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Source: Air, Soil and Water Research, 14(1)

Published By: SAGE Publishing

URL: https://doi.org/10.1177/1178622120988721

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Air, Soil and Water Research
Volume 14: 1–9

DOI: 10.1177/1178622120988721
SAGE

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ABSTRACT: Roads and highways are 1 of the most significant obstacles affecting wildlife movement by fragmenting habitat, altering wildlife migration and use of habitat, while also being a danger to wildlife and humans caused by wildlife-vehicle interactions. To mitigate wildlife mortality on highway sections and to minimize death and injury to motorists as well, road ecologists have proposed structures adapted for the safe passage of wildlife across roads. In this study, photographic sampling was conducted using trail cameras to quantify wildlife activity and use of existing culverts, bridges, and drainages within 2 separate sections of Mexico Federal Highway 2 where previous field assessment had observed high levels of activity. These sections are important areas for the conservation of wildlife, and they are known to be biological corridors for rare species of concern such as jaguar, black bear, and ocelot. The trail cameras were operated for 1 year to document the annual cycle of wildlife movement through the area. With the photographs obtained, a database was created containing the information from each wildlife-culvert interaction. Prior to sampling, an inventory of existing culverts was conducted that measured height, width, volume, and surrounding habitat to assign a hypothesized use quality index. After testing for significant differences in use index among culverts, we recognized that all culverts were equally important for moving wildlife, and that there were no significant differences in the use of culverts by the quality index.

KEYWORDS: Wildlife crossing, encounter rate, road ecology, use index, Sky Islands

RECEIVED: May 31, 2020. **ACCEPTED:** December 29, 2020.

TYPE: Case studies of a grassroots binational restoration collaborative in the Madrean Archipelago Ecoregion (2014-2019) – Original Research

FUNDING: The author(s) disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: Financial support for the research, authorship, and/or publication of this article came from the United States Fish and Wildlife Service.

DECLARATION OF CONFLICTING INTERESTS: The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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Introduction

With the current accelerated urbanization of our world, habitat fragmentation has become the threat with the greatest impact on[1] biodiversity.[2] Habitat connectivity across the landscape is important to ensure the persistence of wildlife populations.[3,4] In a landscape altered by humans, barriers are created that fragment natural habitats, generating serious ecological consequences such as the loss of biological diversity. One of the main causes of habitat fragmentation and loss of connectivity worldwide is caused by the development of road infrastructure.[5,6] Roads hinder the movement of animals; decrease access to resources and shelters; reduce reproductive success, gene flow, and recolonization[7,8]; and are recognized as the infrastructure that has the greatest lasting impact on wildlife.[6,9]

Currently, road networks are ubiquitous, and there is an increase in the number of vehicles each year. In the United States, for example, there are about 6 million km of paved roads and an estimated 1 to 2 million large animal wildlife vehicle collisions a year resulting in hundreds of human fatalities.[10] Mexico, by comparison, has approximately 322 859 km of paved roads. By mid-century, it is estimated that there will be 25 million km of paved roads globally.[11-13] The impacts of roads on wildlife are not only caused by collision with vehicles. Noise, vibration, dust, lighting from vehicle headlights, and human presence and incursion of roads into wild landscapes have been linked to reduced wildlife abundance.[14,15]

In landscapes dominated by human activity, connectivity between habitats can be maintained through biological corridors. Landscape connectivity is essential for the movement of animals between patches of habitat to maintain genetic heterozygosity, and also to assure viable populations in the event of resource depletion and/or stochastic events such as fire or drought.[16,17]

In Mexico, the road network is the most used transportation system for connecting people, products, and services in the country. However, there are few studies on the relationship between organisms and the country's highway system, but it is presumed to be an important cause of mortality for wildlife.[18,19]

Historically, transportation planners in Mexico have been primarily concerned with the economic and structural aspects of road network design, while neglecting the ecological implications of roads.[20] To assess the impacts of road construction on wildlife, it is important to perform detailed studies on the activity of wildlife near existing highways and proposed expansions.

Road ecology in Mexico is a recent field of scientific research. Currently, there are a limited number of published studies (17); most have focused on practical aspects from environmental impact studies and 2 studies are peer-reviewed scientific research articles addressing the effects of roads on biodiversity and a review of works on the subject.[19,21] The first studies on the impact of roads on wildlife were performed by

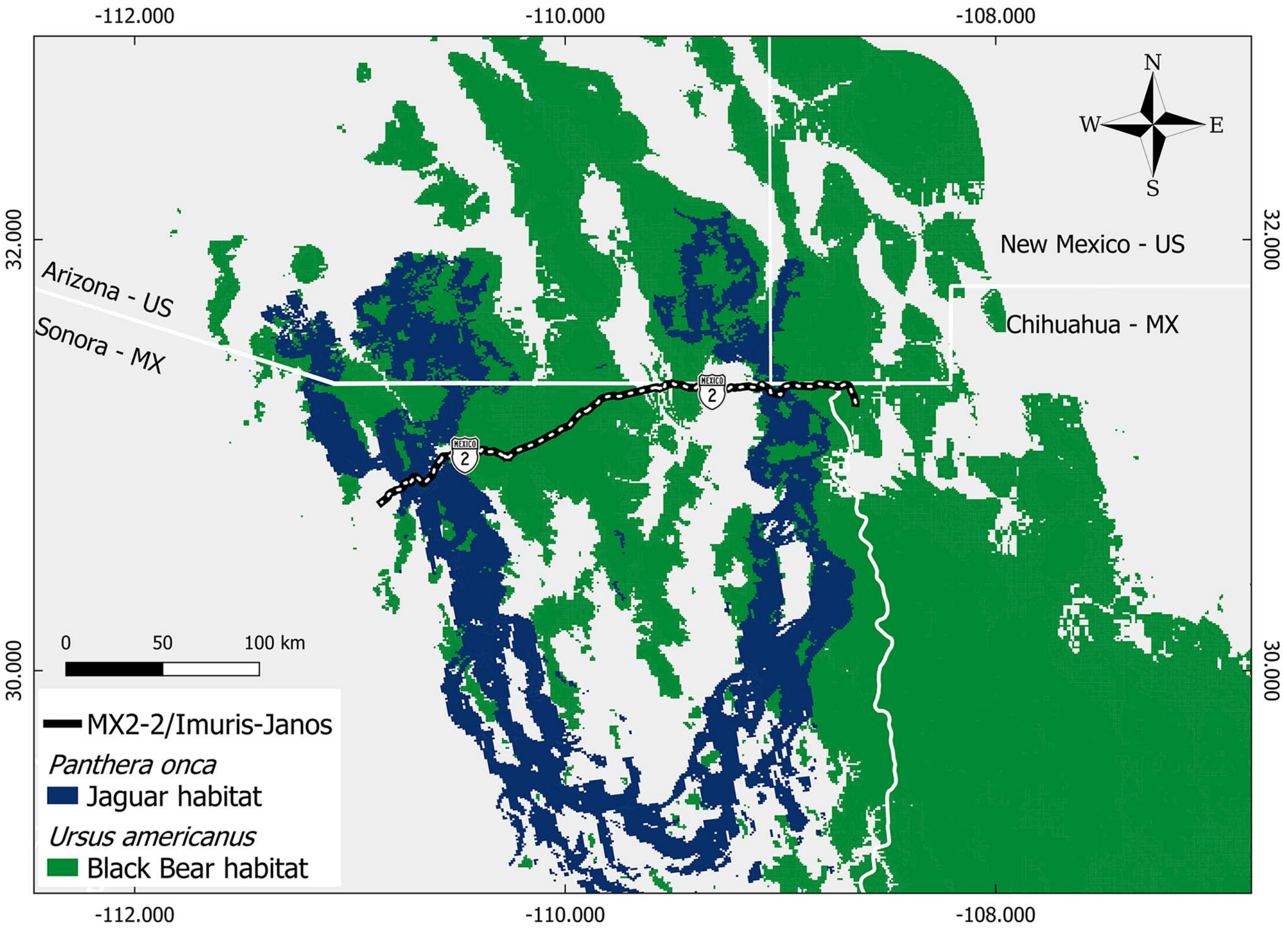

Figure 1. Habitat fragmentation by Federal Highway 2, Imuris- Chihuahua State Limits. Habitat models of two key species in the ky Islands, 1) jaguar habitat in blue and 2) blackbear habitat in green.

Polaco and Guzmán. In the arid zones of northern Mexico, there are only 3 studies: in the Pinacate Biosphere Reserve, the state of Nuevo León, and the Chihuahuan Desert.[22,23] Considering the high biodiversity of the country, there are relatively few wildlife crossings. Currently, throughout the road network of Mexico, there are various proposals for wildlife crossings, as well as for the modification and maintenance of road culvert structures adapted for the passage of wildlife. Furthermore, 2 recent legislative reforms, 1 that requires the creation of wildlife crossings on highways in the state of Chihuahua, and another that requires their inclusion in Regional Territorial Ordinance Plans in the state of Sonora, mark a new standard in the country's environmental legislation. Owing to the limited information that we currently have in the country, appropriate management is difficult to advocate for. This reveals the importance of conducting studies in this field, especially the need to make the information public through peer-reviewed studies in scientific journals.

The objective of this study is to justify the development of mechanisms and strategies to redesign and adapt the road network in Mexico and consider including wildlife crossing structures in future highway projects. We measured wildlife use of existing culverts, tunnels, and bridges in 2 highway sections that bisect portions of high conservation value in the Sky Island region of northwest Sonora, Mexico, over the period of 1 year. With these data, we analyzed each monitored culvert in relation to its importance for wildlife through a usage index.

Materials and Methods

Study area

The study area is located in northern Sonora, Mexico, ranging from the Sierra Azul 15 km east of Ímuris, to the border of Chihuahua in the Sierra San Luis, which is generally considered to be the northernmost mountain range of the Sierra Madre Occidental (Figure 1). This region is known as the Madrean Sky Island Archipelago, a series of about 55 mountain ranges that occur in the borderlands region of the Mexican states of Sonora and Chihuahua and the US states of Arizona and New Mexico. The Madrean Sky Island Archipelago is also exceptional because it lies in a zone of convergence of many biomes, the Sonoran Desert, Chihuahuan Desert, Rocky Mountains, Sierra Madre Occidental, plains grassland, and Neotropical thornscrub. It is the meeting ground of the North American tropical and temperate zones, harboring plants and animals

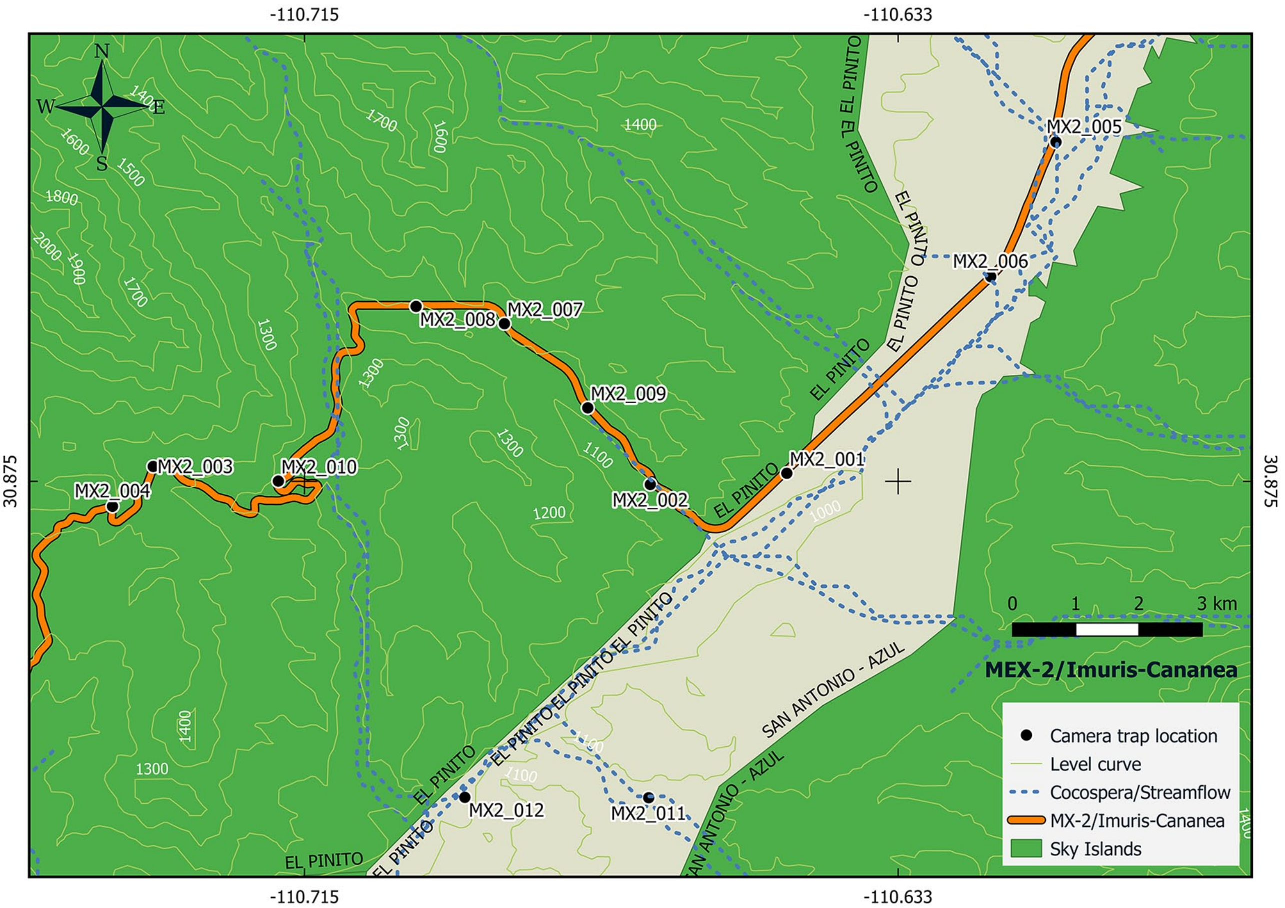

Figure 2. Wildlife camera location, Federal Highway 2, Ímuris – Cananea (IMCA).

from Neotropical and Nearctic influences. It is the land where the black bear and the jaguar share the same trail.[24,25]

This diversity of biotic communities leads to a wide variety of animal species. For example, Neotropical animal species, such as the jaguar, ocelot, and coati, reach their northernmost distribution in this region.[26] Of note, 4 wild felid species have been recorded in the study area: jaguar (*Panthera onca*), mountain lion (*Puma concolor*), ocelot (*Leopardus pardalis*), and bobcat (*Lynx rufus*). Madrean evergreen woodland supports Coues white-tailed deer (*O virginianus couesi*) and white-nosed coati (*Nasua narica*), while pronghorn (*Antilocapra americana mexicana*) and the white-sided jackrabbit (*Lepus callotis gaillardi*) occupy mostly shrub-free semi-desert and plains grassland.

The first stretch of highway, Ímuris-Cananea (IMCA), is located near the municipality of Ímuris; it covers approximately 28 km of highway that comprises 146 to 118 km in an east-west direction (Figure 2). This section contains the mountain complexes El Pinito to the northwest of the highway, San Antonio-Azul to the southeast of it, and La Madera-Cucurpe to the south. The second road section, Agua Prieta-Chihuahua State Limits (APCH), is located near the municipality of Agua Prieta. It covers approximately 6 km of highway that comprises 93 to 109 km in an east-west direction (Figure 3). This section is located in the southern Animas Valley known in Mexico as "El Valle," and bounded by the Peloncillo, Pan Duro, and Sierra San Luis mountain ranges. It is an exceptional expanse of plains grassland dominated by buffalograss (*Buchloe dactyloides*) and blue grama (*Bouteloua gracilis*) that shares a strong floral and faunal affinity to the northern Great Plains.

Camera trap placement

Wildlife cameras were used to record the use of culverts by local wildlife from September 2018 to October 2019. Cameras were placed along the IMCA and APCH highway sections (Figures 2 and 3). The 21 culverts selected were those that we deemed more likely to be used by wildlife. In a previous study, all existing culverts were recorded and measured along 243 km of Federal Highway 2, corresponding to the entire stretch of highway from Ímuris to the state line of Sonora and Chihuahua.[27] In addition to measuring size dimensions, each culvert was assigned to a category (high, moderate, and low) for its potential use by wildlife. This category describes the quality of the surrounding habitat in a radius of 50 m around the culvert. The potential presence of wildlife at each location was determined by sightings, tracks, scat, scrapes, and other spoor. Other variables that could influence wildlife, such as the presence of human settlements, clearing of natural habitat, and

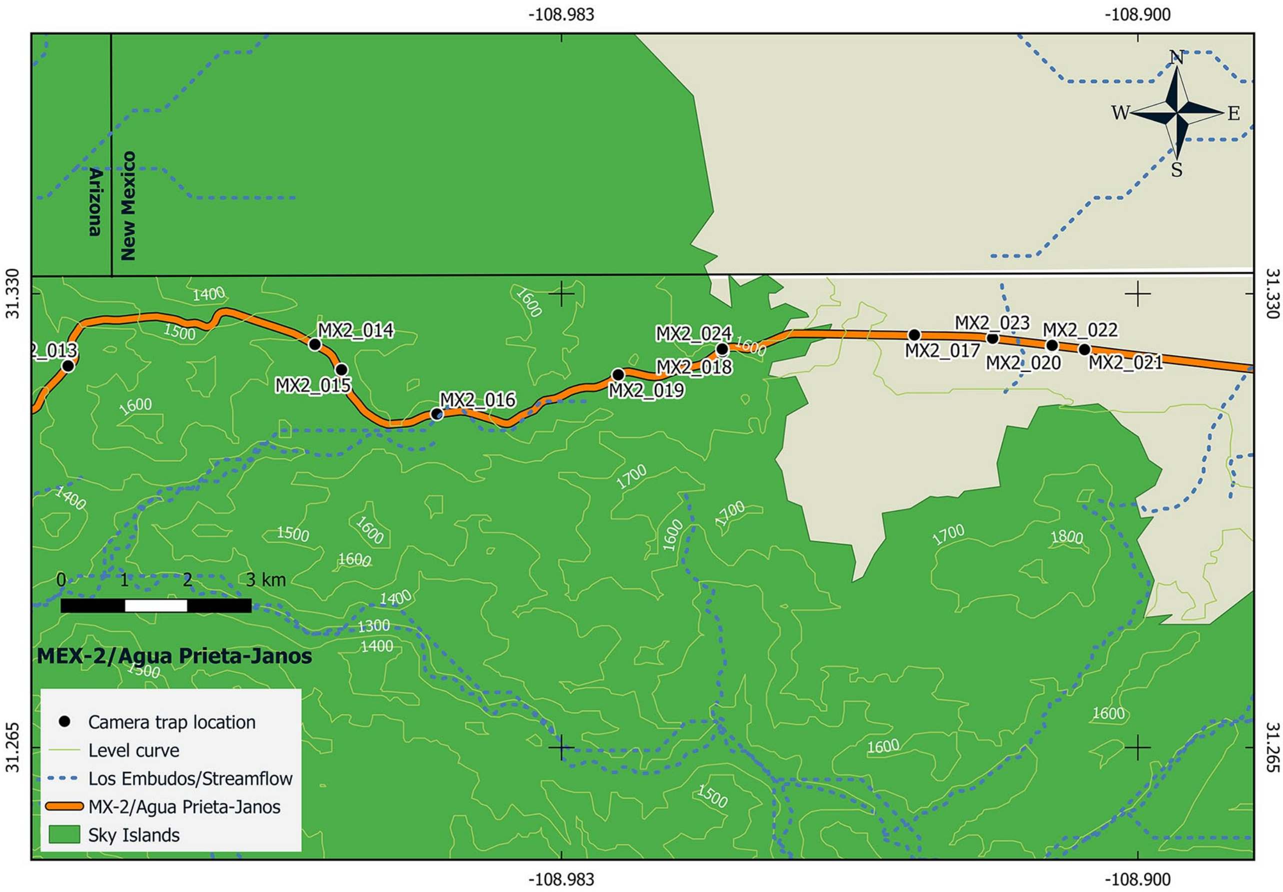

Figure 3. Wildlife camera location, Federal Highway 2, Agua Prieta - Chihuahua State Limits (APCH).

degree of soil disturbance, were also noted. This helped to assign each culvert a category of potential use by wildlife. Based on these categorical values of potential use, culverts with the highest potential values for wildlife use were selected for inclusion in the study for installation of wildlife cameras.

We used the wildlife camera protocol described by Chávez et al. 2013 as a guidance to install the cameras.[28] We used Browning model BTC-6HD-940 cameras. They were programmed using a standardized protocol previously established by Wildlands Network and EcoGrande personnel. From the 21 selected culverts, we were able to place only 18 cameras in highway culverts in the 2 sections: 8 cameras were placed in the IMCA section and 10 were placed in the APCH section (Figures 2 and 3). The cameras were placed either overlooking culverts or bridges or inside these structures, to directly document their use by wildlife (Table 1).

In addition, to maximize the possibility of photographing medium to large wildlife within potential culverts, the sampling sites were chosen based on 2 criteria: the potential for use described in the culvert selection procedure above and the location of culverts within habitat probability models of key species such as the jaguar and the black bear (Figure 1).[29,30] In addition, for the IMCA section, 2 cameras were placed within Rancho el Aribabi, at a distance of 5 km perpendicular from the highway, to document species that are present in the study area. In addition, the presence of any wildlife tracks and scat inside and within 25 m of the culvert structure was documented photographically. However, this article only reports on the results obtained by the wildlife cameras because a methodology was not established to perform a standardized sampling of field observations in each culvert structure. The IMCA section was monitored by EcoGrande personnel, and the APCH section was monitored by Sky Island Alliance with assistance from Cuenca Los Ojos.

Data analysis

For each photo, we documented the species, number of individuals, date, and time.

To demonstrate the diversity of species that directly interacted with the highway, we estimated the percentage of encounters by species and by section where the number of camera triggers was normalized to a percentage to make them comparable between sites, removing the effect of the total number of encounters.

The encounter rate is a standardized measure of the direct interaction between wildlife and the highway. Each photo represents an event where an animal activated the camera sensor. Because the cameras differ in their operation time interval, we adjusted the number of encounters by the number of days they were in operation

Table 1. Location of wildlife cameras by culvert.

WILDLIFE CAMERA	CULVERT	LOCATION		STRETCH
		N	W	
MX2_001	D094	30° 52' 34.0"	–110° 38' 52.7"	IMCA
MX2_002	NA	30° 52' 28.4"	–110° 40' 01.0"	IMCA
MX2_003	D034	30° 52' 37.4"	–110° 44' 09.7"	IMCA
MX2_004	D030	30° 52' 17.5"	–110° 44' 30.1"	IMCA
MX2_005	D116	30° 55' 19.6"	–110° 36' 38.0"	IMCA
MX2_006	D109	30° 54' 12.3"	–110° 37' 10.7"	IMCA
MX2_007	D071	30° 53' 48.8"	–110° 41' 14.0"	IMCA
MX2_008	D063	30° 53' 57.4"	–110° 41' 58.2"	IMCA
MX2_009	D079	30° 53' 06.7"	–110° 40' 32.3"	IMCA
MX2_010	D050	30° 52' 30.1"	–110° 43' 07.1"	IMCA
MX2_011	NA	30° 49' 51.8"	–110° 40' 01.8"	IMCA
MX2_012	NA	30° 49 52.0"	–110° 41' 33.8"	IMCA
MX2_013	D611	31° 19' 10.8"	–109° 03' 11.3"	APCH
MX2_014	D617	31° 19' 21.9"	–109° 01' 04.1"	APCH
MX2_015	D621	31° 19' 08.8"	–109° 00' 50.4"	APCH
MX2_016	D628	31° 18' 45.99"	–109° 00' 01.2"	APCH
MX2_017	D654	31° 19' 26.76"	–108° 55' 55.18"	APCH
MX2_018	D648	31° 19' 18.21"	–108° 57' 34.05"	APCH
MX2_019	D640	31° 19' 06.2"	–108° 58' 27.8"	APCH
MX2_020	D660	31° 19' 20.46"	–108° 54' 44.33"	APCH
MX2_021	D661	31° 19' 19.22"	–108° 54' 27.5"	APCH
MX2_022	D658	31° 19' 21.4"	–108° 54' 44.19"	APCH
MX2_023	D657	31° 19' 25.09"	–108° 55' 14.9"	APCH
MX2_024	D648	31° 19' 19.49"	–108° 57' 34.2"	APCH

Abbreviations: APCH, Agua Prieta-Chihuahua State Limits; IMCA, Ímuris-Cananea.

Encounter rate = number of animals photographed / camera operation days

This equation was used to show the differences between species using the culverts.

The temporal pattern of encounters helps us understand the seasonality of wildlife culvert usage. The objective of this metric is to show the number of direct interactions between wildlife and cameras over the course of the entire year.

To highlight the importance of culverts in each of the highway sections studied, we adapted a usage index for the culvert structures proposed by Mata-Estacio et al.[31] The analysis of this index was performed separately for each section, and only the encounters with the most frequent species and those of greatest importance for conservation were calculated. This index allows us to compare the culvert structures studied by weighing their value according to the number of encounters by species with the equation

$$I.U. = \frac{n_{ij}}{\left(\frac{n_i}{E}\right)}$$

where "n_{ij}" is the number of encounters by species "i" in camera "j," "N_i" is the number of encounters for species "i" in all cameras, and "E" is the total number of cameras.

To detect significant differences between the usage rate with cameras, a Kruskal-Wallis test was performed where the usage rate was the response variable and the camera and species were the explanatory variables.

Results

In both sections (IMCA and APCH), a total of 1579 wildlife sightings were recorded. These are 3.69 sightings/day from September 2018 to September 2019. For the IMCA section, on 10 cameras, a total of 1405 wildlife sightings were recorded using the culvert structures over a period of 410 days. This represents an average of 3.42 sightings/day of any wildlife for the IMCA section. Considering mammals only, we have a total of 1230 mammal sightings for the IMCA section. This represents an average of 3 mammal sightings/day (Figure 4). However, the 8 cameras of the APCH section registered a total of 850 wildlife sightings over a period of 427 days of service. This represents 1.99 sightings/day, on average (Figure 4). Considering mammals only, we have a total of 380 mammal sightings. This is an average of 0.88 mammal sightings/day for APCH. Sightings in the IMCA section include the following mammal species: javelina (*Pecari tajacu*), gray fox (*Urocyon cinereoargenteus*), Coues white-tailed deer (*O virginianus couesi*), white-nosed coati (*N narica*), raccoon (*Procyon lotor*), hooded skunk (*Mephitis macroura*), coyote (*Canis latrans*), and ringtail (*Bassariscus astutus*) (Figure 5, "Aribabi").

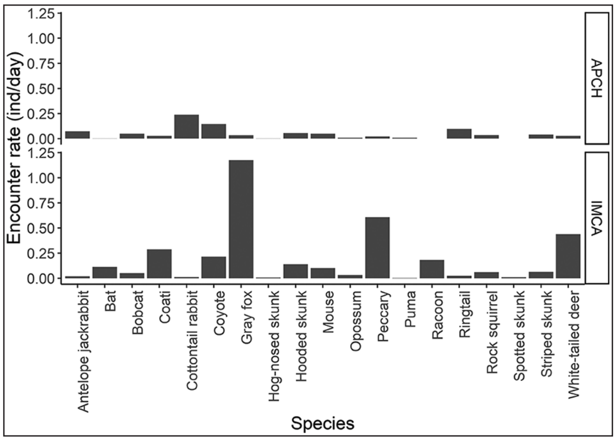

Figure 4. Species encounter rate (number of photos/day) on IMCA and APCH.

Sightings in the APCH section include the following species: ringtail (*B astutus*), coyote (*C latrans*), hooded skunk (*M macroura*), white-nosed coati (*N narica*), Coues white-tailed deer (*O virginianus couesi*), bobcat (*L rufus*), striped skunk (*Mephitis mephitis*), opossum (*Didelphis virginiana*), javelina (also known as collared peccary) (*P tajacu*), gray fox (*U cinereoargenteus*), and finally mountain lion (*P concolor*) (Figure 5, "El Valle").

In general, the observed seasonal pattern is bimodal. Increases in the use of culvert structures are observed in the months of October and May in both sections, but the effect was more pronounced in the IMCA section (Figures 6 to 8).

There were no significant differences in the usage index in either of the 2 sections (Kruskal-Wallis test, $P = .1155$ *IMCA stretch;* $P = .34$, *APCH stretch*); therefore, any of the culverts in both sections are recommended for the construction of a wildlife crossing. Although cameras 1, 4, 6, 14, 17, and 19 appear to be more important, no statistical significance was picked up during the length of the study. They are all equally important to the passage of wildlife.

Discussion

Results can be interpreted as an estimate of the magnitude of a physical interaction between wildlife and highway infrastructure. Most likely, the differences in the magnitude of the

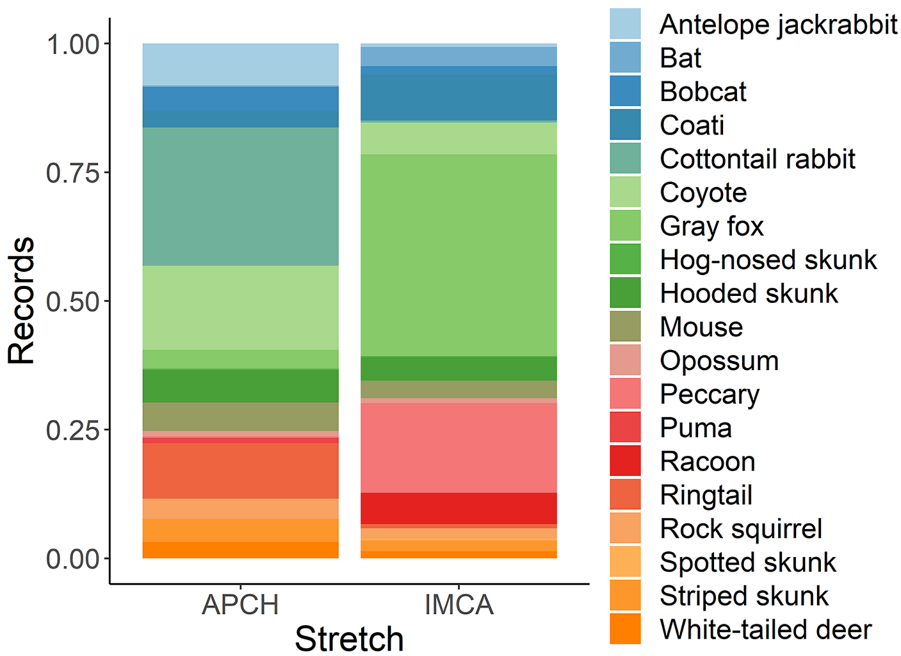

Figure 5. Relative species sightings by stretch.

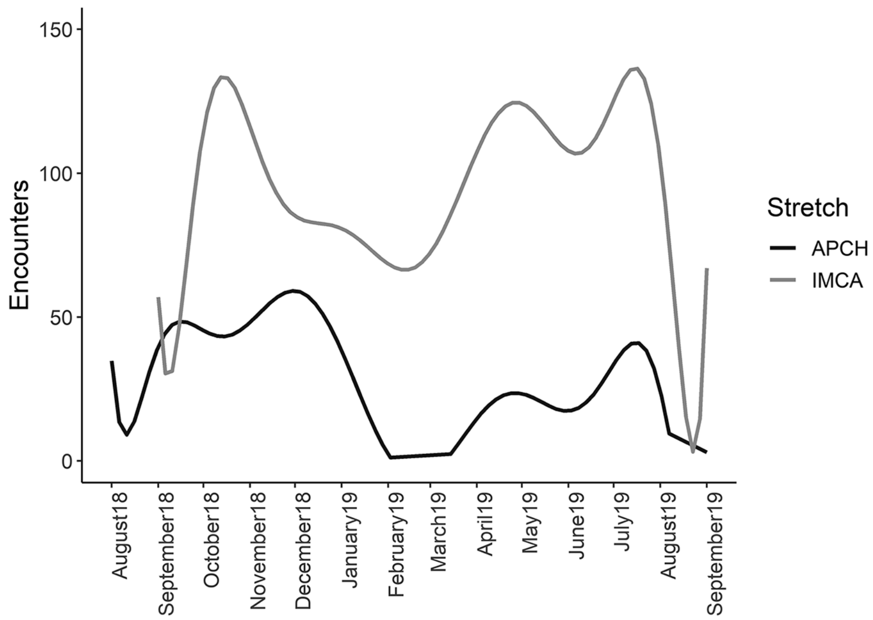

Figure 6. Wildlife occurrence from September 2018 to October 2019.

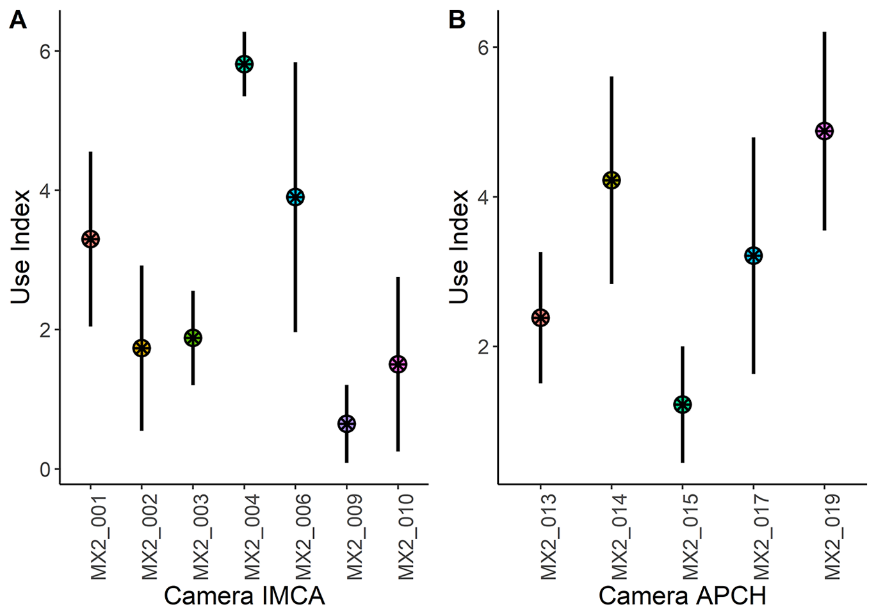

Figure 7. Usage rate by camera.

encounter rate are due to the type of habitat where they are found. Similarly, when examining the diversity of mammals by section (Figure 4), we found 19 species in the IMCA section and 17 species in the APCH section. By examining the encounter rate relative to the number of encounters (Figure 5), we can observe the most frequent species interacting with the road culverts. In the IMCA section, gray fox, white-tailed deer, and javelina are the species that dominate this wildlife-structure interaction. In the APCH section, the most represented species are rabbits, coyote, and coati. Wildlife that crosses these sections of Federal Highway MX2 tend to occur seasonally in a bimodal manner. This bimodality is apparently due to the lack of movement of animals during the winter.

The usage index is a variable of importance that was estimated by species for each of the cameras in the 2 sections separately. Its value was weighted by the frequency and the quantity in which the species occur at each camera, and in the sum of all the cameras in a section (N_i / E in equation 1). That is, the value for each species is limited by the number of animals of that species at that culvert. In general, the most abundant species tend to have less weight per capita. There were no significant differences in the rate of use in the culverts of both sections. This means that all the drains studied are equally important for the passage of wildlife.

A similar study in northwest Spain analyzed the effectiveness of lower and upper structures used by wildlife using an

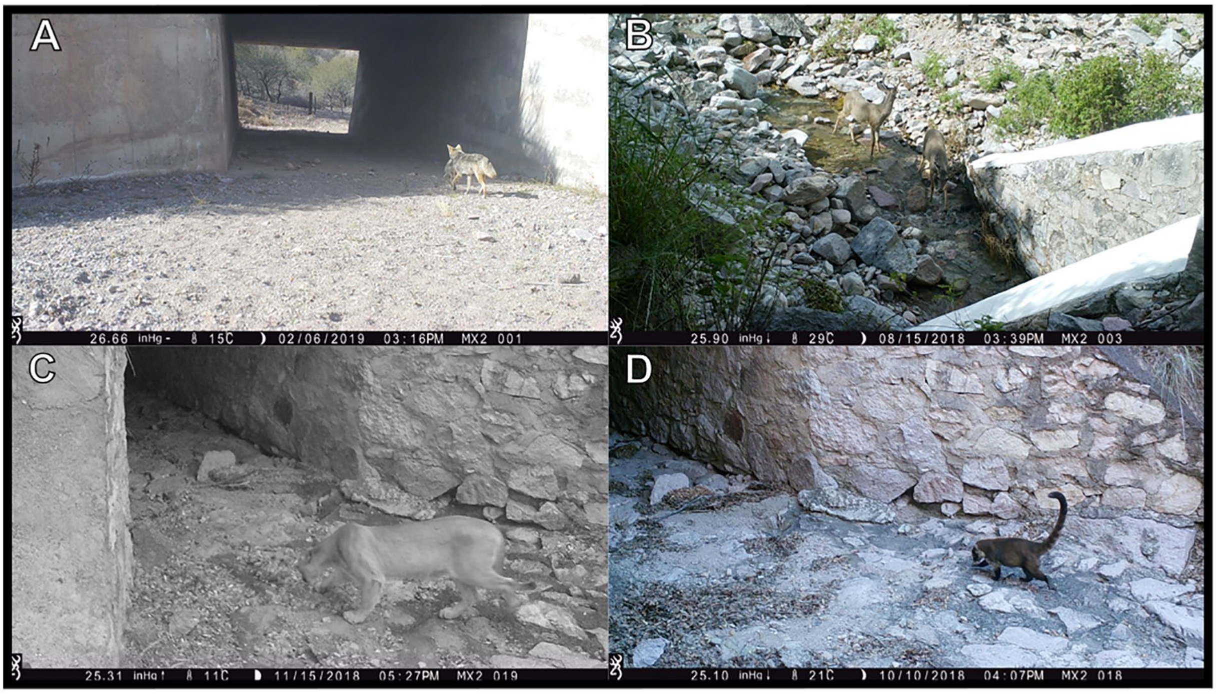

Figure 8. A) Coyote, IMCA, B) White-tailed deer, IMCA, C) Mountail lion, APCH, D) Coati, APCH.

index that is weighted by species density and structure type.[31] In our study, we controlled for the effect of culvert type by choosing to install camera traps in the culverts with more potential of usage by wildlife and therefore eliminating variability between culvert types. Highway MX2 does not have any type of wildlife use-friendly structures, and therefore, variation among culverts is due only to culvert clearance. We modified the usage index used by Mata et al 2006, and our index only considers species density per stretch. The characteristics of this study are similar to ours in the way that biological corridors intersect and their movement routes are affected from moving north-south and vice versa. Despite the fact that the culverts on Highway MX2 are not adapted as wildlife crossings, the species using them are morphologically similar in size to European species[31]: small to medium in size, and larger species such as the white-tailed deer not using underpasses. Because our study area does not have adapted drains or specific wildlife crossings, we cannot directly compare the results regarding structure type. Comparing usage index among culverts regarding wildlife use-only, we did not find significant differences in usage index among culverts in any of the 2 stretches.

To reduce the negative impact caused by Federal Highway MX2 on wildlife, we recommend installing wildlife crossings and modifying existing culvert structures to increase wildlife connectivity. Wildlife funnel fences, regular clearing of vegetation and debris that might obstruct visibility, and exit ramps are some of the recommended improvements.[32] We also recommended establishing a long-term wildlife camera monitoring program monitoring passage rates in these sections where we have collected baseline data. We suggest including a monitoring protocol documenting wildlife use by track and sign, as well as multiyear roadkill surveys along the IMCA-APCH stretch.[33]

Finally, it is important to consider safe wildlife crossings in the planning stages of new highway projects, which will minimize cost by avoiding ad hoc mitigation.[34] To achieve this, highway improvement needs to be addressed urgently from a legislative level in Mexico.

Conclusions

This is the first study in Northwest Mexico that analyzes the potential of existing culverts for use as wildlife crossings. The information gained from this study has helped us understand that existing structures can allow safe wildlife movement across roads. However, existing structures are not always sufficient, for in September 2018, a female black bear (*Ursus americanus*) was killed in the IMCA section, despite there being large culverts on either side of the collision site.[35] In the spring 2020, a black bear was killed on Interstate 10, 30 km east of Tucson, Arizona, above Davidson Canyon, where, too, there existed a sufficient underpass for the bear to cross beneath the highway, yet the bear crossed on an elevated interstate highway. Although there are studies suggesting that black bears frequently use large underpasses,[36] we did not obtain any bear records in the culverts surveyed. In conclusion, this study suggests that although existing culverts have the potential to function as a wildlife underpasses, there is a need to study and better understand the movement patterns and needs of individual species, such as black bears to locate specific priority sites for the implementation of appropriate mitigation structures.

Author Contributions

JCB, MT, EL, and REF-B conceived and designed the study; EL, REF-B, MM-R, and CA-M led fieldwork; EL and REF-B conducted statistical analysis; MM-R, REF-B, and EL wrote the article; and all authors contributed to editing the article and gave the final approval.

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