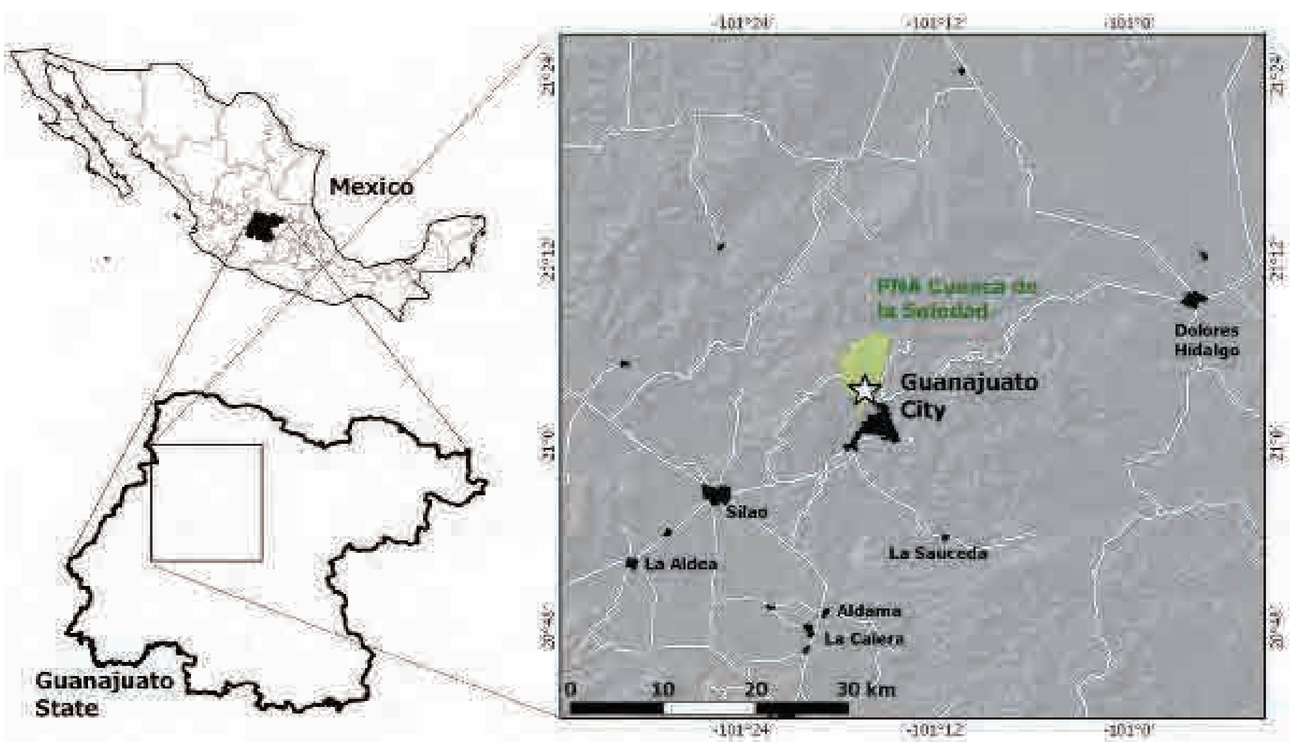

Figure 1. Geographic location of the collection site of *Myotis occultus* in the Protected Natural Area (PNA) Cuenca de la Soledad, Municipality of Guanajuato, Gto., Mexico.

5.13; maxillary toothrow length (MAX): 5.09 - 6.01, = 5.78; condylobasal length (CBL): 11.25 – 12.00, = 11.70; and mandibular toothrow length (MAN: 6.26 - 6.58, = 6.44; Figure 3).

Figure 2. A specimen of *Myotis occultus* collected in the Protected Natural Area (PNA) of Cuenca de la Soledad, Guanajuato, Mexico (Photo Y. Hortelano).

Discussion and Conclusions

The presence of *M. occultus* in the state of Guanajuato is consistent with the potential distribution of the species (Ortega and Arita 2005). Therefore, this is the first record of *M. occultus* for the state supported with voucher specimens. In addition, we uploaded DNA sequences at GenBank for further studies.

Until now, the list of bats in the state of Guanajuato includes 30 species (Sánchez *et al.* 2016). However, this figure is still somehow low compared to that of areas that have undergone major urban development such as the Mexico City Metropolitan Area, where 28 species have been described (Sánchez *et al.* 1989; Hortelano-Moncada *et al.* 2016). Furthermore, the state of Guanajuato encompasses portions of two biogeographic regions: the Neartic and the Neotropical, in an area where the Sierra Madre Oriental, the Trans-Mexican Volcanic Belt, and the Mesa Central come together, a situation that provides a noteworthy diversity. We suggest that more exploration studies be done to obtain a better approximation of its biological diversity.

Figure 3. Dorsal, ventral, and lateral views of skull and lateral view of mandible of an adult male of *Myotis occultus* collected in the Protected Natural Area (PNA) of Cuenca de la Soledad, Municipality of Guanajuato, Gto., Mexico (Photo Carmen Loyola).

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Morphology and stomach content of the Goldman´s diminutive woodrat *Nelsonia goldmani* (Cricetidae: Neotominae)

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Goldman´s diminutive woodrat (*Nelsonia goldmani*) is an endemic rodent that inhabits the temperate and humid environments of the central highlands of Mexico. This species is considered uncommon because of the scarce and dispersed information about specimens collected across the Faja Volcanica Transmexicana. Therefore, it is crucial to generate new information about the basic biology of *N. goldmani*, which so much is unknown thus far. We present the morphological description of the stomach and its content from one specimen of *N. goldmani*. We performed a longitudinal bisection and washing of stomach from one adult male collected at the Natural Park "Las Peñas" in the municipality of Jilotepec, Estado de México, Mexico. The macroscopic structures of the stomach were described by observation in a stereoscopic microscope. The stomach content was mounted on sliders and its components were identified and photographed with an optical microscope. According to the gastric glandular epithelium distribution, *N. goldmani* have a bilocular-discoglandular stomach with a characteristic *fornix ventricularis* slightly elongated horizontally. The stomach content was composed by several plant items: epidermal tissues of steam and leaves of angiosperms, pollen of gymnosperms, spores of ferns, fungi and animal tissue like mouth appendages and legs of insects. The stomach morphology was similar to the *N. neotomodon* and the peromyscine *Neotomodon alstoni* described previously, but the flattened and elongated *fornix ventricularis* found in *N. goldmani* was distinctive. Nonetheless, this structure can vary between individuals and mainly depends of the amount of food before dissection. The plant material found in the stomach was similar to that reported in other rodents that inhabit in similar environments to the highlands of central Mexico, such as *Peromyscus aztecus*, *P. difficilis* and *Reithrodontomys fulvescens*.

La rata enana michoacana (*Nelsonia goldmani*) es un roedor endémico que habita en los ambientes templados y húmedos de la zona montañosa del centro de México. Esta especie es considerada como poco común debido a la escasa información disponible para los especímenes colectados en la Faja Volcánica Transmexicana. Por lo tanto, es indispensable generar información nueva de la biología básica de *N. goldmani* mucha de la cual se desconoce hasta el momento. Presentamos la descripción morfológica del estómago y su contenido en un ejemplar de *N. goldmani*. Se realizó un corte longitudinal y un lavado del estómago de un macho adulto colectado en el Parque Natural "Las Peñas" en el Municipio de Jilotepec, Estado de México, México. Se describieron las estructuras macroscópicas del estómago por observación en un microscopio estereoscópico. El contenido estomacal fue montado en laminillas y sus componentes fueron identificados y fotografiados en un microscopio óptico. Según la distribución del epitelio glandular gástrico, *N. goldmani* tienen un estómago bilocular-discoglandular con un característico *fórnix ventricularis* ligeramente alargado horizontalmente. El contenido estomacal estuvo compuesto por varios elementos vegetales: tejido epidérmico de tallos y hojas de angiospermas, polen de gimnospermas, esporas de helechos, hongos y tejido animal de aparatos bucales y patas de insectos. La morfología estomacal fue similar a la de *N. neotomodon* y el peromyscino *Neotomodon alstoni* descritos previamente, pero el *fórnix ventricularis* aplanado y alargado encontrado en *N. goldmani* fue distintivo. Sin embargo, esta estructura puede variar entre individuos y depende principalmente de la cantidad de alimento antes de la bisección. El material vegetal estomacal encontrado fue similar al reportado en otros roedores que habitan en ambientes similares a las zonas altas del centro de México como *Peromyscus aztecus*, *P. difficilis* y *Reithrodontomys fulvescens.*

Keywords: Cloud forest; Estado de México; feeding; rare species; rodentia.

Introduction

Mexico hosts a wide diversity of rodents with an estimated number of 233 species, which 140 belongs to the family Cricetidae (Ramírez-Pulido *et al.* 2014). Nevertheless, most of these species have scarce information about its basic biology, such as demography, ecological role in their communities, and history life, to mention some examples (Fernández *et al.* 2014). Furthermore, some of these rodents are listed under some category of extinction risk, and for which the lack of information is critical, for instance, for the development of plans aimed to their habitat conservation (SEMARNAT 2010).

One of these species is the Goldman´s diminutive woodrat *Nelsonia goldmani* Merriam 1903, an endemic taxon that inhabits temperate and humid regions of highlands of central Mexico. It is considered an uncommon species due to few specimens collected after being discovered and its fragmented geographical distribution across the biogeographical region known as Faja Volcánica Transmexicana (León-Tapia 2013). As well, this species is under "Special

Protection" by the NOM-059-SEMARNAT-2010 (SEMARNAT 2010) in Mexico and "Endangered" by the by the International Union for Conservation of Nature (IUCN; Álvarez-Castañeda and Castro-Arellano 2008).

Most of the basic aspects of the biology of this species is unknown, mainly related to the few biological material available of this rodent and making difficult to carry out some studies such as particular internal morphology and the diet. Therefore, in this study we describe for first time the macroscopic stomach morphology and its content from one individual of *N. goldmani* in order to provide information related to the digestive system morphology and the food items consumed by the rodent.

Materials and Methods

One adult male of *N. goldmani* was collected with the authorization FAUT-0002 on October 16 2010 and vouchered in the Colección Nacional de Mamíferos (CNMA 46291), at the Natural Park "Las Peñas", Jilotepec, Estado de México, Mexico (Figure 1; León-Tapia 2013). The vegetation at the collecting locality was mainly oak forest (*Quercus sp.*) with some areas with fir and montane cloud forests (Engstrom *et al.* 1992; Carleton *et al.* 2002).

Figure 1. Geographic location of the specimen of *Nelsonia goldmani* (CNMA 46291) collected at the Faja Volcánica Transmexicana in México (yellow polygon). The green dot shows the collecting locality of the specimen close to the main towns of the region in purple polygons.

From this specimen we removed the stomach *in situ* cutting the end of the esophagus and duodenum, immediately preserved it in 70 % alcohol, and posteriorly stored it at -72 °C. For the stomach description and the terminology of the structures, the classical methodology stated by Careton (1973) was used. The stomach was placed in a laboratory dish and a ventral bisection was made in a horizontal plane, the stomach content was extracted with 70% alcohol and conserved at 4 °C for posterior analyses. We carried out the observation of stomach in a stereoscopic microscope (Nikon SMZ445) and the illustration of stomach and structures was made.

The stomach content was analyzed according to the procedure used to determinate the diet of *Peromyscus difficilis* (Morales-Medina 2010; Peralta-Juárez 2015). The stomach material was disaggregated on a glass slide, covered with a coverslip and sealed for its preservation. Subsequently, observations were carried out in an optic microscope Zeiss Axiophot 7082 in order to locate the stomach items. During this process, pictures were taken with a digital microscope camera Moticam 2000 and the software Motic Image Plus 2.0. We used different colored filters in addition to black and white filters for color investment in order to highlight the desired stomach items. Finally, the pictures were edited for emphasizing some structural features of such items in order to taxonomically identify them using specialized literature (Linconff 1988; Andrews and Caballero 1989; Phillips 1991; Guzmán 1998; Palacios-Vargas *et al.* 2014).

Results

The specimen of *N. goldmani* was taxonomically identified by both external and internal characteristics (Figure 2). According to the color nomenclature (Ridgway 1912), we determined that it was dark dorsally (buffy olive at tips and blackish slate at base), white ventrally, laterally olive lake and the hind feet white with olive lake color at the upper parts. The tail was large with long liver brown hairs dorsally

Figure 2. Photographs of the *Nelsonia goldmani* (CNMA 46291). From top to bottom: individual feeding on seeds of oak (*Quercus* sp.), dorsal, ventral and lateral view of the skull.

and cream color ventrally, but no completely bicolored and with a distinctive terminal tuft of hair. The external measurements in mm were: total length 260, tail length 140, hind foot length 25 and ear length 24. The skull was flattened with the presence of the diagnostic anteorbital zygomatic notch and the singular prismatic molar pattern. The cranial measurements according to González-Cózatl *et al.* (2016) were: greatest length of skull 32.8, length of rostrum 13.11, breadth of zygomatic plate 3.76, mastoid breadth 14, zygomatic breadth 16.67, interorbital constriction 4.63, breadth of rostrum 5.51, width of interparietal bone 10.24, breadth across molars 6.33 and depth of skull 10.39.

Stomach description. According to the gastric glandular epithelium arrangement, *N. goldmani* has a bilocular-discoglandular stomach (Figure 3). The *incisura angularis* is deep and proyected beyond the esophageal opening forming the stomach bilocular condition well defined. *The fornix ventricularis* archs are located further from the esophageal opening and slightly curved to the esophagus. The size of *corpus*, mainly in *fornix ventricularis* section, is elongated horizontally. The *antrum* walls are muscular, more visible near the pyloric opening. A bordering fold surrounds the glandular epithelium, and restricted by a small curved area

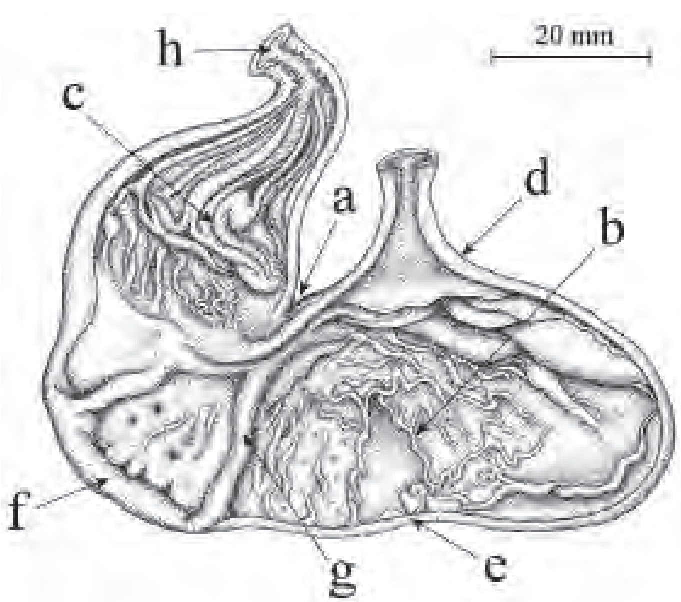

Figure 3. Illustration of the macroscopic bilocular-discoglandular stomach structures of the Nelson and Goldman's woodrat *Nelsonia goldmani* (CNMA 46291). a) *incisura angularis*, b) *corpus*, c) *antrum*, d) *fornix ventricularis*, e) cornified squamous epithelium, f) glandular epithelium, g) bordering fold, and h) anterior end of duodenum.

on the left side of the grandular zone at the bottom of the stomach. The cornified squamose epithelium coats the remaining areas of the *antrum* and *corpus*.

Stomach content. Several food items of plant origin were detected in the stomach content, such as epidermal tissue mainly consisting of stems and leaves, pollen of gimnosperms and spores of ferns and fungi. Likewise, animal tissues were observed, such as mouth appendages and legs of insects (Figure 4). The plant tissues were most frequent in the analyzed samples, and the spores were scarce.

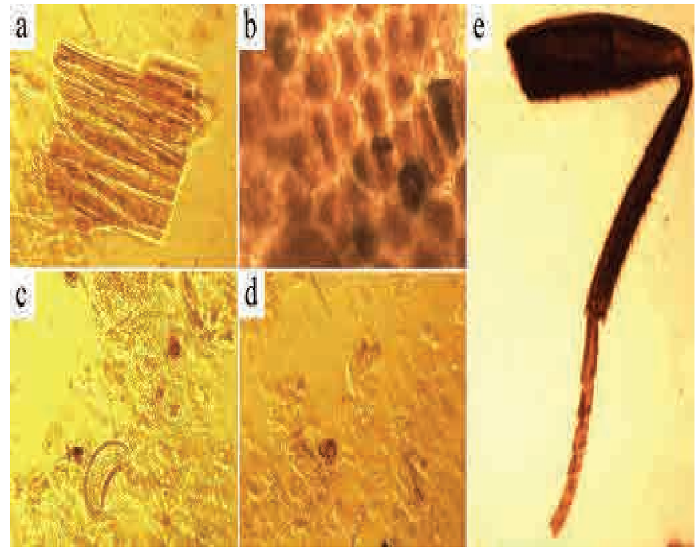

Figure 4. Pictures from optical microscopy at 40x of food items found in a stomach of the Nelson and Goldman's woodrat *Nelsonia goldmani* (CNMA 46291). a) epidermal tissue from stems, b) epidermal tissue from leaves, c) fern spores, d) gymnosperms pollen, and e) leg of an insect.

Discussion

The stomach morphology of *N. goldmani* was similar to its sister species *N. neotomodon*, described previously by Careton (1973), with the exception of the *fornix ventricularis*, which is further flattened and elongated. Nonetheless, it has been shown that this structure can vary between individuals and mainly depends of the amount of food before dissection. Hooper (1954) pointed out that some stomach structures like the pronounced *incisura angularis* in rodents as *N. goldmanik,* with a bi1ocular stomach, could increase the area for intensive mixing and reworking of the food bolus. However, this information is limited to make inferences about feeding.

Regarding to the stomach content, it was possible to identify that this individual ingested mainly plant items. Similarly, the insect items could indicate that were occasionally ingested by the specimen. These results were similar to the observations reported for *N. neotomodon* from one specimen from San Luis, where the mainly food items were a green mass with the appearance of fir (*Pseudotsuga menziesii*) or junipers (*Juniperus* sp.) needles (Hooper 1954).

Prior to the capture of the specimen of *N. goldmani*, we observed the individual feeding on lichens on the bark of trees and photographed it eating seeds of *Quercus* sp. (Figure 2). These observations could indicate that this species likely eats a wide plant resource available in the locality. However, it is indispensable to carry out specific studies throughout the years in order to have an approximation about its diet.

The diet for the two species of the genus *Nelsonia* is unknown mainly due to the difficulties to capture these rodents. Even so, the findings in this study contribute to the knowledge of the basic biology of *N. goldmani* and the results are comparable to those reported for other rodent species that inhabit environments apparently similar in the highlands of central Mexico, such as *Peromyscus aztecus*, *P. difficilis* and *Reithrodontomys fulvescens*. These rodents feed preferentially upon plant material of different composition according to the conservation condition of their habitat (Vázquez *et al.* 2004; Peralta-Juárez 2015).

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Potential distribution of *Sphaeronycteris toxophyllum* in Colombia and new record

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The visored bat, *Sphaeronycteris toxophyllum* Peters, 1882, is a species listed as Data Deficient by the International Union for the Conservation of Nature, being considered rare throughout its geographical range due to the small number of specimens recorded. This study reports a new record that broadens the distribution range of the species in Colombia to the central mountain range and upper valley of the Magdalena river. One adult male of *S. toxophyllum* was captured after a sampling effort of 3,870 net-hours over 36 nights in the Andean region, at the upper valley of the Magdalena river, Department of Tolima, Colombia. Its diet was analyzed and a model for Colombia was built. The morphometric analysis of the specimen captured yielded values lower than those previously reported for Colombia. The analysis of stomach contents identified two floristic elements as part of its diet, corroborating the frugivorous feeding habits of this species. Separately, the application of species distribution modeling techniques established that *S. toxophyllum* has a potential geographic range that continues north of the Chocó-Magdalena province and west of the Orinoco and Guayana biogeographical provinces of Colombia. Based on these data, we propose that the extended distribution range reported here may be associated with a current connection between these geographical provinces.

El murciélago de vísera *Sphaeronycteris toxophyllum* Peters, 1882, está catalogado con Datos Deficientes por la Unión Internacional para la Conservación de la Naturaleza y es considerado raro a lo largo de su distribución, debido al escaso número de ejemplares conocidos. Este estudio presenta un nuevo registro que extienden la distribución geográfica de la especie en Colombia, hacia la cordillera Central y el valle alto del río Magdalena. Durante 36 noches y un esfuerzo de muestreo de 3,870 horas-red en la región andina en el valle alto del río Magdalena en el departamento de Tolima, Colombia, se capturó un macho adulto de *S. toxophyllum*. Se analizó su dieta y se construyó un modelo de distribución potencial para Colombia. El espécimen capturado registró medidas morfológicas craneales inferiores a las reportadas en trabajos previos para Colombia. El análisis del contenido estomacal permitió identificar dos elementos florísticos como parte de la dieta de la especie, información que permite corroborar su frugivoría. A través de la aplicación de técnicas de modelaje de distribución de especies fue posible establecer que *S. toxophyllum* presenta un intervalo geográfico potencial que se extiende al norte de la provincia Choco-Magdalena y al oeste de las provincias biogeográficas de la Orinoquia y de la Guayana de Colombia. Los datos permiten proponer que la ampliación de distribución presentada aquí puede estar asociada a la conexión existente entre estas provincias geográficas.

Key words: Chiroptera; distribution; geographic range; tropical dry forest; rare species.

Introduction

Sphaeronycteris toxophyllum Peters, 1882, is one of the least known species of fruit bats in the Neotropics due to the small number of collections throughout its distribution range (Emmons and Feer 1990). The reported distribution includes Colombia (Sanborn 1941; Cuervo-Díaz *et al.* 1986; Gardner 2008; Rodríguez and Gonzales 2012); Bolivia (Koopman 1976; Anderson 1997); Brazil (Piccinini 1974; Peracchi 1986), Ecuador (Albuja and Mena 1991); Peru (Rehn 1901; Solari *et al.* 1998) and Venezuela (Thomas 1898; Handley 1976). It has been observed from the Amazon basin to 3,000 masl in secondary, cloud, and deciduous forests, as well as in open areas (Angulo *et al.* 2008). However, these are isolated records involving a few individuals.

In Colombia, the species has been recorded in the Departments of Amazonas (Rodríguez-Posada and Cárdenas-González 2012); Boyacá (Gallardo *et al.* 2014); Caquetá (Montenegro and Romero 1999); Casanare (Rodríguez-Posada and Cárdenas-González 2012); Cundinamarca (Muñoz 2001; Solari *et al.* 2013); Guainía (Sanborn 1941); Magdalena (Cuervo-Díaz *et al.* 1986); Meta (Rodríguez-Posada and Cárdenas-González 2012); Norte de Santander (Sanborn 1941), Vaupés and Vichada (Rodríguez-Posada and Cárdenas-González 2012; Solari *et al.* 2013). These records correspond of a number of different natural regions of the country, including tropical humid forests, dry forests, montane forests, savannas and urban centers (Rodríguez-Posada and Cárdenas-González 2012).

Despite the broad distribution of *S. toxophyllum* according with the International Union for the Conservation of Nature (IUCN 2018), it is classified as Deficient Data and there is concern that this status may shift to Near Threatened in the future. The distribution range reported for this species comprises the Central mountain range in the upper valley of the Magdalena river and the Tropical Dry Forest (BST) of Colombia, a highly fragile and threatened ecosystem. This study discusses the rarity of *S. toxophyllum* based on existing records, as well as its potential distribution area.

It incorporates the first dietary records of the species for BST, filling previous information gaps.

Materials and Methods

Sampling was carried out in 2014 to assess the trophic structure of the bat fauna at Centro Universitario Regional del Norte, located in the upper valley of the Magdalena river, in the municipality of Armero Guayabal, Department of Tolima, Colombia (5° 0′ 21.49″ N, -74° 54′ 28.42′′W; Figure 1). This locality has an altitude of 240 masl, with a mean annual temperature of 28 °C, and a precipitation of 1,791 mm (CORTOLIMA 2007).

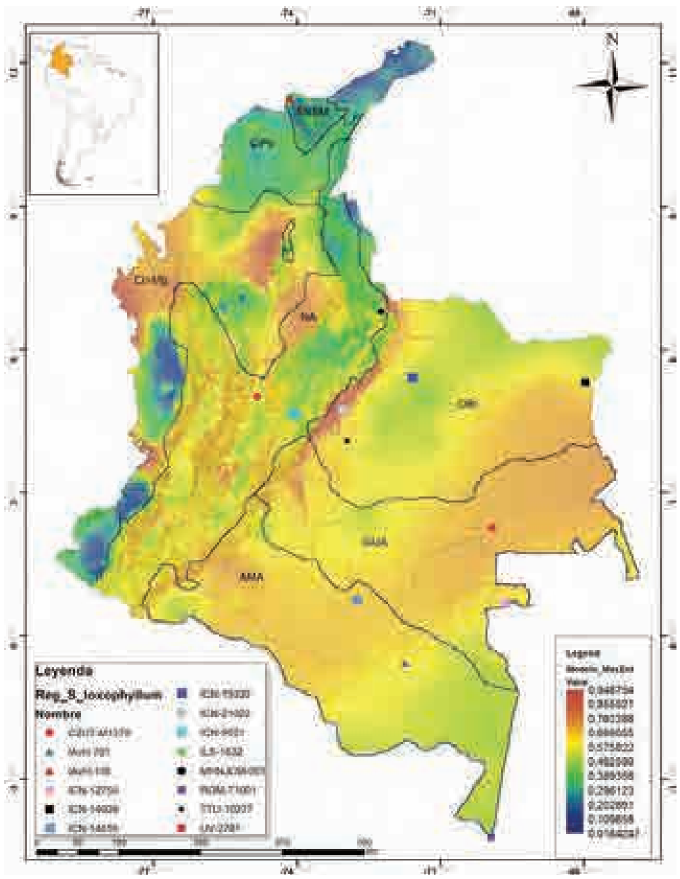

Figure 1. Model of potential distribution of *Sphaeronycteris toxophyllum* in Colombia and records in the collection of mammals of the Instituto de Ciencias Naturales at Universidad Nacional de Colombia ICN (squares), Instituto de Investigación de Recursos Biológicos Alexander von Humboldt IAVH (triangles), Museo La Salle ILS (pentagon), Museo de Historia Natural José Celestino Mutis at the Universidad de Pamplona MHNJCM (black circle), Royal Ontario Museum ROM (purple square), Texas Tech University TTU (star), collection of mammals of the Universidad del Valle UV (red square) and a new locality reported of the Zoological Collection at Universidad del Tolima CZUT (red circle). Black lines show the boundaries of biogeographical regions according to Hernández-Camacho et al. (1992); CiPe, peri-Caribbean arid belt; SNSM, Sierra Nevada de Santa Marta; Ch-Mg, Chocó-Magdalena; NA, North-Andean; ORI, Orinoco; GUA, Guayana; AMA, Amazon.

One adult male of *S. toxophyllum* was captured after a sampling that involved six mist nets measuring 6 x 2.5 m and three measuring 12 x 2.5 m, with a sampling effort of 3,870 net-hours over 36 nights. The specimen captured was handled according to the recommendations of the American Society of Mammalogists (Sikes *et al.* 2011) regarding capture and handling procedures. A series of standard morphometric measurements (Simmons and Voss 1998) of this specimen were determined with a Mitutoyo gauge accurate to 0.1 mm (Table 1), and the weight was measured with an electronic scale accurate to 0.1 g. Age was estimated based on the degree of ossification of epiphyseal growth plates of finger phalanges (Dietz *et al.* 2009), and the reproductive condition was determined according to Kunz *et al.* (1996).

The individual was identified using the taxonomic keys proposed by Gardner (2008). It was preserved in skin and skull in the Zoological Collection of Universidad de Tolima, Mammalogy Section, under catalog number CZUT-M 1,379.

A distribution model was elaborated for *S. toxophyllum* from 14 presence records in various regions of Colombia, in the Departments of Amazonas, Boyacá, Casanare, Caquetá, Cundinamarca, Magdalena, Meta, Norte de Santander, Tolima, Vaupes and Vichada; provisions were taken to ensure that data be obtained from reliable information sources: scientific literature and biological collections (Table 2). Each individual datum was recorded systematically, including geo-referenced information (latitude and longitude), Department, and locality. The information was screened using the Google Earth Pro application to determine the geographical location of each individual record; only properly georeferenced data were used.

The potential habitat of *S. toxophyllum* was modeled using the software Maxent v3.4.1. This program determines the relationship between climatic variables and records of the species (Guisan and Thuiller 2005), and assumes that the climate in the observation points of a species represents its environmental range; therefore, climate was used as a model predictor (Jarvis *et al.* 2005). We used historical values of 19 climatic variables (Table 3) from the global database WorldClim version 2 (http://worldclim.org) accessed on 23 June 2018. The model was evaluated statistically by analyzing the area under the curve (AUC), which ranges from 0 to 1; values close to 1 indicate has good performance of the model, while values near or below 0.5 indicate that the model has no predictive power (Guisan and Thuiller 2005).

Table 1. External and cranial measurements of the male of *Sphaeronycteris toxophyllum* from Armero Guayabal, Department of Tolima (Tolima), data from a male by Rodriguez and Gonzales (2012; RyG) and a females form Gallardo *et al.* (2014). mesaurementes in milimeters (min. - max.).

Variables	Tolima	Rodríguez y Gonzales	Gallardo *et al.*
Total length	53.64	47.70 - 54.00	54.00
Forearm length	37.67	37.00 - 38.07	38.00
Tibia length	15.27	16.90 - 18.76	16.80
Ear length	8.25	8.85 - 15.00	11.20
Maximum skull length	15.76	15.80 - 16.14	16.50
Condyle-incisive length	12.47	14.16 - 14.47	14.46
Condyle-canine length	12.09	13.59 - 14.22	14.12
Postorbital width	5.23	5.29 - 5.78	5.94
Zygomatic width	11.63	11.74 - 12.07	11.40
Skull width	8.15	8.76 - 9.10	9.60
Mastoid width	9.33	9.94 - 10.12	10.12
Length of maxillary tooth row	4.35	4.37 - 4.56	4.45

Results

The presence of *S. toxophyllum* in the central mountain range of Colombia is reported herein for the first time. This record is based on a single specimen collected in the northern portion of the Department of Tolima, in the upper valley of the Magdalena river. This record broadens the known distribution of *S. toxophyllum* to approximately 110 km west of the nearest known locality in the Department of Cundinamarca (ICN- 9521, Figure 1).

The collection locality shows vegetation elements typical of Tropical Dry Forest (BST), with predominance of clayey, silt-clayey and loamy soils, with trees measuring less than 8 m in height and shrubs up to 3 m in height. Some areas show vegetation in early successional stages, such as *Acacia farnesiana*, *Acalypha macrostachya*, *Calliandra pittier*, *Cecropia peltata*, *Centrosema pubescens*, *Cordia alliodora, Crataeva tapia, Crotalaria incana, Croton schiedeanus, Cnidoscolus urens, Chiococca alba, Henriettella fissanthera, Maclura tinctoria, Melochia pyramidata, Mimosa pigra*, *Myrtus communis*, *Piper aduncum, Piper bogotense, Pithecellobium dulce, Sapium glandulosum, Solanum betaceum, Solanum nigrum, Solanum quitoense, Tabernaemontana grandiflora* and *Vernonanthura brasiliana*.

The specimen of *S. toxophyllum* collected has all of the diagnostic characters of the species (Husson 1958; Emmons and Feer 1997; Gardner 2008; Angulo *et al*. 2008). However, the majority of its morphological measures are smaller than those reported previously (Table 1; Gallardo *et al*. 2014; Rodríguez-Posada and Cárdenas-González 2012). The specimen has long and tri-color pelage: dorsum brown to dark brown with a grayish shade and individual hairs whitish in the center of the dorsum, white spot on the shoulder near the wing; anterior part lighter than the posterior, and abdomen brown.

Rostrum short, less than half the braincase length; nasal and maxillary bones projected upwards, oriented vertically in the same plane as the palatine process; nasal opening located at the base of the skull, which resembles a primate in lateral view; anterior margin of the orbit extends to form a conspicuous plate; palatal emargination V-shaped, shallow and extended approximately to half of second upper molar (Figure 2).

The analysis of the stomach contents of *S. toxophyllum* revealed 70 % plant cell tissue (pulp) and 30 % seeds, belonging to *Chiococca alba* (*n* = 7; 23.1%) and *Henriettella fissanthera* (*n* = 2; 6.6%). The following taxa were reported together with the capture of *S. toxophyllum*: Emballonurinae: *Saccopteryx bilineata*, *S. leptura*. Phyllostomidae: *Carollia brevicauda*, *C. perspicillata*, *Desmodus rotundus*, *Glossophaga soricina*, *Phyllostomus discolor*, *P. hastatus*, *Trachops cirrhosus*, *Artibeus lituratus*, *A. planirostris trinitatis*, *Dermanura phaeotis*, *Mesophylla maconnelli*, *Platyrrhinus helleri*, *Uroderma convexum*. Vespertilionidae: *Eptesicus brasiliensis*, *Myotis nigricans*, *M. riparius*, *Rhogeessa io.* Molossidae: *Molossus molossus*.

The distribution model for *S. toxophyllum* yielded an average AUC of 0.94 (range: 0.926-0.949), indicating a good performance with low levels of commission errors and identifying all the localities where the species has been reported (Figure 1). The empirical evaluation allows establishing the usefulness and efficiency of the model to find potential sites of occurrence of the species.

The model predicts a distribution in four of the nine biogeographical provinces proposed for Colombia by Hernández-Camacho *et al.* (1992). The highest probability of occurrence is recorded in the northern part of the Chocó-Magdalena biogeographical provinces in the Departments of Antioquia, Chocó, southern Bolivar and Sucre, and east of

Table 2. Records of the Colombian localities with records of *S. toxophyllum* used for the model of potential distribution of the species. *New record.

Acronym	Locality	Latitude	Longitude
IAvH 701	Amazonia, corregimiento La Chorrera	-0.56663	-71.74994
ROM 71001	Amazonia, Leticia	-3.78472	-69.94056
MHNJCM-001	Boyacá, Cubará	7.04716	-72.16939
ICN- 9521	Bogotá, Ciudad universitaria.	4.64360	-74.14406
ICN- 21022	Casanare, San Jorge trail	4.72086	-73.04997
ICN- 19320	Casanare, El Banco de la Cañada trail	5.41661	-71.59850
ICN 14616	Caquetá	0.74737	-72.73729
UV 2781	Guainía, corregimiento Morichal	2.26370	-69.91847
IAvH 116	Magdalena, Sierra Nevada de Santa Marta	11.25000	-74.16339
TTU 10277	Meta, municipality of Puerto López	4.08500	-72.95528
ILS 1632	Norte de Santander, municipality of Pamplona	7.37093	-72.65237
CZUT-M 1379*	Tolima, municipality of Armero Guayabal	4.64360	-74.14406
ICN 12750	Vaupés, municipality of Taraira	0.56474	-69.63411
ICN 14009	Vichada, Tuparro Natural National Park	5.30750	-67.97128

Table 3. Climatic variables used in the program MaxEnt to produce the map of potential distribution of *S. toxophyllum* for Colombia.

Climatic Variables
1. Mean annual temperature (°C)
2. Diurnal temperature variation (°C)
3. Isothermality (ratio between parameters 2 and 7)
4. Temperature seasonality (coefficient of variation %)
5. Mean maximum temperature of the warmest season (°C)
6. Mean minimum temperature of the coldest season (°C)
7. Annual temperature variation (difference between parameters 5 and 6)
8. Mean temperature of the rainiest four-month period (°C)
9. Mean temperature of the driest four-month period (°C)
10. Mean temperature of the warmest four-month period (°C)
11. Mean temperature of the coldest four-month period (°C)
12. Annual precipitation (mm)
13. Precipitation of the rainiest period (mm)
14. Precipitation of the driest period (mm)
15. Precipitation seasonality (coefficient of variation %)
16. Precipitation of the rainiest four-month period (mm)
17. Precipitation of the driest four-month period (mm)
18. Precipitation of the warmest four-month period (mm)
19. Precipitation of the coldest four-month period (mm)

Figure 2. Dorsal, ventral and side view of skull and jaw of *Sphaeronycteris toxophyllum* (CZUT-M 1379) male from Armero Guayabal, Department of Tolima, Colombia. A) nasal and maxillary projected upwards with nasal openings at the base of the skull.

Córdoba; west of the Orinoco biogeographical province in the Departments of Arauca and Casanare, and north of the biogeographical province of Guayana in the Department of Meta, the northwestern zone of the North Andean biogeographical province in the Department of Santander; the rest of the latter province records a medium-to-low probability of occurrence of this species.

The annual temperature range was the variable that contributed more information to the model when used individually, being the one containing the greatest amount of information, unlike any other variable. In general, the variables that contributed most information to the distribution model of *S. toxophyllum* are those related to temperature (temperature seasonality and isothermality).

Discussion

The available information on *S. toxophyllum* in Colombia reveals its distribution in contrasting environments. To date, however, there were no records of its presence in the Cordillera Central, nor in the inter-Andean valleys of the Magdalena and Cauca rivers. The existing records of the species correspond to very dry forests, as those that cover the northwestern portion of the North-Andean biogeographical province in northern Santander (Sanborn 1941), as well as in humid tropical forests, in Boyacá (Gallardo *et al.* 2014). These latter authors reported the presence of the species in the boundary between Cundinamarca and Tolima corresponding to the specimen with voucher ID ICN-Mastozoologia 9521. However, the review of the information provided revealed that this is a wrong location on the distribution map, as the report was gathered in the locality Ciudad Universitaria, Department of Cundinamarca, Bogotá D. C., 11 September 1986; this specimen is deposited in the "Alberto Cadena García" mammal collection of the Institute of Natural Sciences at the National University of Colombia.

To note, the record reported here not only represents the first report of the species for the Central Cordillera, but also implies an extension of the ecological range, for being located in a region with environmental and climatic characteristics that differ from those that characterize the Eastern Cordillera, where the closest previous record is located (Cundinamarca, ICN- 9521).

The analysis of the stomach contents of *S. toxophyllum* revealed the presence of two plant elements that are the first evidence of the diet of this species and confirm its frugivorous status. However, the limited number of samples analyzed restrain a reliable determination of its feeding habits, but nonetheless contributes information to the diet of this species by filling information gaps, at least for the BST biome.

The modelled distribution of *S. toxophyllum* comprises the northern Chocó-Magdalena, western Orinoco, and northern Guayana biogeographical provinces. The analysis of the available information suggests that this distribution and the average probability of occurrence of this species in the north-Andean biogeographical province (particularly in the central Cordillera) is established by the ecological association between the Chocó region and the valleys of the Magdalena and Cauca rivers, being a zone where the biological elements of these areas are exchanged with Cis-Andean elements and those from the Magdalena valley, using the Orinoco foothills as a biological corridor, crossing the Burbúa depression, and reaching the Catatumbo basin (Hernández-Camacho *et al.* 1992). The species studied may occur along the Magdalena dry areas and the Orinoco and Guayana provinces.

The model does not predict the presence of *S. toxophyllum* in the Macizo de la Sierra Nevada de Santa Marta, despite the fact that Cuervo-Díaz *et al.* (1986) report its presence in this locality. It is worth noting that many of the biological elements in this area derive from lowlands of Andean origin, and move with relative ease toward the Serranía del Perijá, since a considerable number of common plant and animal species have been established in both of these formations (Hernández-Camacho *et al.* 1992).

Likewise, the model does not predict the presence of this species in the Amazon biogeographical province; however, Rodriguez-Posada and Cárdenas-González (2012) and Montenegro and Romero (1999) have reported its presence in the Departments of Amazonas and Caquetá, this being the southernmost distribution known in Colombia. Also, the model does not predict the presence of *S. toxophyllum* in the northern area of the Department of Tolima, a record reported herein.

The model of potential distribution of *S. toxophyllum* for Colombia carried out in this study contrasts with the model proposed for this same species by Angulo *et al.* (2008), who reported that part of the Department of La Guajira and the north-Andean biogeographical province and the totality of the Orinoco, Guayana and Amazon biogeographical provinces are highly suitable for the presence of the species. It should be stressed that the potential model proposed by Angulo *et al.* (2008) was designed for all South America. However, an aspect to bear in mind is that different methods lead to different results. Furthermore. the distribution model presented here takes into account only the relationship with climate and does not consider the presence of geographic barriers, ecological interactions or habitat requirements, all of them aspects that might also influence the distribution of the species.

The application of distribution modeling techniques showed that *S. toxophyllum* probably has a continuous potential geographical range from the northern Chocó-Magdalena to the western Orinoco provinces. Temperature was determined to be the climate variable with the greatest influence on the occurrence of this species, a fact that supports conservation strategies not only for these areas, but also for the biotic elements that govern temperature changes.

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Record of American black bear (*Ursus americanus*) in Durango, Mexico

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In Mexico, the American black bear (*Ursus americanus*) has withstood a number of anthropogenic pressures such as habitat loss, illegal poaching, poor legislation regarding conservation, management and exploitation, etc. Consequently, these have been the major causes of the decline of large carnivorous mammals in Mexico. The new technologies currently available have provided tools that facilitate their study, documenting their presence and distribution, in addition to expanding the biological and ecological information of this group of predators. During the 2009 winter season, intensive field surveys were conducted in southeastern Chihuahua and western Durango. In each area surveyed, 40 fixed monitoring stations were set, fitted with a camera trap and an scent station, distributed at distances ≥ 1 km between stations across the study areas. A total sampling effort of 2,400 days/trap resulted in one record of a juvenile American black bear (*Ursus americanus*; Figure 1) at Ejido "Lobos y Pescaderos", in the municipality of Tepehuanes, Durango, in Sierra Madre Occidental, Mexico (Figure 2). This record was obtained at 2436 masl, representing the second reliable report of the presence of the American black bear at these altitudes. The photographic record currently available underscores the need to implement and intensify long-term surveys that contribute to determine the presence of American black bear in the northwestern states of Mexico, specifically in Sierra Madre Occidental in the state of Durango, in sites already identified as having temperate forests habitat conditions that are potentially suitable for the American black bear.

En México, los osos negros americanos (*Ursus americanus*) se han enfrentado a diversas presiones antropogénicas como la pérdida de hábitat, cacería ilegal, falta de legislación hacia la conservación, manejo y aprovechamiento, etc., siendo estas las principales causas de disminución de los mamíferos carnívoros de gran talla en México. En la actualidad las nuevas tecnológicas han permitido contar con herramientas que faciliten su estudio, documentar su presencia y distribución, además de incrementar la información biológica y ecológica de este grupo de depredadores. Durante la temporada invernal de 2009 se realizaron prospecciones intensivas de campo en el sureste de Chihuahua y noroeste de Durango. En cada zona de prospección se colocaron 40 estaciones de monitoreo fijas, dotadas de una cámara-trampa y una estación olfativa, distribuidas a distancias ≥ a 1 km entre estaciones en las áreas de estudio. Con un esfuerzo de muestreo total de 2400 días/trampa, se obtuvo el registro de un oso negro americano (*Ursus americanus*) juvenil (Figura 1), en el Ejido "Lobos y Pescaderos", en el municipio de Tepehuanes, Durango, en la Sierra Madre Occidental de México (Figura 2), el registro se obtuvo a una altitud de 2436 msnm, lo que representa el segundo reporte fidedigno de la presencia de oso negro americano a estas altitudes. El presente registro fotográfico hace patente la necesidad de implementar e intensificar prospecciones a largo plazo que contribuyan a conocer la presencia de oso negro americano en los estados del noroeste de México, específicamente en la Sierra Madre Occidental del estado de Durango, sobre todo en aquellas entidades en las que se han identificado las condiciones de hábitat adecuadas de bosques templados y que favorezcan la presencia del oso negro americano.

Key words: black bear; carnivores; Durango; photographic record; México; survey.

Introduction

The American black bear (*Ursus americanus*, Pallas, 1780) is the largest carnivorous mammal in Mexican territory. There is incomplete information regarding its geographical distribution in Mexico, partly because this species has disappeared from many areas of its original range (Deflín-Alfonso *et al.* 2011; Juárez-Casillas and Varas 2013; Scheick and McCown, 2014; Monroy-Vilchis *et al.* 2016). It should be noted that ecological information is even more scarce for black bear populations inhabiting Sierra Madre Occidental, which, according to Hall (1981), belong to two subspecies: *U. a. machetes* and *U. a. amblyceps*.

In Mexico, black bear have withstood a number of anthropogenic pressures, the most significant being the decline of wild populations by the end of the decade of 1950s as a result of campaigns aiming to the extermination of carnivores promoted by the Federal Government (Villa 1960; Baker and Greer 1962). Other factors made the situation worse, such as hunting, habitat loss, and fragmentation caused by the promotion of livestock and agriculture from 1950 across the country (Leopold 1959; Moctezuma and Doan-Crider 2005). Likewise, the social perception of rural communities about large predators, sometimes coupled with the lack of environmental legislation and poor law enforcement, among others, have historically contributed to the few records and lack of robust scientific information about large carnivorous mammals, including the black bear (Delfín-Alfonso *et al.* 2012; Juárez-Casillas and Varas 2013, Servín 2013). In recent years, the incorporation of new technologies to the study of wildlife, including camera traps, genetic analysis and telemetry, has started to yield reliable data on these mammals, whose unique biological characteristics of large-scale movements and nocturnal habits, as well as low population densities, make them hard to spot, so they frequently go unnoticed in the wild (Delfín-Alonso *et al.* 2011; Juárez-Casillas and Varas 2013; Monroy-

Vilchis *et al.* 2016; Camargo-Aguilera *et al.* 2017).

Recently, historical and current records on the presence of black bear in Mexico have been reviewed (Deflín-Alfonso *et al.* 2011; Juárez-Casillas and Varas 2013; Monroy-Vilchis *et al.* 2016). These reviews found seven records of black bear for the State of Durango, distributed in four municipalities. One record corresponds to the municipality of Canatlan, evidenced by a skin in 1989; four records, to the municipality of Mezquital, through the recovery of one skull and one partial skeleton in 1956 (MSU 817), one stool sample and two cubs captured in 1957; a direct sighting in 2005; one specimen was recorded in the municipality of Ocampo, from a skull in 1903; and, finally, one record was obtained from the municipality of Tepehuanes through direct sighting in 2004 (Baker and Greer 1962; Deflín-Alfonso *et al.* 2011).

Materials and Methods

During the winter (22 December to 20 March) of 2009, intensive field surveys were conducted in Sierra Madre Occidental ranging from southwestern Chihuahua to northwestern Durango, in regions covered by well-preserved temperate forests, through camera traps (Wildview® Xtreme4 model). These were distributed at a distance of approximately one kilometer between stations across the study areas, along old roads used for timber extraction, bridle paths, and trails. Each area surveyed included 40 fixed monitoring stations equipped with one camera trap (placed at a height of approximately 50 cm, depending on the characteristics of the land) plus an scent station; bait consisted of sardines in tomato sauce, vanilla extract, and apple. Cameras were operating 24-hours a day for 30 days, capturing 3 images per motion detection event. Sampling effort was calculated by multiplying total number of monitoring stations by total number of sampling days (Chavez *et al.* 2013).

Results

A total sampling effort of 2400 days/trap resulted in one record of an American black bear (*Ursus americanus*) standing upright on its two hind legs; based on body size, it corresponds to a juvenile individual. The animal was photographed at 08:40 hrs on 2 February 2009 (Figure 1), at 25° 09′ 45.14″ N and 105° 57′ 15.90″ W, in pasture land of Ejido "Lobos y Pescaderos", municipality of Tepehuanes, Durango (Figure 2). The record was obtained at 2,436 meters above sea level, representing the second reliable report of the presence of American black bear at these altitudes in the Western Sierra Madre, as the altitudinal distribution of this species ranges between 1,015 and 2,809 masl (Deflín-Alfonso *et al.* 2011). The full identification of the American black bear individual involved an exchange of opinions of specialists, where six observers determined that the images correspond to a black bear.

Based on this record, and given that the specimen photographed is a juvenile, is could not be determined whether this individual belongs to an established population inhabiting the municipality of Tepehuanes or whether it is a juvenile individual in a dispersal process. The same camera trap station also recorded the presence of other wildlife species, such as coyote (*Canis latrans*), white-tailed deer (*Odocoileus virginianus*), rabbit (*Sylvilagus* spp.), hare (*Lepus californicus*), and even creole cattle. There were no further records of *U. americanus* over the course of the survey.

Figura 1. Individuo de oso negro americano (*Ursus americanus*) fotografiado en los terrenos de agostadero del Ejido "Lobos y Pescaderos", en el Municipio de Tepehuanes, Durango, México

The area where the record was obtained is characterized by pine and pine-oak forest, at an altitude of 2,436 masl. The local climate is temperate, semi-cold with cool and long summer, Cb (w2), with summer rainfall; precipitation ranges between 873 and 1,200 mm. The maximum temperatures recorded in the area vary between 24 °C and 34 °C, and the minimum from -8 °C to -16 °C (INEGI, 2016; Medina *et al.* 2005). The local topography is mountainous, with areas of gullies to the northwest of the ejido, with elevations ranging from 1,900 to 2,800 masl. in mountainous zones (INEGI, 2016). The study area comprises a broad extension of perennial and intermittent runoff, the latter primarily during the rainy season.

Discussion

Except for two records, one involving direct sighting in 2004 in Sierra de la Candela, municipality of Tepehuanes, Durango, at 2,809 meters a.s.l. in a pine forest (pers. comm., Jorge I. Servin in Deflín-Alfonso *et al.* 2011), and one capture in 2014 in the town of Felipe Carrillo Puerto, municipality of Guadalupe Victoria, where it was determined that this individual was dispersing from Sierra Madre Oriental (Camargo-Aguilera *et al.* 2017), no other recent study had formally reported the presence of American black bear in the State of Durango.

In Mexico, the historical range of the species covered forested regions of northwestern and northeastern States (Leopold 1959; Baker and Greer 1962; Hall 1981). It is currently acknowledged that not all areas within its historical distribution range in Mexico has been properly explored, so that there is a geographic gap regarding its current dis-

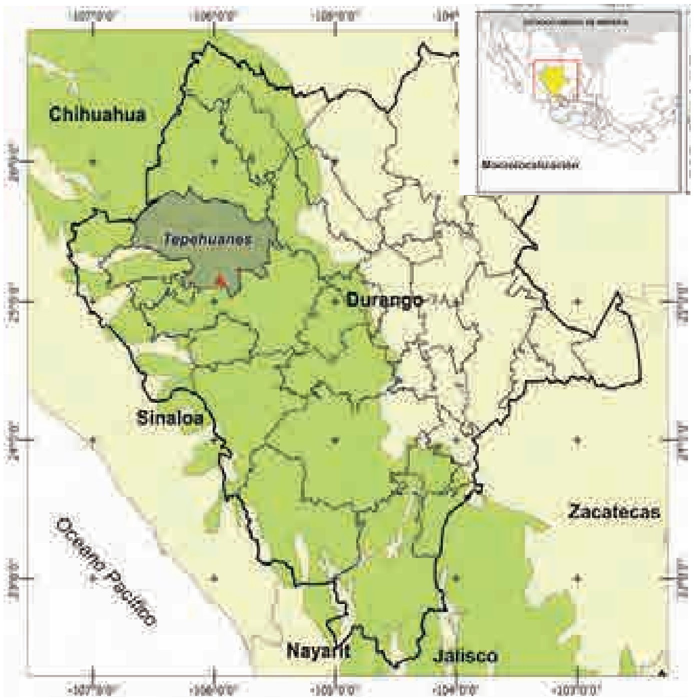

Figura 2. Mapa mostrando el área de estudio en donde se efectuaron las prospecciones en el año 2009, en el Ejido de Lobos y Pescaderos, municipio de Tepehuanes y donde se obtuvo el registro de Oso negro americano (*Ursus americanus*). Ubicación de la fotocolecta (triangulo rojo). Municipio de Tepehuanes (achurado en gris). División del estado (línea oscura). Sierra madre Occidental (achurado verde).

tribution, mainly in the States of Durango, Aguascalientes and Zacatecas. These gaps, together with insufficient field work aimed at the monitoring of the species, have led to consider *U. americanus* as locally absent or extirpated from these regions (Delfín-Alonso *et al.* 2011; Monroy-Vilchis *et al.* 2016).

The present photographic record have determined its presence and underscores the need to implement and intensify field surveys spanning at least one full year, or better still, develop mid- and long-term projects contributing to monitor the presence and even the habitat use and activity patterns of the American black bear in the northwestern regions of Mexico, particularly in the State of Durango on Sierra Madre Occidental. These surveys should be conducted in areas that have been identified as having suitable habitat conditions that favor the presence of this bear as in the municipalities of Guanacevi, Tepehuanes, Topia, Tamazula, Otaez, Santiago Papasquiaro, Durango, Canelas, Pueblo Nuevo, Mezquital and Suchil, just to mention a few.

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Records of two species of felines in Oaxaca, México

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The State of Oaxaca is home to six felid species. There are regions, such as in the Valles Centrales, where there is no evidence of their presence. This scientific note records two feline species listed in the Norma Oficial Mexicana 059. Two species of felines were recorded in the municipalities of San José del Progreso and San Pedro Totolapan. The photographs are deposited in the Colección de Fotocolectas Biológicas at UNAM (IBUNAM-CFB), plus one specimen in the Colección Nacional de Mamíferos, UNAM. Historical records were sought in the literature and databases. The species were determined based on literature descriptions, collection vouchers, and assisted by expert opinions. The first two records refer to *Herpailurus yagouaroundi* in oak forest and tropical deciduous forest in San José del Progreso and San Pedro Totolapan. The third is *Leopardus wiedii*, in a pine forest in San José del Progreso. *Herpailurus yagouaroundi* has been recorded on the Pacific and Gulf of Mexico slopes. The specimen closest to the ones reported here is located at 63.5 km from San José del Progreso and 76 km from San Pedro Totolapan. *Leopardus wiedii* has been spotted to the north, south and east of the state. The record that is closest to the one of San José del Progreso is located 48 km to the north. These records contribute to the knowledge of the Valles Centrales of Oaxaca.

En el estado de Oaxaca se distribuyen seis especies de felinos. Existen regiones como en los Valles Centrales, en las que no se cuenta con evidencia de su presencia. Esta nota científica registra dos especies de felinos incluidos en la Norma Oficial Mexicana 059. Se registran dos especies de felinos en los municipios de San José del Progreso y San Pedro Totolapan. Las fotografías se encuentran en la Colección de Fotocolectas Biológicas, UNAM (IBUNAM-CFB) y un ejemplar en la Colección Nacional de Mamíferos, UNAM. Se buscaron registros históricos en bibliografía y bases de datos. Para la determinación de las especies se revisó bibliografía, ejemplares de colección y se contó con la opinión de expertos. Los dos primeros registros son de *Herpailurus yagouaroundi* en bosque de encino y selva baja caducifolia en San José del Progreso y San Pedro Totolapan. El tercero es un *Leopardus wiedii*, en bosque de pino en San José del Progreso. *Herpailurus yagouaroundi* se ha registrado en las vertientes del Pacífico y del Golfo de México. El más cercano a los reportados aquí se ubica a 63.5 km de San José del Progreso y 76 km de San Pedro Totolapan. *Leopardus wiedii* ha sido registrado al norte, sur y este del estado. El más cercano al de San José del Progreso se encuentra a 48 km al norte. Estos registros contribuyen al conocimiento de los Valles Centrales de Oaxaca.

Key words: dry forest; *Herpailurus yagouaroundi; Leopardus wiedii*; oak forest; pine forest; Valles Centrales of Oaxaca.

Introduction

Mexico has documented the presence of six species of felines (Villa and Cervantes 2003), all with records in the State of Oaxaca (Briones-Salas and Sánchez-Cordero, 2004; Santos-Moreno 2014; Briones-Salas *et al.* 2016). However, there are regions within the State where there is no documented evidence of their presence. One example regards the districts of Ocotlán de Morelos and Tlacolula, both in the region of the Valles Centrales. Ocotlán de Morelos, despite its location just 30 km south of the city of Oaxaca and with an area of 857.9 km^2, is characterized by a remarkable lack of information on wild mammals. Thus, from a collection of 17,670 georeferenced and confirmed records of wild mammals of Oaxaca deposited in scientific collections, only four corresponded to the whole District. The Tlacolula District spreads across 3,324.14 km^2, with records for 33 mammal species (Monroy-Garcia 2009).

This note contributes to the knowledge of the geographical range of felines in the State of Oaxaca, specifically in the region of the Valles Centrales. This is crucial, as the two species recorded, namely the onza or jaguarundi (*Herpailurus yagouaroundi*) and the margay (*Leopardus wiedii*; Álvarez-Castañeda and Gonzalez-Ruiz 2018) are listed in the Norma Oficial Mexicana 059 (SEMARNAT 2010), under the category of Threatened and Endangered species, respectively, and in Appendix II of the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES; www.cites.org).

Materials and Methods

H. yagouaroundi and *L. wiedii* were recorded through photographs and one specimen preserved as skin and skeleton. Photographs were provided by private individuals and are deposited in the Colección de Fotocolectas Biológicas at Universidad Nacional Autónoma de México (UNAM; IBUNAM-CFB). The jaguarundi was donated by private individuals and was deposited in the Colección Nacional de Mamíferos (CNMA) at Instituto de Biología, UNAM. Besides, the VertNet (www.vertnet.org/index.html) and CONABIO (www.gob.mx/Conabio) databases were reviewed, as well as the monograph of mammals of Oaxaca (Goodwin 1969) and recent publications, in search of records of both species for the State (Briones-Salas and Sánchez-Cordero, 2004; Santos-Moreno 2014; Briones-Salas *et al.* 2016). The determination of the specimen and the identification of photographs were based on Hall (1981), Aranda (2012); also, identifications were supported by comparison with reference specimens deposited at CNMA, and by expert opinions.

Results

The first record corresponds to one jaguarundi found on 11 May 2013 at 18:00 hrs at "Latajo", 3.7 km south of La Garzona (16.64° N, -96.64° W, 2,362 masl), municipality of San José del Progreso (IBUNAM-CFB-38860). The local vegetation is oak forest (Figure 1a). The second jaguarundi specimen is a female, which was found dead on 3 December 2015, 1.6 km NE to El Chacal (16.69° N, -96.12° W, 1,141 masl), municipality of San Pedro Totolapan. The local vegetation is tropical deciduous forest. The somatic measures of the specimen, in millimeters, are: total length 675; vertebral tail 295, right hind leg 114, and ear 40; its weight was 1,000 g (CNMA 47966). This species displays two color phases, red and gray (Reid 1997); both specimens recorded were gray in color. The third record corresponds to a margay male, found on 24 March 2016 at 15:00 hrs at "El Hueco de la Hiesca", 4.7 km SSE of La Garzona (16.63° N, -96.62° W, 2,153 masl), municipality of San José del Progreso (IBUNAM-CFB-38861). The local vegetation is pine forest (Figure 1b).

Discussion and Conclusions

The jaguarundi is considered as a common species in Oaxaca (Goodwin 1969). However, only 12 individuals have been recorded on the Pacific and Gulf of Mexico slopes. The most recent records have expanded its distribution range to the northwest of the State (Briones-Salas and Sánchez-Cordero, 2004; CONABIO 2010a; Briones-Salas *et al.* 2016) and to the Tehuacán-Cuicatlán Biosphere Reserve (Botello *et al.* 2013). The record closest to those described in this work is located to the south, in San Miguel Suchixtepec (CONABIO 2010a), at 63.5 km from San José del Progreso and 76 km from San Pedro Totolapan (Figure 2).

Two subspecies of margay have been historically recorded in Oaxaca: *L. wiedii oaxacensis*, which inhabits the upper parts of the center of the state, and *L. w. yucatanica*,

Figure 1. Photographs of the specimens registered in the Municipality of San José del Progreso, Oaxaca, Mexico, a) onza or jaguarundi (*Herpailurus yagouaroundi*). b) margay (*Leopardus wiedii*).

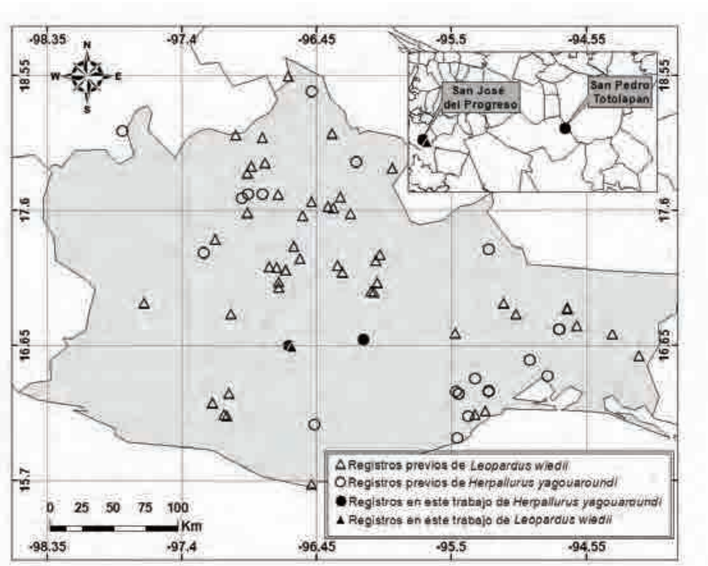

Figure 2. Records of onza or jaguarundi (*Herpailurus yagouaroundi*) and margay (*Leopardus wiedii*) in Oaxaca, Mexico

distributed toward the Isthmus of Tehuantepec in the arid tropical region (Goodwin 1969). Subsequent works have documented their presence to the north of the State, in temperate humid and semiwarm climates: mountain cloud forests, pine forests, and oak forests (Cinta 2007; Perez 2008; CONABIO 2010b; Cinta-Magallon *et al.* 2012), and in the coastal region (Meraz *et al.* 2010). The margay in tropical deciduous forest in Oaxaca was first recorded in the Tehuacán-Cuicatlán Biosphere Reserve (Botello *et al.* 2006).

Only five records belong to the center of the state of Oaxaca, located in the districts Centro (Goodwin 1969; CONABIO 2010b), Etla (CONABIO 2010b), and Mixe, where the presence of margay was recorded from footprints in a pine forest (Lavariega *et al.* 2012). The record closest to the one of San José del Progreso, district of Ocotlán de Morelos, is located 48 km to the north, in the municipality of Oaxaca de Juárez (Figure 2; CONABIO 2010b).

The information reported here contributes to the knowledge of the current geographical distribution of two species that are key for conservation, in two districts that have been relegated in mammal research and which now could be essential for understanding the displacement of species. These data provide baseline information for planning biological corridors, which are required given the accelerated changes of land use across large areas of Mexico in general and of the State of Oaxaca in particular, forcing wildlife individuals to search for new habitats to thrive.

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Biological field stations and scientific knowledge: the case of mammals in forests of the Chiapas highlands, México

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The San José Biological Station (SJBE) is the first temperate station in the Chiapas Highlands dedicated to preserve the last wildlife refuges in the area. Comprehensive studies in biological stations are a top priority to generate scientific knowledge, as well as to manage, maintain and conserve both the species and the ecological systems to which they belong. Field sampling was carried out in 2007, 2010, 2015, and 2016 at SJBE. Rodents were captured with Sherman traps; shrews, with Pitfall traps; and bats, with mist nets. Medium-sized and large mammals were recorded by direct observation. The species recorded were listed by their common name in Spanish and Tzotzil. In addition, a bibliographic search was conducted and the databases of the Global Biodiversity Information Facility and the Mammal Collection at El Colegio de la Frontera Sur were used. The conservation status of each species was identified based on the red list of the International Union for the Conservation of Nature (IUCN) and the Mexican Official Norm 059 (NOM-059) by SEMARNAT. The list of mammals recorded at SJBE includes 23 species in 8 orders and 13 families. Of the total number of species, two are endemic to Chiapas, *Peromyscus zarhynchus* and *Cryptotis griseoventris*. In the IUCN, *Cryptotis griseoventris* is listed as endangered (with few records for 62 years), and *Peromyscus zarhynchus* as vulnerable. In NOM-059, *Reithrodontomys microdon* is listed as threatened and *P. zarhynchus* as subject to special protection. Oak-pine forests at SJBE are essential for the maintenance of biodiversity in the region, offer resources and preserve the local native fauna. The creation of spaces for research on the biota is a key tool to understand the impacts and threats that currently affect forests in the Chiapas Highlands.

La Estación Biológica San José (EBSJ) es la primera estación de clima templado en los Altos de Chiapas con un enfoque dedicado a la conservación de los últimos refugios de vida silvestre en la zona. Los estudios integrales en estaciones biológicas son de prioridad para generar conocimiento científico, así como manejar, mantener y conservar las especies y los sistemas ecológicos a los que pertenecen. Los muestreos en campo en la EBSJ se realizaron en 2007, 2010, 2015 y 2016. La captura de roedores se realizó con trampas Sherman, de musarañas con trampas Pitfall y de murciélagos con redes de niebla. Mediante observación directa se registraron mamíferos medianos y grandes. Las especies registradas fueron listadas por su nombre común en español y Tzotzil. Además, se realizó búsqueda bibliográfica y se utilizaron las bases de datos de Global Biodiversity Information Facility y la Colección Mastozoológica de El Colegio de la Frontera Sur. El estado de conservación de cada especie se identificó con base en la lista roja de la Unión Internacional para la Conservación de la Naturaleza (IUCN) y la Norma Oficial Mexicana 059 (NOM-059) de la SEMARNAT. El listado de los mamíferos de la EBSJ contiene 23 especies comprendidas en 8 órdenes y 13 familias. Del total de especies, dos son endémicas a Chiapas, *Peromyscus zarhynchus* y *Cryptotis griseoventris*. En la IUCN *Cryptotis griseoventris* está en la categoría de peligro (con pocos registros desde hace 62 años) y *Peromyscus zarhynchus* como vulnerable. En la NOM-059, *Reithrodontomys microdon* está amenazada y *P. zarhynchus* sujeta a protección especial. Los bosques de encino-pino son indispensables para el mantenimiento de la biodiversidad de la región, ofrecen disponibilidad de recursos y conservan la fauna nativa. La creación de espacios para la investigación de la fauna y flora se convierten en una herramienta clave para entender los impactos y amenazas que actualmente afectan a los bosques de Los Altos de Chiapas.

Key words: conservation; mammals; oak-pine forest; San José Biological Station.

Introduction

Protected natural areas include biological field stations, whose objectives are to conduct scientific research and preserve nature (Langholz and Lassoie 2001). Examples of biological field stations in Mexico are Los Tuxtlas in Veracruz and Chamela in Jalisco, both managed by Universidad Nacional Autónoma de México (UNAM), as well as Piedra Herrada at Michilia, Durango, managed by Instituto de Ecología, A. C. (INECOL). In Chiapas, Natura and Ecosistemas Mexicanos, A. C., manage the Chajul and Tzendales biological field stations, dedicated to the study and conservation of the Lacandon forest. The research conducted in biological field stations is highly relevant for the knowledge and conservation of both the local species and the ecological systems they belong to. However, biological field stations involve complex issues, such as land tenure conflicts, lack of monitoring and management plans, and scarce information on the biotic resources in them (Smith and Ruiz-Cedillo 2009).

Although Mexico is a country where tropical and semiarid climates prevail, it should be noted that a large proportion of its territory is covered by temperate forests such as the pine-oak forest (Rzedowski 1978). This vegetation type is home to a large number of plant and animal species, many of which are currently threatened largely due to human disturbance (Flores-Villela and Geréz 1994; Toledo and Ordóñez 1998).

The San Jose Biological Field Station (EBSJ), managed by Secretariat of Environment and Natural History (SEMANH) of the State of Chiapas, was declared as such on 29 January 2015. It is the first temperate biological field station in the Chiapas Highlands dedicated to the conservation of the last wildlife refuges in the area. A number of biological studies have been carried out in this station, mainly reporting lists of bird (González-Ortega and Pérez-Suasnavar 2007) and amphibian (Aranda-Coello *et al.* 2018) species. Given the limited information on biodiversity in the Chiapas Highlands and in order to encourage research at national and international levels to advance the scientific knowledge about the region, the objective of this investigation is to communicate the importance of oak-pine forests and their associated mammalians in a highly relevant site such as EBSJ and its importance for conservation, in order to develop management and protection strategies.

Materials and Methods

Study Area. The San Jose Biological Field Station (EBSJ) is located southwest of San Cristóbal de Las Casas, a city located in the municipality of the same name in the State of Chiapas, at coordinates 16.723333 °N and -92.698333 °W (Figure 1). It is located in a mountainous area at altitudes between 2,350 and 2,380 m, stretching across 16 hectares of oak-pine- forest (Aranda-Coello *et al.* 2018), within the ecoregion of temperate sierras of Mexico (Arriaga *et al.* 2000). EBSJ receives approximately 12,200 visitors from various municipalities across the Chiapas Highlands region.

Field Work. Rodents were monitored and recorded using 100 Sherman traps baited with oats and vanilla essence, and arranged in various linear transects covering a total area of six hectares; traps were separated from each other by 10 meters. In the case of shrews, 50 Pitfall traps were placed in the same area. For bats, three mist nets (12 x 2 m in length) were placed, which remained open for six hours every night from 18:00 hours; nets were reviewed at 20-minute intervals. Thirty samplings were carried out, with a duration of five days each, from March to November 2007, January to December 2010, April to July 2015, and January 2016. Individuals captured were identified using specialized keys (Álvarez-Castañeda *et al.* 2015). Species of medium-sized and large mammals (cingulates, carnivores, lagomorphs and artiodactyla) were recorded through direct observation by the authors, local inhabitants (who provided the names in Tzotzil), and EBSJ staff. The common names of the species recorded were obtained from Álvarez-Castañeda and Gonzalez-Ruiz (2018).

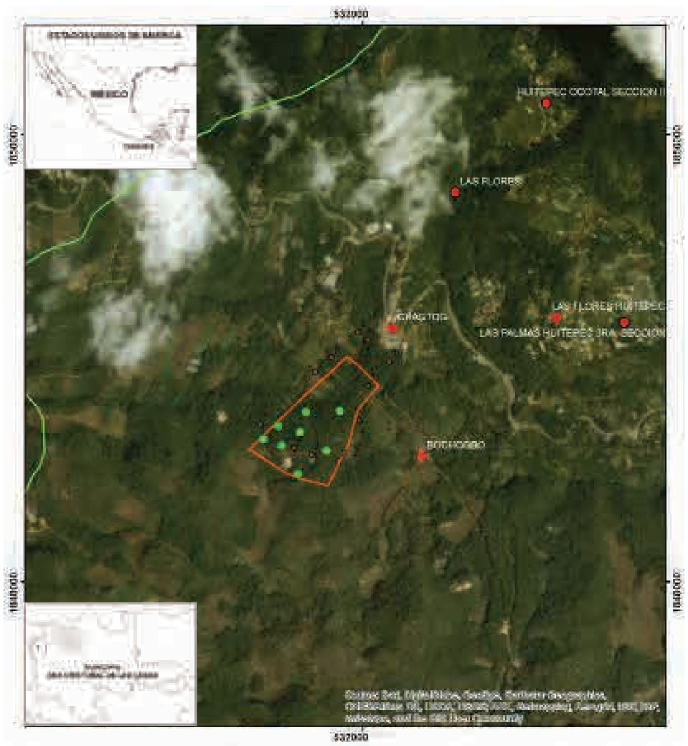

Figure 1. Geographic location of mammal sampling sites (green dots) in San Jose Biological Field Station (EBSJ), San Cristobal de Las Casas, Chiapas, Mexico. Human settlements near this station are shown (red dots).

The records of species spotted in the study area were confirmed and extended using the databases of Global Biodiversity Information Facility (GBIF 2013; www.gbif.org) and the Collection of Mammals at El Colegio de la Frontera Sur in San Cristóbal de Las Casas, Chiapas (ECO-SC-M). The species endemic to the state of Chiapas were identified, as well as the conservation and protection status of each species, based on the red list of the International Union for the Conservation of Nature (IUCN 2017) and the Mexican Official Norm 059 (NOM-059; SEMARNAT 2010). Also, we used the taxonomy proposed by Lorenzo *et al.* (2017) and Spradling *et al.* (2016).

Results

A total of 23 species were recorded, comprising 8 orders and 13 families (Table 1). The orders with the highest number of species were Rodentia (7), Chiroptera (5), and Carnivora (4). Of the total number of species, two are endemic to Chiapas: the Chiapan deer mouse, *Peromyscus zarhynchus,* and the Guatemalan broad-clawed shrew, *Cryptotis griseoventris*. With regard to the conservation status, both endemic species are listed by the IUCN (2017): *Cryptotis griseoventris* as endangered, and *P. zarhynchus* as vulnerable. In NOM-059 (SEMARNAT 2010), *Reithrodontomys microdon* is listed as threatened and *P. zarhynchus* is listed as subject to special protection.

Discussion

The Chiapas Highlands, where EBSJ is located, is the subprovince of Chiapas with the greatest amount of non-endemic (*n* = 157) and endemic (*n* = 3) mammal species (Lorenzo *et al.* 2017); therefore, it constitutes an important biodiversity sanctuary in the region (March and Flamenco 1996). The total number of records obtained in EBSJ represents 57.5% of the species reported for the municipality of San Cristóbal de Las Casas (*n* = 40 spp.) and 10.9% of those for the state of Chiapas (*n* = 211 spp.), which illustrates the importance of EBSJ in terms of species diversity and richness (Lorenzo *et al.* 2017; Porter *et al.* 2017). In particular, the oak-pine forests located in this station are important from the mammal ecology perspective (Horváth and Sarmiento-Aguilar 2001; García-Méndez *et al.* 2014), since they offer plentiful resources including food, shelter and breeding

Table 1. List of wild mammal species reported for the San Jose Biological Field Station, San Cristobal de Las Casas, Chiapas, Mexico.

	Species	Common Name in Spanish	Name Tzotzil	NOM-059	IUCN	Record
ORDER DIDELPHIMORPHA						
Family Didelphidae	*Didelphis marsupialis*	tlacuache sureño	uch	NC	LC	C
	Didelphis virginiana	tlacuache norteño	uch	NC	LC	OD
ORDER CINGULATA						
Family Dasypodidae	*Dasypus novemcinctus*	armadillo de nueve bandas	mail-chon	NC	LC	OD
ORDER SORICOMORPHA						
Family Soricidae	*Cryptotis griseoventris*	musaraña tropical de Guatemala	ch'o	NC	EN	C
	Sorex salvini	musaraña del sureste	ch'o	NC	LC	OD
ORDER CHIROPTERA						
Family Phyllostomidae	*Desmodus rotundus*	vampiro común	sotst	NC	LC	BE
	Anoura geoffroyi	murciélago sin cola gris	sotz'	NC	LC	BE
	Sturnira hondurensis	murciélago de charreteras mayor	totz	NC	LC	C
Familia Vespertilionidae	*Myotis elegans*	murciélago ratón elegante	sotz'	NC	LC	C
	Lasiurus intermedius	murciélago cola peluda amarillo del norte	sotz'	NC	LC	C
ORDER LAGOMORPHA						
Family Leporidae	*Sylvilagus floridanus*	conejo del este	ttul	NC	LC	OD
ORDER RODENTIA						
Family Sciuridae	*Sciurus aureogaster*	ardilla de vientre rojo	Chuch	NC	LC	OD
Family Geomyidae	*Heterogeomys hispidus*	tuza gigante tropical	Ba	NC	LC	OD
Family Cricetidae	*Peromyscus aztecus*	ratón azteca	choo	NC	LC	C
	Peromyscus levipes	ratón de patas ágiles	choo	NC	LC	C
	Peromyscus zarhynchus	ratón de Chiapas	choo	Pr	VU	C
	Reithrodontomys microdon	ratón cosechero dientes pequeños	choo	A	LC	C
	Reithrodontomys sumichrasti	ratón cosechero de montaña	choo	NC	LC	C
ORDEN CARNÍVORA						
Familia Canidae	*Urocyon cinereoargenteus*	zorra gris	vet	NC	LC	OD
Familia Mephitidae	*Mephitis macroura*	zorrillo rayado sureño	pay	NC	LC	GBIF
	Conepatus leuconotus	zorrillo espalda blanca	pay	NC	LC	BE
Family Mustelidae	*Mustela frenata*	comadreja	j´cuch baca	NC	LC	GBIF
ORDER ARTIODACTYLA						
Family Cervidae	*Odocoileus virginianus*	venado cola blanca	tetikal chij	NC	LC	OD

Mexican Official Norm 059 (NOM-059): A = threatened, Pr = subject to special protection, NC = not classified. International Union for Conservation of Nature (IUCN): LC = least concern, VU = vulnerable, EN = endangered. Record: C = in-field catch, OD = direct observation, BE = ECO-SC-M database, GBIF = Global Biodiversity Information Facility database.

sites. The mammal species favored by this type of habitat in EBSJ include those with restricted and fragmented distribution due to extensive deforestation in other areas within their distribution range, such as *C. griseoventris,* a species with only 11 known individuals since it was first collected by F. L. Burnet in 1956 (Guevara *et al.* 2014a), *P. zarhynchus* and *R. microdon* (IUCN 2017; Guevara *et al.* 2014b; Lorenzo *et al.* 2017). The mammals listed in the risk category by NOM-059 require immediate actions for conservation, such as the development of a management program to preserve both the habitat and the species living in it, and the creation of a regional network for the elaboration of strategies aiming at the protection of their habitat. Currently, there are few areas in the Chiapas Highlands region covered by oak-pine forests in a good state of conservation, which are limited to small scattered patches (Horváth and Sarmiento-Aguilar 2001).

On the other hand, the loss and fragmentation of natural ecosystems in Chiapas facilitate the movement of opportunistic mammal species (*e. g.,* rodents, domestic cats and dogs) that, in addition to competing for the same space and food resources with native mammals, are predators of native fauna and potential carriers of diseases, both for wildlife and humans (Naranjo *et al.* 2016). The above indicates that these are potential threats in EBSJ, since these opportunistic animals have been observed given their proximity to Tzotzil communities, such as Nachig, San Felipe, Zacualpa and Navenchauc.

Therefore, we believe that the creation of natural spaces is a key tool for conducting research on the local flora and fauna, and also to understand the responses of wild mammal populations to the impacts and threats currently affecting forests in the Chiapas Highlands. Thus, biological and ecological studies should be encouraged in oak-pine forests in the region, including regular evaluations of the fauna; particularly in EBSJ, it is essential to conduct short-, medium- and long-term monitoring of mammals to determine the state of their populations. Specialized sampling with aerial and camera traps will allow to elaborate a more comprehensive list of those species that are potential inhabitants in the area but are rarely captured at ground level (*e. g.,* the rodents *Habromys lophurus*, *Nyctomys sumichrasti* and *Glaucomys volans*). Furthermore, the influence of nearby human settlements on forest structure and composition and on mammal populations should be assessed, including the introduction of exotic species (*i.e*., dogs, cats, rats and mice), as well as an assessment of the convenience of creating biological corridors connecting with other protected natural areas, and the development of programs in partnership between municipal governments and society to implement urgent actions for management and conservation of the local habitats, plants and animals. EBSJ currently conducts environmental education campaigns in schools within the region as well as in the field station itself, where we use *P. zarhynchus* as a symbol of conservation of the area, and undertake research projects on the ecology and distribution of the red-lipped arboreal aligator lizard (*Abronia lythrochila*, Reptilia: Squamata) and the unspotted saw-whet owl (*Aegolius ridgwayi*, Birds: Strigiformes).

The conservation and proper management of the oak-pine forest in EBSJ are appropriate strategies to promote the survival of most mammal species (Naranjo *et al.* 2016; Lorenzo *et al*. 2017). For this reason, this note sets the grounds for developing scientific knowledge and continue with the biological studies, as EBSJ is home to species of great ecological and conservation importance.

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