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Wildlife Use of Road Mitigation Structures in Relation to Their Construction, Structural Characteristics, and Environmental Factors along a South Texas Highway

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WILDLIFE USE OF ROAD MITIGATION STRUCTURES IN RELATION TO THEIR CONSTRUCTION, STRUCTURAL CHARACTERISTICS, AND ENVIRONMENTAL FACTORS ALONG A SOUTH TEXAS HIGHWAY

A Thesis

by

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ABSTRACT

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Roads are known to negatively impact wildlife by fragmenting habitat and mortality caused by wildlife-vehicle collisions. Road mitigation structures, such as wildlife crossing structures (WCS), wildlife guards (WG), and fencing are commonly used to address the issue of roads. In South Texas, such structures were built or modified along State Highway (SH) 100 in Cameron County as an effort to conserve the endangered ocelot (*Leopardus pardalis*). Camera traps were deployed to monitor these structures as a way to assess their effectiveness in restricting wildlife entry into the roadway with fencing and WG and conveying wildlife movement across roads through WCS. By examining changes in wildlife interaction with these structures from their construction to post construction and examining the relationship between wildlife movement through WCS and their structural characteristics and environmental factors, this thesis provides baseline insight into the effectiveness of road mitigation along SH 100.

DEDICATION

I dedicate this thesis to my wonderful husband, Austin Roy, who was an endless source of support and motivation throughout this entire journey.

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CHAPTER I

INTRODUCTION

Road Ecology

Roads are ubiquitous across the United States, with a network of over 6.7 million km of public roads (FHWA 2018). While they are a crucial infrastructural element in the transportation of people and goods, they can have adverse effects on the natural world. Road density and roads that extend into rural areas affect the extent to which natural systems and processes are altered (Forman and Hersperger 1996, Forman et al. 1997). Some environmental effects of roads include alterations in hydrology, erosion, transport of pollutants, noise pollution, changes in wind and temperature patterns, and importantly, impacts on wildlife (Forman and Alexander 1998, Coffin 2007).

Roads have long been known to influence wildlife distributions, movements, and survival (Forman and Alexander 1998). Not only are roads themselves a direct contributor to habitat loss and modification, but they also fragment habitat, thus creating and isolating smaller populations by limiting dispersal (Andrews 1990, Forman and Alexander 1998, Coffin 2007). Population isolation can limit gene flow, resulting in eventual inbreeding and populations that may be more susceptible to stochasticity, disease, and catastrophic events (Gilpin and Soulé 1986, Forman and Alexander 1998). Other long-term effects of fragmentation include reduction in species richness and changes in community composition and ecosystem functions (Haddad et al. 2015).

One of the most visible consequences of roads are mortalities caused by wildlife-vehicle collisions. Wildlife road mortalities are widespread and occur in both urban and rural areas (Huijser et al. 2008). These mortalities can have direct negative effects on wildlife abundance and distribution, population size, and population genetics (Forman and Alexander 1998, Gerlach and Musolf 2000, Forman et al. 2003, Fahrig and Rytwinski 2009. These effects have a more profound and noticeable impact on rare species (Forman and Alexander 1998, Forman et al. 2003). In a report to Congress, Huijser et al. (2008) identified 21 federally listed species, ranging from large mammals such as Florida panthers (*Puma concolor coryi*), to medium-sized carnivores such as ocelots (*Leopardus pardalis albescens*), to small amphibians such as Houston toads (*Anaxyrus houstonensis*), whose populations are threatened by road mortalities.

In order to minimize mortality events and increase population connectivity, it is important to select appropriate mitigation measures to reduce the overall negative effects of roads. Such measures often include road mitigation structures, which have been developed to both restrict wildlife from entering roadways and safely facilitate their movements across roads (Forman et al. 2003, Reuer 2007, Klar et al. 2009, Clevenger and Huijser 2011). Road mitigation structures have taken many different designs to fit both their function and target species, and include exclusion fencing, overpasses, underpasses, and wildlife guards (WG; Clevenger and Huijser 2011, Sott 2012). Overpasses include large landscape bridges, which may benefit large mammals, multi-use overpasses, which are intended for use by humans and wildlife, and canopy crossings, which may benefit arboreal species (Clevenger and Huijser 2011, Sott 2012). Underpasses include culverts, bridges, or tunnels, all of which may benefit all types of species depending on the size of the underpass (Clevenger and Huijser 2011, Sott 2012). Furthermore, overpasses and underpasses often vary in design based on surrounding topography, target species, and other intended uses (Clevenger and Huijser 2011). Where gaps in exclusionary fencing exist, due to road intersections and driveways, WG serve as a possible solution for road mitigation much in the same way that cattle guards limit livestock to a particular area (Reed et al. 1974).

Monitoring the use of mitigation structures is a preliminary step in examining the effectiveness of these mitigation measures (van der Grift et al. 2013). Use of underpasses and overpasses are often monitored with the use of camera traps or track beds, though camera traps aid in reducing observer bias as tracks can be difficult to discern (Hardy et al. 2003, van der Grift and van der Ree 2015). These methods can also be applied to monitor activity at WG and fence ends.

Road Ecology and Ocelots (*Leopardus pardalis albescens*) in South Texas

South Texas is home to two small populations of the federally endangered ocelot, found in the Lower Rio Grande Valley, that are isolated both spatially and genetically (Haines et al. 2005*a*, Janečka et al. 2008, US Fish and Wildlife Service (USFWS) 2016). Furthermore, road mortalities have been identified as a direct threat to the populations in these areas (Haines et al. 2005*a*). To address these threats to ocelots, it is important to increase connectivity between the populations and provide a way for them to cross roads safely (Haines et al. 2005*b*). In response, the USFWS Recovery Plan for the Ocelot (2016) listed the building of road mitigation structures as a recovery action to reduce the effects of human activity and loss of habitat connectivity in addition to road mortalities.

On State Highway (SH 100) in Cameron County at the southern end of the Texas ocelot range, three ocelot mortalities occurred between 2010 and 2014, following the installation of concrete traffic barriers (TxDOT 2015). In response, TxDOT constructed and modified mitigation measures from 2016 to 2018, including exclusion fencing, WG, and wildlife crossing structures (WCS; TxDOT 2015).

Road mitigation structures have been built along many major highways in the US, Canada, Portugal, and Spain, among other countries, and the effectiveness of many of these structures has been documented (Clevenger and Waltho 2000, Barnum 2003, Peterson et a. 2003, Mata et al. 2004, Reuer 2007, Grilo et al. 2008). This type of research is lacking in South Texas but is crucial in determining the potential for road mitigation structures to aid in ocelot conservation.

This thesis is a component of on-going monitoring efforts to examine the effectiveness of road mitigations structures in South Texas. By using camera traps to record wildlife interaction with WCS and WG, this thesis will evaluate changes in wildlife interactions between the during construction and post construction periods, and structural characteristics and environmental factors will be examined as well. This study is a continuation of previous graduate research by Cogan (2018) and seeks to offer further examination of road mitigation structures and their effects on wildlife along SH 100.

Thesis Organization

Chapter I: Introduction

This chapter discusses background information on the threats of roads to wildlife, common methods of mitigating those threats, and the particular issue of mitigating ocelot (*Leopardus pardalis albescens*) road mortalities in South Texas. Study area is also included in this chapter.

Chapter II: Changes in wildlife interaction with road mitigation structures from their construction to post construction

The objective of this chapter is to examine how wildlife use of mitigation structures changes from during construction to post construction periods.

Chapter III: The influence of structural characteristics and environmental factors on wildlife use of wildlife crossing structures

The objective of this chapter is to analyze select variables that may be having an effect on wildlife use of WCS in the post construction period.

Chapter IV: Conclusion

This chapter includes conclusions drawn from chapters II and III and a discussion on the relevance of this research.

CHAPTER II

CHANGES IN WILDLIFE INTERACTION WITH ROAD MITIGATION STRUCTURES FROM THEIR CONSTRUCTION TO POST CONSTRUCTION

Introduction

One of the fundamental ways to determine the effectiveness of road mitigation structures is to examine how wildlife interact with them over time. The general expectation would be that wildlife crossing structures (WCS), whether underpasses or overpasses, would be used by wildlife to cross roads, and fencing, wildlife guards (WG), and other exclusionary structures would discourage wildlife entrance into roadways. McCollister and van Manen (2010) and Seidler et al. (2018) documented that the construction of fencing and WCS contributed to a decrease in wildlife road mortalities in Washington County, North Carolina and western Wyoming, respectively. Additionally, Dodd et al. (2004), Braden et al. (2008), and Alonso et al. (2014) reported an increase in WCS use from pre-construction to post-construction of mitigation structures. Increases in wildlife use of WCS have led to a supposition that wildlife become more comfortable with WCS over time, demonstrating WCS effectiveness in increasing connectivity where roads disrupt habitat (Braden et al. 2008, Gagnon et al. 2011, Alonso et al. 2014). Belant et al. (1998) and Sebesta et al. (2003) found that cattle guards and deer guards with straight crossbars were effective at restricting ungulate movement, and Peterson et al. (2003) and Allen et al. (2013) found that deer and wildlife guards with crossed or bridge grating configurations were also effective for ungulates. Conversely, Allen et al. (2013) did find that WG were less effective for restricting movement of carnivore species.

Setting up a study with a before-after-control-impact (BACI) to monitor areas before and after mitigation and at sites unaffected by human impact (control) and those that are (impact) has been described as the optimal study design to measure mitigation structure effectiveness (Roedenbeck et al. 2007, van der Grift et al. 2013, van der Grift and van der Ree 2015). When a full BACI study is not possible, a before-after (BA) study may take its place (Roedenbeck et al. 2007; van der Grift et al. 2013; van der Grift and van der Ree 2015). Several studies have documented changes in wildlife use of crossing structures from pre-construction of further road mitigation to post construction (Dodd et al. 2004, Braden et al. 2008, McCollister and van Manen 2010, Alonso et al. 2014); however, few studies have examined use of mitigation structures during construction (Cramer 2012, Seidler et al. 2018).

Monitoring the number of successful crossings by wildlife is insufficient for evaluating effectiveness of crossing structures (van der Grift et al. 2013, van der Grift and van der Ree 2015). Use of crossing rates and performance ratios using minimum expected rate of use supplies a better comparison of the potential for wildlife to use crossing structures and the actual use of crossing structures (Clevenger and Waltho 2005, van der Grift et al. 2013, van der Grift and van der Ree 2015). Control plots are typically used with crossing use measurements to calculate the minimum expected rate of use (van der Grift and van der Ree 2015); however, when control plots are not available, crossing rates have been used to measure performance of WCS (Kintsch et al. 2017).

Newly constructed and modified WCS, WG, and exclusionary fencing along SH 100 in South Texas allow for an opportunity to examine how wildlife interactions with these structures change over time. Behaviors related to the effectiveness of the WCS that have been observed at these five sites have been classified into two major categories: crossing and refusal (Cogan 2018). Crossings occur when the WCS has successfully facilitated wildlife movement from one side of the road to the other, and refusals occur when wildlife movement does not end on the opposite side of the road (Cogan 2018). Similarly, two main behaviors observed at WG sites are crossings and repels (Cogan 2018). A repel occurs when a WG has successfully blocked access to the road, and wildlife do not cross. Crossings occur when animals are not hindered by the structure and are able to enter the roadway. Overall, crossings are the desired behavior at WCS, and repels are the desired behavior at WG (Kintsch et al. 2017, Cogan 2018). Understanding this will aid in determining their effectiveness in facilitating wildlife movement and potentially reducing wildlife road mortalities.

The objective of this chapter is to examine how wildlife use of mitigation structures changes from during construction to post construction periods. The hypotheses to be tested are as follows:

1. Wildlife crossing rates at WCS will increase post construction.
2. Species richness of wildlife using the WCS will increase post construction.
3. Repel rates of wildlife interacting with pipe WG (PWG) and bridge grating WG (BGWG) will increase post construction.
4. Species richness wildlife interacting with PWG and BGWG will increase post construction.

Study Area

This study was conducted along an 11.9 km stretch of State Highway (SH) 100 between Los Fresnos and Laguna Vista in Cameron County, Texas (Fig. 1). The road is flanked by

USFWS protected Laguna Atascosa National Wildlife Refuge (LANWR) and agricultural and residential land. The study area (Fig. 1) is located within the Rio Grande Delta physiographic zone (Leslie 2016). The surrounding habitat is characterized by Texas coastal grassland, Tamaulipan saline thornscrub, and Texas coastal high salt marsh (USGS 2011, Leslie 2016). The diversity of ecological systems and land use along SH 100 result in varying amounts of vegetative cover, from open, sea-ox-eye daisy (*Borrichia frutescens*) flats, to smooth cordgrass (*Spartina alterniflora*) fields, to thornscrub patches with canopy cover (TPWD 2014, Leslie 2016). The area receives an average annual rainfall of 66 cm, with highs in the summer reaching an average of 33° C and lows in the winter getting down to an average of 12° C (NOAA 2020).

Construction of road mitigation structures resulted in a 1.8 m tall chain link fence spanning an 11.9 km length of SH 100, on the north and south sides of the highway, along five WCS (Fig. 2) and 18 WG (Fig. 3). WCS1 and WCS2 are large concrete box culverts, each located in drainage ditches with permanent water (Table 1). WCS1 and WCS2 had previously been constructed with a wildlife crossing; a 41-cm round culvert pipe was placed above the water line to enable use (TxDOT 2015). WCS3A is a small concrete box culvert with a dirt substrate and ephemeral water presence (Table 1). WCS3A was already in existence prior to construction of the other WCS along SH 100 and was only modified during the construction period with fence replacement and cutting back vegetation. The WG are modified cattle guards, nine of which were constructed with 7.6 cm metal pipe (PWG) and the other nine constructed with metal bridge grating (BGWG; TxDOT 2015). Construction began 9 September 2016 and was completed on 10 May 2018.

Methods

Data Collection

Species activity at mitigation structures was monitored using active infra-red (AIR) and passive infra-red (PIR) triggered camera traps (Reconyx PC900 HyperFire™ and Reconyx HS2X HyperFire 2™; Reconyx LLP, Holmen, WI, USA) placed at each WCS and WG. Monitoring occurred from 10 January 2017 to 10 May 2019 at WCS and from 28 April 2017 to 10 May 2019 at WG . Each WCS had two cameras on each side of the WCS, with one camera trap facing toward the opening and one facing away (Fig. 4). One camera was equipped with both PIR and AIR triggers to capture wildlife interacting with the WCS. A second PIR-only camera trap faced away from the WCS to capture wildlife in the immediate surrounding areas that may not interact with the WCS (Fig. 4). The PIR trigger is activated within the camera by changes in radiation emitted in the camera's field of view, while AIR trigger is activated by an externally triggered infrared beam deployed in the field of view (Welbourne et al. 2016). Additionally, video cameras (Bushnell Trophy Cam HD Model 119874, Bushnell Corporation, Overland Park, KS, USA and Reconyx HyperFire 2™) were set up at each WCS entrance to supplement still photographs in wildlife-WCS interaction analysis. Additional cameras were set up at larger WCS openings to ensure that the full extent of the opening is captured and that the number of individual animals missed by the cameras is reduced.

Permission was granted by landowners to monitor 16 of the 18 WG. Each of the 16 WG were monitored with one AIR-equipped camera facing the WG toward the road side and one PIR-only camera facing the WG toward habitat (Fig. 5).

Each camera in this array was checked every two weeks to exchange memory cards for empty ones, change batteries as needed, and check for any maintenance issues.

Data Management

To process and organize all photographs and videos, a suite of software programs developed by Sanderson and Harris (2013) was used. Each picture was relabeled with its timestamp using the programs Renamer and Special Renamer. The pictures from each location were then sorted into file folders based on species, then sorted again by number of individuals observed in each picture. The program DataOrganize was used to process the sorted file folder structure to catalog all pictures. This ensured that all sorting errors were addressed prior to analysis. The last step in this process was to run DataAnalyze, which used the output from DataOrganize to produce summary statistics of the data.

Data Analysis

To determine how wildlife interacted with WCS, behaviors were categorized into four different groups: 1) "crossing," in which individuals completely cross from one end of the WCS to the other; 2) "entry" and exit on the same side, where individuals do not complete movement to the opposite end; 3) "approach" without entry; 4) "nearby," where individuals are in the vicinity of a WCS but do not interact with it. Entry, approach, and nearby were considered "refusal" behaviors. Crossing, entry, approach, and nearby were used to calculate a crossing rate for WCS using the following formula:

$$\text{crossing rate} = \frac{\sum \text{no. of crossings}}{\sum(\text{no. of crossings} + \text{no. of entry} + \text{no. of approach} + \text{no. of nearby})}$$

Crossing, entry, and approach were used to calculate a repel rate for WG using the following formula:

$$\text{repel rate} = \frac{\sum(\text{no. of entry} + \text{no. of approach})}{(\sum \text{no. of crossing} + \text{no. of entry} + \text{no. of approach})}$$

Nearby interactions were not included in WG repel rates as wildlife may have been traveling parallel to the fence without intention to enter the roadway where they are meant to be deterred. Directional movements were also recorded at WG and categorized into habitat-road and road-habitat. Only habitat-road interactions were included in repel rates because entry and approach behaviors could not be observed from the road side of WG.

Monitoring was broken into two time periods: during construction (January 2017-May 2018 at WCS; April 2017-May2018 at WG) and post construction (May 2018-May2019). When cameras were occasionally temporarily removed from sites due to flooding or needing repair, interactions with mitigation structures could not be observed. Only data from dates when at least one camera was active on both sides of a WCS were used in data analysis. Additionally, only data from dates when at least one camera was facing toward a WG were included in analysis. Only wildlife that were identifiable to species were used in analysis of WCS and WG (Table 2); birds, rodents, and unidentifiable individuals were excluded. All herpetofauna that were not of conservation concern were grouped together. Reptile species of conservation concern consist of the Texas tortoise (*Gopherus berlandieri*) and Texas indigo snake (*Drymarchon melanurus erebennus*) and were documented as individual species.

Data were binned by monthly increments. For comparing WCS use between during and post construction, only WCS1, WCS2, and WCS3A were included in analysis. Monitoring at newly constructed WCS3 and WCS4 did not begin until post construction, and thus they were not included in analysis. All means are reported as mean ± standard error. To gain a basic understanding of whether daily wildlife activity was different between during and post construction periods and between WCS, a permutational analysis of variance (PERMANOVA) was performed using daily counts of crossings, and another PERMANOVA was performed using daily counts of refusals.

To determine whether there was an increase in crossing rates between the during and post construction periods, a two-way analysis of variance (ANOVA) was conducted using program R 3.6.1 comparing monthly crossing rates to site and construction period (R Core Team 2019). Crossing rates were arranged into a matrix and imported into PRIMER v7 (Primer-e, Ltd. Plymouth Marine Laboratory, UK) for analysis. Monthly bins by locations served as samples, and species and species group served as variables. A Bray-Curtis similarity matrix was calculated in PRIMER with untransformed data and a dummy variable of one as a zero-adjustment (Clark and Gorley 2015). Then, a permutated analysis of variance (PERMANOVA) was used to compare crossing rates of wildlife communities between construction periods nested within WCS. A post-hoc pairwise PERMANOVA was used to compare crossing rates of communities between during and post construction periods for each individual WCS. A test for homogeneity of dispersion (PERMDISP) was performed in PRIMER to test dispersion of samples across WCS and construction period (Anderson et al. 2008). Using a similarly constructed matrix to that which was used for PERMANOVA, with monthly counts of crossings for each species and species group, a similarity percentages (SIMPER) analysis was performed in PRIMER to determine which species contributed most to similarities of communities crossing observed within the during and post construction periods for WCS1, WCS2, and WCS3A (Clark and Gorley 2015). Monthly species richness, defined as the count of the number of species present (Morrison et al. 2008), was calculated in PRIMER for species crossing in each construction period at each WCS. A two-way ANOVA was performed to compare species richness between WCS and construction period using R, and a post-hoc Tukey test was performed for pairwise comparisons.

To gain a basic understanding of whether daily wildlife activity was different between during and post construction periods and between PWG and BGWG, a PERMANOVA was performed using daily counts of repels, and another PERMANOVA was performed using daily counts of crossings .To determine whether there was an increase in repel rates at WG between the during and post construction periods, a Wilcoxon rank sum test was conducted comparing monthly repel rates between the two periods using R (R Core Team 2019). Repel rates were entered into a matrix in PRIMER. Species and species groups served as variables, and monthly data bins by WG type and construction period served as samples. A Bray-Curtis similarity matrix was then calculated with untransformed data and a dummy variable of one in PRIMER for subsequent analyses (Clark and Gorley 2015). Using PERMANOVA, wildlife community repel rates were compared between the during and post construction periods nested within WG type. A post-hoc pairwise PERMANOVA was then used to determine whether there was a difference in community repel rates between the during and post construction periods within each WG type. A test for homogeneity of dispersion (PERMDISP) was performed in PRIMER to test dispersion of samples across WG type and construction period (Anderson et al. 2008).Using counts of repels, a SIMPER analysis was used in PRIMER to determine the amount to which each species contributed to similarities within communities being repelled by each WG and each construction period. Monthly species richness was calculated for each WG type for each construction period in PRIMER for species that were repelled by WG. A two-way ANOVA was used to compare differences in species richness between each construction period and each WG type.

Results

Wildlife Crossing Structures

Cameras were deployed for 1,078 survey days during construction and 1,019 survey days post construction at WCS1, WCS2, and WCS3A combined (Table 3). At WCS1, WCS2, and WCS3A, 4,512 interactions were recorded during construction, and 3,369 interactions were recorded post construction (Table 3). During construction, WCS1 had a mean of 1.34±0.15 crossings and 0.81±0.07 refusals occurred per survey day; post construction, a mean of 0.54±0.05 crossings and 0.48±0.05 refusals occurred per survey day were observed (Fig. 6). At WCS2 during construction, a mean of 2.22±0.12 crossings and a mean of 2.05±0.13 refusals occurred per survey day. A mean of 1.98±0.19 crossings and a mean of 1.21±0.09 refusals per survey day post construction (Fig. 6). During construction at WCS3A, there were 3.45±0.14 crossings and 2.49±0.09 refusals occurred per survey day; a mean of 3.09±0.16 crossings and 2.46±0.09 refusals occurred per survey day post construction (Fig. 6). A PERMANOVA showed that mean daily number of crossings per survey day were significantly different between WCS1, WCS2, and WCS3A ($Pseudo\text{-}F_2$=184.6, P=0.001) and between during construction and post construction ($Pseudo\text{-}F_1$=62.48 P=0.001; Fig. 6). The interaction between site and construction period was also significant ($Pseudo\text{-}F_2$=6.05, P=0.002). A PERMANOVA showed that mean daily number of refusals per survey day were significantly different between WCS1, WCS2, and WCS3A ($Pseudo\text{-}F_2$=251.3, P=0.001) and between during construction and post construction ($Pseudo\text{-}F_1$=26.3, P=0.001). The interaction between site and construction period for mean daily refusals was significant ($Pseudo\text{-}F_2$=13.3, P=0.001).

The mean monthly crossing rate for all species at WCS1 during construction was 0.59±0.04, and the mean monthly crossing rate post construction was 0.51±0.05 (Fig.7). For all species at WCS2, the mean monthly crossing rate during construction was 0.52±0.04, and the mean monthly crossing rate was 0.49±0.07 (Fig. 7). The mean monthly crossing rate for all species at WCS3A was 0.56±0.04 during construction and 0.53±0.05 post construction (Fig. 7). A two-way ANOVA showed that there was no significant difference in crossing rate between WCS or between the during and post construction periods on a monthly basis, for all species combined (Site: F_2=0.338, P=0.714; Period: F_1=1.17, P=0.282; Fig. 7), and there was no significance found in the interaction of site and period (F_2=0.170, P=0.844). PERMANOVA results revealed that there were significant differences between the communities crossing between the three WCS (Pseudo-F_2=12.79, $P \leq 0.001$) and between construction periods nested within site (Pseudo-F_3=3.29, $P \leq 0.001$; Fig. 8). Further pairwise testing showed that the communities crossing at each individual WCS were significantly different between the two time periods (WCS1: t=1.91, P=0.010; WCS2: t=1.63, P=0.030; WCS3A: t=1.73, P=0.015; Fig. 8). In a follow-up test for PERMDISP, it was found that dispersion among samples was significantly different between sites ($F_{2,75}$=26.26, $P \leq 0.001$), but this was not true between the two construction periods ($F_{1,76}$=2.19, P=0.180). The results of the PERMANOVA were accepted, as a significant PERMDISP likely indicates a combination of position and dispersion in the data (Anderson et al. 2008).

Results from SIMPER showed that only raccoon used WCS1 to cross SH 100 during construction; raccoon, opossum and bobcat all used WCS1 post construction (Table 4). Similarly, at WCS2, only raccoon crossed during construction, and raccoon, opossum, and bobcat used WCS2 post construction (Table 4). Multiple species used WCS3A both during and post construction, and javelina began to use WCS3A to cross post construction (Table 4).

A two-way ANOVA showed that monthly species richness of wildlife crossing at WCS was significantly different between WCS (F_2=134.6, P≤0.001) and between during construction and post construction (F_1=18.2, P≤0.001; Fig. 9). There was no significant interaction between site and construction period (F_2=0.42, P=0.658). A post-hoc pairwise Tukey test showed that mean monthly species richness of wildlife crossing was significantly higher at WCS3A than WCS1 (P≤0.001) and higher at WCS3A than WCS2 (P≤0.001), but no difference was found between WCS1 and WCS2 (P=0.704; Fig. 9).

Wildlife Guards

Cameras at WG were deployed for a total of 3,465 survey days during construction and 3,918 survey days post construction at PWG and BGWG combined. For both PWG and BGWG combined, 2,570 interactions were recorded during construction, and 3,368 interactions were recorded post construction (Table 5). At PWG during construction, a mean of 2.09±0.09 repels and a mean of 0.67±0.06 crossings were observed per survey day, and post construction, 2.57±0.11 repels and 1.07±0.09 crossings occurred per survey day (Fig. 10). At BGWG during construction, 1.35±0.09 repels and 1.22±0.08 crossings occurred per survey day, and post construction, 2.28±0.11 repels and 1.92±0.12 crossings occurred per survey day (Fig. 10). A PERMANOVA showed that mean number of repels per survey day were significantly different between PWG and BGWG (*Pseudo*-F_1=34.4, P=0.001) and between during and post construction (*Pseudo*-F_1=47.1, P=0.002; Fig. 10). There was a significant interaction between WG type and construction period (*Pseudo*-F_1=10.2, P=0.002). A PERMANOVA showed that mean crossings per survey day were significantly different between PWG and BGWG (*Pseudo*-F_1=47.1, P=0.001) and between during and post construction (*Pseudo*-F_1=27.9, P=0.001; Fig.

10). There was no significant interaction between WG type and construction period (*Pseudo-*F_1=1.66, P=0.195).

A comparison of repel rates between during construction and post construction showed no significant difference (W=7958, P=0.195; Fig. 11), but there was a significant difference in repel rates between BGWG and PWG across both time periods, with greater repel rates at PWG (W=6946, P=004; Fig. 11). PERMANOVA results showed that the communities of species being repelled by WG were significantly different between WG types (Pseudo-F_1=7.60, P=0.012) and between the during and post construction periods nested within WG type (Pseudo-F_2=2.42, P=0.010; Fig. 12). Further pairwise testing showed no significant difference in communities between the during and post construction periods at BGWG (t=1.39, P=0.085); however, there was a significant difference between communities during construction versus post construction at PWG (t=1.70, P=0.022; Fig. 12). Results of PERMDISP did not show any significant differences in dispersion of samples between the two WG types ($F_{1,263}$=0.68, P=0.455) or between the during and post construction periods ($F_{1,263}$=1.22, P=0.338).

SIMPER results showed that during construction and post construction, the species that contributed the most to the community being repelled by PWG were raccoons, coyotes, black-tailed jackrabbits, opossums, eastern cottontails, white-tailed deer, striped skunks, domestic cats, and bobcats (Table 6). At BGWG, opossums, coyotes, raccoons, domestic cats and sheep, eastern cottontails, white-tailed deer, and striped skunks characterized the community of species being repelled during construction, which remained quite similar into the post-construction period (Table 6).

A two-way ANOVA showed no significant difference in monthly species richness between the PWG and BGWG (F_1=1.72, P=0.197); however, there was a significant difference between during and post construction (F_1=5.11, P=0.029, Fig. 13). There was no interaction between WG type and construction period for monthly species richness (F_1=0.21, P=0.649).

Discussion

The results of this study did not support the hypothesis that crossing rates for all species would increase after construction had ended. While not statistically significant (P=0.282), the average crossing rate decreased between the during and post construction periods for WCS1, WCS2, and WCS3A, which may be due to the overall reduction in activity at WCS1 and WCS2, as shown in Fig. 6 as well as the construction of two new structures, WCS3 and WCS4. This does not, however, take the communities using the WCS into consideration. The PERMANOVA results showed that, not only are the communities crossing at each WCS different, but they also differed between the two construction periods at each location.

The hypothesis that species richness of wildlife crossing at WCS post construction was supported by findings from analyses. Similarity percentages (SIMPER) provided further detail into changes in species activity and showed that each species that used each WCS showed its own increase, decrease, or lack of change in use from during to post construction. For example, raccoon crossings decreased, while bobcat and opossum crossings increased at both WCS1 and WCS2. Overall use of WCS3A increased, and increases in use by bobcat, javelina, and Texas indigo snake at WCS3A contributed to the change in community (Table 4). It is likely that the increase in use of WCS3A was, in part, due to the completion of exclusionary fencing along State Highway (SH) 100. This would be consistent with findings that adding fencing along the right-of-way helps increase wildlife crossings via underpasses and overpasses (Dodd et al. 2004, Braden et al. 2008, McCollister and van Manen 2010, Alonso et al. 2014, Seidler et al. 2018).

Increases in WCS use may also be due to habituation (Simpson et al. 2016). While WCS3A had been in place the longest out of the three WCS analyzed on SH 100 and already experienced relatively high wildlife traffic, species such as javelina were likely still becoming habituated to this structure. Javelinas had not been observed crossing at WCS3A at any point during the monitoring period prior to completion of construction. There had been many observations of entry-and-exit and approach behaviors, but there were no full crossings observed until about four months post construction. Use by javelina four months after construction demonstrates that even if structures are not used immediately, use may change over time. This was also seen in elk using newly constructed WCS in central Arizona (Gagnon et al. 2011) and in squirrel gliders using canopy bridges in southeast Australia (Soanes et al. 2013). Van der Grift and van der Ree (2015) hypothesized that WCS will increase into the future until the species using the WCS closely resembles the community surrounding the structure.

Habituation to WCS1 and WCS2 by bobcats and opossums likely occurred, as the newly widened catwalks provided a dry path to allow them to cross. Other studies have also shown that similar dry ledges have been beneficial in facilitating wildlife crossings (Meaney et al. 2007, Villalva et al. 2013). Our observations, as well as those of the aforementioned studies provide evidence that dry pathways are an important consideration in the construction and configuration of WCS that may have a seasonal or permanent flow of water.

It is unclear what is the likely cause of the decrease in raccoon activity between the two time periods at both WCS1 and WCS2. As these WCS have a nearly constant flow of water, the presence of aquatic animals, such as small fish, crabs, and crayfish have provided foraging opportunities for raccoons. Foraging was a frequently observed behavior of raccoons in and around these structures, mostly prior to completion of construction; however, raccoon activity declined after flooding at WCS1 and WCS2 resulting from heavy rainfall that occurred post construction. Furthermore, increased bobcat activity at WCS1 and WCS2 may have influenced the decrease in raccoon activity, as raccoon may have been avoiding predators.

The hypothesis that repel rates at WG would increase from during construction to post construction was disproven. Despite the lack of statistical significance, there was an overall pattern of decrease in repel rates and increase in crossing rates. These results are similar to what was observed for carnivore species encountering and crossing WG in northwestern Montana (Allen et al. 2013). The Wilcoxon rank sum test did not take repel rates of the communities interacting with these structures into account. PERMANOVA did show a difference in repel rates of communities between the during and post construction periods for both PWG and BGWG combined, and the post-hoc pairwise test did reveal a difference between the construction periods for PWG but not for BGWG.

The hypothesis that species richness would increase post construction was supported. Further insight into changes in species being repelled were provided by SIMPER analysis. The SIMPER analysis showed that at PWG, repels of raccoons decreased and repels of opossums increased from during to post construction, suggesting that these two species were the main drivers of differences observed at PWG (Table 6). At BGWG, increases in repel behavior by opossum and domestic cat likely drove the differences in communities from during to post construction (Table 6).

It is probable that wildlife started to become habituated to WG and learned how to cross. Not only were more wildlife approaching WG to inspect and test them out, but they eventually became familiar enough with the structures to cross over or through them. VanCauteren et al. (2009) also documented habituation to deer-guards by white-tailed deer and reported deer jumping across and using crossbars to walk across the guards. The same behaviors were observed of the two large ungulate species along SH 100, white-tailed deer and nilgai, though crossing events (n=10) were infrequent compared to repel events (n=126). On several occasions, attempts to jump across WG were unsuccessful, which was likely due to the length of the WG. Sebesta et al. (2003) had also reported deer-guard length as an important factor in preventing deer from crossing at these structures.

Bobcat, coyote, and opossum crossings at WG noticeably increased and are consistent with the findings of Allen et al. (2013) that WG are not a substantial barrier to multiple carnivore species, including bobcats and coyotes. On only one observed occasion, an individual ocelot encountered a PWG and crossed into the roadway without any apparent hesitation. This suggests that WG may not be an effective road mitigation strategy for ocelot or other felids, although caution should be used in drawing conclusions regarding endangered species from small sample sizes.

Habituation was demonstrated by more than just crossing behaviors. At PWG, several species, including domestic cats, opossums, long-tailed weasels, and several squamate species, were observed slipping between pipes into the excavated areas increasingly in the post construction period. This was likely due to frequent small rodent activity in and around WG as a source of prey, though it is speculated, based on observations in this study, that some wildlife sought these excavated areas as cover.

Wildlife guards installed along SH 100 were designed to deter wildlife from entering the road and to allow the passage of emergency vehicles. Pipe wildlife guards were constructed with 7.6 cm pipe and were supported by 15.2 cm wide steel beams. The size of the pipes and the underlying beams provided ample footing for animals to walk across PWG. Despite expectations that PWG would be effective in preventing crossings, several bobcats and the individual ocelot crossed into the roadway using the crossbars as footpaths. Another unintentional footpath that was created was the cement pad between side panel fencing and the WG. Although a similar design to BGWG was reported as the most effective design for deterring Florida Key deer (Peterson et al. 2003), this does not seem to be the case for coyotes, opossums, domestic cats, or domestic sheep along SH 100.

The original hypotheses, that crossing rates at WCS would increase post construction (1) and that repel rates at both WG types would increase post construction (3), were not supported by the findings of this study. Habituation of individuals over time likely played a factor in driving the observed WG results. However, the data showed increases in species richness over time using the WCS and being repelled at WG, supporting the hypothesis that species richness of wildlife crossing would increase during the post construction period (2) and species richness of wildlife being repelled would increase post construction (4).

Future Research

Overall, certain aspects of this study could be addressed to produce more meaningful results and information for advising management and other similar research. Several more years of monitoring would be beneficial for documenting patterns with habituation, seasonality, and other environmental factors. Additionally, it may be useful to analyze other types of behaviors besides movement, such as hesitation, foraging, species interactions, and day-bedding. The on-going monitoring of these mitigation structures will allow questions such as these to be addressed.

CHAPTER III

THE INFLUENCE OF STRUCTURAL CHARACTERISTICS AND ENVIRONMENTAL FACTORS ON WILDLIFE USE OF WCS

Introduction

In examining wildlife use of wildlife crossing structures (WCS), it is important not only to look at frequency of use and the differential use of WCS by multiple species but also to find possible reasons why these differences are being observed. Variables in the form of structural characteristics and environmental factors are important in attempting to explain differences in crossing structure effectiveness (Clevenger and Waltho 2000, 2005; van der Grift et al. 2013; van der Grift and van der Ree 2015).

Previous studies have considered dimension (Foster and Humphrey 1995, Clevenger and Waltho 2000, Mata et al. 2005), presence of water (Craveiro et al. 2019), noise (Clevenger and Waltho 2000, 2005), human activity (Clevenger and Waltho 2000, 2005), and nearby vegetation (van Vuurde and van der Grift 2005, Grilo et al. 2008) as important factors that may influence effectiveness of WCS. Studies have linked differential species use of underpasses to the openness ratio, which is the calculation of width × height / length (Yanes et al. 1995; Clevenger and Waltho 2000, 2005), as well as the presence of flowing water (Craveiro et al. 2019) and vegetative cover (van Vuurde and van der Grift 2005). Understanding the relationship between effectiveness of mitigation measures and their structural characteristics and environmental factors can aid in informing optimal placement and design of future mitigation structures (Bissonette and Adair 2008, Clevenger and Huijser 2011).

Wildlife crossing structures of various size and design were constructed by the Texas Department of Transportation on SH 100 located in South Texas to address ocelot (*Leopardus pardalis albescens*) mortality, improve connectivity among wildlife populations, and reduce wildlife-vehicle collisions (TxDOT 2015). Wildlife crossing structures along SH 100 provide an opportunity to examine how differential characteristics of WCS influence their use by wildlife. While they have been camera monitored (Cogan 2018), characteristics of the WCS and effect of surrounding landscape have not previously been related to wildlife use. Wildlife crossing structure dimensions, such as openness ratio, and presence of water have been shown to affect frequency of wildlife crossings (Clevenger and Waltho 2000, 2005; Craveiro et al. 2019), and these factors may be important to consider for WCS use in South Texas. Other factors such as distance to vegetation and daily precipitation may also be important factors affecting WCS use. Some species, such as ocelots, require dense vegetation (USFWS 2016) and likely a certain proximity of vegetation to WCS entrances. Additionally, excess rainfall occasionally creates flooded conditions which may reduce WCS use. Such inundation occurred at one WCS along SH 100 due to heavy rainfall and provided a unique opportunity to examine how water presence influences WCS use. An understanding of how and why wildlife differentially use WCS based on structural characteristics will better inform wildlife management in this unique system which supports multiple federal and state endangered species, including ocelot, Texas tortoise (*Gopherus berlandieri*), and Texas indigo snake (*Drymarchon melanurus erebennus*; TPWD 2011).

The objective of this chapter is to analyze structural characteristics of WCS (Table 1) and landscape variables that may be having an effect on wildlife use of WCS in the post construction period. In this chapter, the following hypotheses will be tested:

1. Counts of crossings at WCS will decrease at WCS with high openness ratios versus WCS with high openness ratios.
2. Counts of crossings at WCS will decrease as distance to nearest vegetative cover increases.
3. Counts of crossings at WCS will decrease as daily precipitation increases.
4. Crossing rates at WCS3 will decrease during periods of temporary flooding.

Study Area

This study was conducted along an 11.9 km stretch of State Highway (SH) 100 between Los Fresnos and Laguna Vista in Cameron County, Texas (Fig. 1). The road is flanked by USFWS protected Laguna Atascosa National Wildlife Refuge (LANWR) and agricultural and residential land. The study area is located within the Rio Grande Delta physiographic zone (Leslie 2016). The surrounding habitat is characterized by Texas coastal grassland, Tamaulipan saline thornscrub, and Texas coastal high salt marsh (USGS 2011, Leslie 2016). The diversity of ecological systems and land use along SH 100 result in varying amounts of vegetative cover, from open, sea-ox-eye daisy (*Borrichia frutescens*) flats, to smooth cordgrass (*Spartina alterniflora*) fields, to thornscrub patches with canopy cover (TPWD 2014, Leslie 2016). The area receives an average annual rainfall of 66 cm, with highs in the summer reaching an average of 33° C and lows in the winter getting down to an average of 12° C (NOAA 2020).

Construction of road mitigation structures resulted in a 1.8 m tall chain link fence spanning an 11.9 km length of SH 100, on the north and south sides of the highway, along five

WCS (Fig. 2) and 18 WG (Fig. 3). WCS1 and WCS2 are large concrete box culverts, each located in drainage ditches with permanent water (Table 1). WCS1 and WCS2 had previously been constructed with a wildlife crossing; a 41-cm round culvert pipe was placed above the water line to enable use (TxDOT 2015). WCS3 is a newly constructed bridge underpass with a natural substrate and ephemeral water presence (Table 1). WCS3A is a small concrete box culvert with a dirt substrate and ephemeral water presence (Table 1). WCS3A was already in existence prior to construction of the other WCS along SH 100 and was only modified during the construction period with fence replacement and cutting back vegetation. Lastly, WCS4 is a newly constructed medium concrete box culvert with a concrete substrate and no water presence (Table 1). Construction began 9 September 2016 and was completed on 10 May 2018.

Methods

Data Collection

Species activity at mitigation structures was monitored using active infra-red (AIR) and passive infra-red (PIR) triggered camera traps (Reconyx PC900 HyperFire™ and Reconyx HS2X HyperFire 2™; Reconyx LLP, Holmen, WI, USA) placed at each WCS and WG. Monitoring occurred from 10 January 2017 to 10 May 2019 at WCS and from 28 April 2017 to 10 May 2019 at WG . Each WCS had two cameras on each side of the WCS, with one camera trap facing toward the opening and one facing away (Fig. 4). One camera was equipped with both PIR and AIR triggers to capture wildlife interacting with the WCS. A second PIR-only camera trap faced away from the WCS to capture wildlife in the immediate surrounding areas that may not interact with the WCS (Fig. 4). The PIR trigger is activated within the camera by changes in radiation emitted in the camera’s field of view, while AIR trigger is activated by an externally triggered infrared beam deployed in the field of view (Welbourne et al. 2016).

Additionally, video cameras (Bushnell Trophy Cam HD Model 119874, Bushnell Corporation, Overland Park, KS, USA and Reconyx HyperFire 2™) were set up at each WCS entrance to supplement still photographs in wildlife-WCS interaction analysis. Additional cameras were set up at larger WCS openings to ensure that the full extent of the opening is captured and that the number of individual animals missed by the cameras is reduced.

To determine how WCS characteristics influence wildlife use, structural characteristics and environmental factors for each WCS along SH 100 were compiled. Wildlife crossing structure dimensions (length, width, and height) and openness ratio were previously obtained by TxDOT (2015). To determine influence of vegetation on WCS use, distance to the nearest patch of native vegetation from the WCS entrances was measured for WCS3, 3A, and 4 using a measuring tape. To calculate an average distance to vegetation at each crossing opening, three distance measurements were taken at each WCS opening: two following the lateral fence edges from the opening to vegetation and one from the center of the opening directly to the vegetation. Because WCS1 and WCS2 were situated in drainage ditches, the distance from the WCS opening to the top of the ditch and from the top of the ditch to the nearest large patch of native vegetation was measured. Lastly, daily precipitation and daily low temperatures were obtained from NOAA Climate Data Online (2020).

To understand how standing water influenced WCS use, the amount of water at WCS3 was categorized for comparison against wildlife crossing rates. At this WCS, water was pumped intermittently to remove pooled water using a gas-powered pump and hose (Model 100113, Champion Power Equipment, Santa Fe Springs, California, USA). The level of water pooling was separated into three categories: “0,” with very little to no water pooled under the WCS; “1,” with some water pooled and enough dry pathway for wildlife to cross under the WCS; “2,” with water pooled at its highest level and no room for wildlife to cross under the WCS (Fig. 14).

Data Management

To process and organize all photographs and videos, a suite of software programs developed by Sanderson and Harris (2013) was used. Each picture was relabeled with its timestamp using the programs Renamer and Special Renamer. The pictures from each location were then sorted into file folders based on species, then sorted again by number of individuals observed in each picture. The program DataOrganize was used to process the sorted file folder structure to catalog all pictures. This ensured that all sorting errors were addressed prior to analysis. The last step in this process was to run DataAnalyze, which used the output from DataOrganize to produce summary statistics of the data.

Data Analysis

To determine how wildlife interacted with WCS, behaviors were categorized into four different groups: 1) “crossing,” in which individuals completely cross from one end of the WCS to the other; 2) “entry” and exit on the same side, where individuals do not complete movement to the opposite end; 3) “approach” without entry; 4) “nearby,” where individuals are in the vicinity of a WCS but do not interact with it. Crossing, entry, approach, and nearby were used to calculate a crossing rate for WCS using the following formula:

$$\text{crossing rate} = \frac{\sum \text{no. of crossings}}{\sum(\text{no. of entry} + \text{no. of approach} + \text{no. of nearby})}$$

Data from all five WCS in only the post construction period were used in analyses, as it is assumed that all newly constructed mitigation structures were fully functioning post construction.

A permutational multivariate analysis of variance (PERMANOVA) was used to compare crossing rates of wildlife communities between the five WCS. To accomplish this, crossing rates were arranged in a matrix with species as the variable and monthly data bins by site as samples, and this matrix was imported into PRIMER-e v7. A Bray-Curtis similarity matrix was calculated for use in the PERMANOVA, and post-hoc pairwise PERMANOVA was used to compare communities between each pair of WCS.

To find potential relationships between environmental factors and full crossings at WCS, a generalized linear model (GLM) with a Poisson error distribution was performed using the *stats* package in Program R 3.6.1 (R Core Team 2019). The global model included openness ratio (0.06-0.54 m), height (1.2-2.1 m), length (22.6-54.9 m), width (1.8-6.1 m), daily precipitation (0-11.9 cm), daily low temperature (2.2-27.2 °C), daily high temperature (3.9-38.9 °C), and distance from WCS entrance to nearest large vegetation patch (0-83.9 m) as factors (Table 7), and daily counts of successful crossings of all species were the dependent variable.

Individual species models were used to find relationships between the factors tested in the global model and counts of successful crossings of the five most frequently observed species: coyote (*Canis latrans*), bobcat (*Lynx rufus*), raccoon (*Procyon lotor*), opossum (*Didelphis virginianus*), and nine-banded armadillo (*Dasypus novemcinctus*). Because daily counts of crossings for each species were low, often “1” or “0,” individual species counts of crossings were converted to binomial notation, and a binomial distribution was used for each species model. To determine which factors to include in each of these models, the dredge function from the MuMIn package was used as a model-selecting tool based on lowest change in Akaike information criterion, or ΔAICc (Barton 2019).

Differences in wildlife crossing rates for all species combined at the three assigned water levels at WCS3 were tested with a Kruskal-Wallis test, followed by a post-hoc Dunn's test with a Bonferroni correction to discern differences between pairs of water levels. These calculations were performed in Program R; the Dunn's test was performed using the FSA package (Ogle et al. 2019).

Results

A PERMANOVA showed that the wildlife communities crossing at WCS1, WCS2, WCS3, WCS3A, and WCS4 differed significantly in the post construction period (Pseudo-F_4=20.89, $P \leq 0.001$; Fig. 15). Further pairwise testing showed that each WCS had significantly different wildlife communities (Table 8).

Environmental Factors

The global generalized linear model (GLM) showed significant negative relationships between counts of crossings and openness ratio (GLMz_{4226}=-17.36, $P \leq 0.001$), precipitation (GLMz_{4226}=-2.16, P=0.031), length (GLMz_{4226}=-15.23, $P \leq 0.001$), and daily low temperature (GLMz_{4226}=-8.36, $P \leq 0.001$; Table 9). There were significant positive relationships between counts of crossings and WCS height (GLMz_{4226}=9.67, $P \leq 0.001$) and distance to vegetation (GLMz_{4226} = 9.60, $P \leq 0.001$; Table 9). This model had an R^2 of 0.15. Daily high temperatures were found to be correlated with daily lows (r=0.88), as was width with openness ratio (r=0.99), so daily high temperatures and width were omitted from the model.

As a result of model selection, the bobcat model included daily low temperature and precipitation as factors, and there was a significant negative relationship between counts of bobcat crossings and daily low temperature (GLMz_{266}=-2.03, P=0.042; Table 10). This model had a McFadden's pseudo-R^2 of 0.02.

The best-fitting model for coyote crossings included only WCS height, and there was a significant negative relationship between counts of coyote crossings and this factor (GLMz_{196}=-7.26, $P \leq 0.001$; Table 10). This model had a McFadden's pseudo-R^2 of 0.36.

The best-fitting raccoon model included openness ratio and distance to vegetation as factors. There were significant negative relationships between counts of raccoon crossings and openness ratio (GLMz_{433}=-3.73, $P \leq 0.001$) and vegetation distance (GLMz_{433}=-2.54, P=0.011; Table 10). This model had a McFadden's pseudo-R^2 of 0.04.

The best-fitting model for opossum crossings included openness ratio, WCS height, WCS length, precipitation, daily low temperature, and distance to vegetation as factors. There were significant negative relationships between counts of opossum crossings and openness ratio (GLMz_{1258}=-3.08, P=0.002), WCS length (GLMz_{1258}=-3.12, P=0.002), precipitation (GLMz_{1258}=-2.31, P=0.021), and daily low temperature (GLMz_{1258}=-3.78, $P \leq 0.001$; Table 10). There was a significant positive relationship between opossum crossings and vegetation distance (GLMz_{1258}=5.42, $P \leq 0.001$; Table 10). This model had a McFadden's pseudo-R^2 of 0.19.

Lastly, the best-fitting nine-banded armadillo model, as a result of model selection, only included WCS height as a factor, and there was a significant negative relationship between counts of armadillo crossings and WCS height (GLMz_{325} =-6.89, $P \leq 0.001$; Table 10). This model had a McFadden's pseudo-R^2 of 0.19.

Water levels at WCS3

A Kruskal-Wallis test determined that there was a significant difference in daily crossing rates at the three different water levels at WCS3 (χ^2_2=57.19, $P \leq 0.001$; Fig. 16). A Dunn test using a Bonferroni correction showed that there was no difference in daily crossing rates between water levels 0 and 1 (Z=0.25, P=1.00), but there were significant differences in daily crossing rates between water levels 1 and 2 (Z=6.93, P≤0.001) and levels 0 and 2 (Z=6.89, P≤0.001; Fig. 16).

Discussion

All factors in the global generalized linear model (GLM) were determined to be predictors of overall wildlife crossings, but these varied by species. Because there was a negative relationship between counts of wildlife crossings and increases in WCS openness ratio, the hypothesis that crossings would decrease at a high openness ratio versus a low openness ratio was supported. This negative association with openness ratio was also found to be true of black bears and mountain lions in Banff National Park, Canada (Clevenger and Waltho 2005) and small mammals and mustelids in Northwest Spain (Mata et al. 2004). When going further to examine effects of individual dimension parameters of WCS on wildlife crossings, it was found that crossings increased with increases in WCS height but decreased with increases in WCS length. Mule deer use of underpasses in Utah showed the same relationship with height and length (Cramer 2012), though Clevenger and Waltho (2000; 2005) found no significant relationship between carnivore underpass usage and these individual dimensions.

The hypothesis that crossings would decrease with increases in precipitation was also supported, and when examining effects of other weather variables, such as temperature, it was found that increases in minimum temperatures, or typically nighttime temperatures, had a negative effect on crossings as well. The hypothesis that wildlife crossings would increase as distance from nearest large vegetation patch to WCS entrance decreased, as was described of mustelids in the Netherlands (van Vuurde and van der Grift 2005), was not supported by the global model, as the opposite was found to be true.

Although statistically significant factors, global model estimates of daily low temperature, vegetation distance, and WCS length were low, thus suggesting low chances of influencing wildlife crossings. The likelihood of precipitation influencing wildlife crossings was also minimal. Estimates for openness ratio and WCS height suggested the highest chances of influencing wildlife crossings and may therefore be the most important factors for the species community using WCS. While the global model provides good baseline information, it is important to examine the influences of structural characteristics and environmental factors on individual species' use of WCS. Not all factors in the global model were important predictor variables in each species' use of WCS.

For bobcats, the best predictors of their crossing were the weather parameters. Only daily low temperature was significant, but the estimate for this factor suggested that chances of crossings changing due to this factor were relatively low. Bobcats are described as habitat generalists and are highly adaptable, so it is possible that cover, in the form of either WCS or vegetation, may not have been an important factor influencing their use of WCS (Schmidly 2004). Murphy-Mariscal et al. (2015) also found that structural characteristics played a minor role in bobcat use of underpasses, and bobcat use was more related to human activity and prey availability, which was not measured in this study.

WCS height was shown to be the only significant variable influencing coyote crossings, and the estimate suggested a high likelihood of a decrease in crossings as height increased. It is unclear why WCS height would influence coyote use of WCS, as there is scarce literature that suggests this. Coyotes have been documented using a wide range of WCS (Ng et al. 2004, Murphy-Mariscal et al. 2015), and multiple road mitigation handbooks suggest that most type of underpasses are suitable for coyotes (Huijser et al. 2008, Clevenger and Huijser 2011). It is possible that coyote followed smaller animals, or potential prey, into WCS with shorter heights, but such interactions were not well documented in this study.

Height was also shown to affect armadillo use of WCS. Use of WCS with lower heights by armadillos may be explained by their tendency toward brushy areas (Schmidly 2004, Reid 2006), and WCS with lower heights, such as WCS3A, may be a potential substitute for cover. Additionally, armadillos were only observed at WCS3A and WCS3, which are the only WCS within the study area with natural soil substrate. Armadillos tend to roll in wet mud to cool off during warm temperatures, and when soils harden, this provides opportunities for armadillos to forage for insects (Schmidly 2004). Both of these behaviors were observed in this dataset and may be influencing factors in armadillo use of WCS.

Increases in both openness ratio and distance to vegetation had significant negative effects on raccoon crossings, allowing us to accept the hypotheses regarding these parameters, but the low estimate of distance to vegetation suggested that it had a relatively low likelihood of causing raccoon crossings to decrease. Openness ratio had a greater likelihood of causing raccoon crossings to decrease. Raccoons crossed at all five WCS. Despite their use of WCS1 and WCS2 having decreased from during construction to post construction, raccoons crossed the most at these two WCS post construction, followed by WCS3A. Their high usage of WCS1 and WCS2 may have been due the regular flow of water through these structures and raccoons' association with water (Schmidly 2004). Raccoon use of WCS3A may have been due to the age of the structure and their established familiarity with this WCS. Although the negative relationship between raccoon crossings and openness ratio are consistent with the findings of Mata et al. (2004) that smaller animals tend to choose smaller culverts, other attributes such as presence of water are likely influencing raccoon use.

The opossum model included all the global model variables. This may be due to opossum crossings comprising of nearly a third of total post construction wildlife crossings across all WCS. Similar trends were observed in terms of directionality and significance with these variables, with the exception of WCS height, which in the opossum model, had a statistically insignificant negative effect on opossum crossings. The estimate for openness ratio from the model suggests that this factor has a high likelihood of causing a decrease in opossum crossings as openness ratio increases. This finding is also consistent with that of Mata et al. (2004), as opossums' smaller body size may be related to their crossing through smaller WCS. The estimate for precipitation suggests a moderate likelihood that this factor negatively influences opossum crossings. While high amounts of precipitation resulting in WCS flooding may have temporarily kept opossums from using them, it is also possible that pooled water following rain resulted in a lesser need for opossums to go in search of a water source.

Clevenger and Waltho (2000), Mata et al. (2004), Ng et al. (2004), Grilo et al. (2008), and Gagnon et al. (2011) reported WCS dimensions as some of the most important factors influencing certain species' crossings; however, this was not always the case in this study. As previously noted, bobcats in this study were not influenced in their crossing frequency by structural characteristics. Contrary to the above cited studies, Murphy-Mariscal et al. (2015) found that structural dimensions of underpasses were minorly influential in carnivore use, but instead, other factors such as habitat fragmentation and human activity appeared to have had an impact on bobcat and coyote use.

Many studies that attempt to describe factors that affect wildlife use of WCS include variables other than structural dimensions, such as noise (Clevenger and Waltho 2000, Jackson and Griffin 2000), surrounding habitat type and quality (Ng et al. 2004, Grilo et al. 2008), proximity to other WCS (Clevenger and Waltho 2005), human activity (Clevenger and Waltho 2000, Gagnon et al. 2011, Murphy-Mariscal et al. 2015), proximity to towns (Clevenger and Waltho 2000, 2005), flow of water through underpasses (Serronha et al. 2013, Craveiro et al. 2019), and existing wildlife home ranges and life history (Mata et al. 2004, Serronha et al. 2013, Martinig and Bélanger-Smith 2016). The results of this study were likely influenced by several of these factors, and it may be beneficial to include them in future aspects of this project.

Another variable that has been reported to influence wildlife use of underpasses is flooding (Craveiro et al. 2019, Serronha et al. 2013). For this study, the hypothesis that wildlife crossing rates would decrease during periods of flooding at WCS3 was partially supported. When flooding was at its highest, crossing rates for all species dropped significantly. As water levels receded, crossing rates increased. A pathway developed along the edges of water that was concentrated toward the center and low point of WCS3, and this allowed smaller species, such as opossums, raccoons, and cottontails to pass under the road. White-tailed deer eventually became accustomed to the shallow, pooled water and were able to cross through the WCS as well. This is consistent with the findings of Craveiro et al. (2019) and Serronha et al. (2013) that intermediate levels of water in underpasses pose a minimal barrier to wildlife. Given this, it is important to consider WCS3's construction flaw in that water does not drain away on its own. In order to reach an intermediate level of water following significant flooding, this underpass must be at least partially drained by pumping out the water. This restores function to WCS3, but if personnel are not immediately available to complete this task, then wildlife movement will be hindered.

WCS1, WCS2, and WCS3A have also experienced flooding. In some cases, the water level has been high enough to completely bar animals from approaching or entering. At more intermediate water levels, wildlife have been observed continuing to use WCS, including at WCS3A where a dry pathway is not available. On several occasions, raccoons, coyotes, and even bobcats have been observed using WCS3A at chest-deep levels of flooding. At WCS1 and WCS2, the catwalks provide a dry path for wildlife to use. The major difference between these WCS and WCS3, however, is that water drains away quickly, and flooding is short-lived. Because of this, there were not enough data to examine the effects of flooding on wildlife crossing rates at these WCS in this study, though with several more years of data, it may be a possibility.

One of the major limitations of this study is that it is the result of only one year of data. With only five WCS of four different designs, there is a lack of replicates, and in this situation, time seems to be the best substitute for this. Several more years of monitoring WCS would elucidate the more telling relationships between environmental factors and wildlife crossings. Gagnon et al. (2011) stresses the importance of long-term studies to take wildlife habituation into consideration and to better understand temporal changes in WCS use. Patterns of seasonality, of both wildlife activity and weather, could be revealed by continuing this study beyond one year.

Nevertheless, a longer study period would not completely make up for the lack of replicates. This is important in determining what type of WCS is used by which species. The relationships between environmental factors and wildlife crossings were only examined in five species because these species were observed using two or more WCS, and sample sizes for each species were large enough. As an example, white-tailed deer only crossed at WCS3 during this study, which is insufficient as it only provides one set of criteria to examine in relation to their WCS use. Even for species observed at more than one WCS, with the exception of WCS1 and

WCS2, only having one of each WCS design makes it difficult to tease out which parameters influence differential use of WCS.

Though the lack of replicates in this study cannot be amended, and one could compile a seemingly infinite number of possible variables that affect wildlife use of WCS, improvement is not out of the realm of possibility. Extending this study beyond one year of data and including other likely influential factors would be important ways to make the most out of monitoring WCS and determining their effectiveness.

The hypotheses that increases in openness ratio (1), distance to vegetation (2), daily precipitation (3) had negative effects on wildlife crossings at WCS were partially supported by the findings of this study. This is due to the fact that individual species' responses to these factors were varied. The hypothesis that pooling of water at WCS3 would cause a decrease in wildlife crossing rates (4) was also partially supported in that intermediate levels of water allowed access to dry pathways, and high levels of water did not. Overall, the findings of this study provide a baseline of information about factors that may influence effectiveness of WCS and also a framework for continuing to study these factors and possibly a few others. The continuation of this research would allow for a better understanding of the relationship between wildlife and WCS.

CHAPTER IV

CONCLUSION

This thesis shows the progression of how wildlife crossing structures (WCS) and wildlife guards (WG) on State Highway (SH) 100 are performing from their construction or modification through their first year of full functionality. The continuation of monitoring and evaluating wildlife use of WCS and avoidance of WG over several more years is key in getting the best insight regarding the effectiveness of these structures. In studying the new mitigation structures along SH 100, more possible explanatory variables should be considered as factors affecting use of WCS.

As suggested in Braden et al. (2008), Huijser and McGowen (2010), and McCollister and van Manen (2010), just having WCS and fencing in place is important in increasing connectivity lost from the construction of roads. The most effective situations were where frequent WCS and multiples of different types, which provided opportunities for a variety of taxa, from amphibians to large ungulates and carnivores, to safely cross roads without getting struck by vehicles (Mata et al. 2004, Bissonette and Adair 2008). Although a variety of structure designs were placed along SH 100, specifically for ocelots, such an extensive network of WCS along SH 100 is not likely a possibility. However, more WCS are being constructed in South Texas, providing an opportunity to increase connectivity elsewhere in the region. Even though most of these mitigation structures are placed for the benefit of threatened and endangered species, other species may benefit as well. Certainly, as budgets may be restrictive, the best strategy is to understand target species' home ranges and habitat requirements and apply that to WCS placement and construction (Ng et al. 2004, Grilo et al. 2008, Murphy-Mariscal et al. 2016).

As WG continue to age, wildlife are expected to continue to become habituated to them, and their effectiveness may reduce over time. Because WG on SH 100 are not planned to be removed in the foreseeable future, it would be beneficial to examine variables, such as nearby vegetation and habitat, that may affect how wildlife interact with them. Again, continuing the monitoring of WG will provide greater insight into repel and crossing rate patterns at these sites.

Not only is this thesis a snapshot of the "lives" of road mitigation structures on SH 100, but it is also part of a broader, long-term project involving research on other aspects of road ecology on this highway. This research includes examining patterns of wildlife road mortalities, surrounding wildlife communities, and other mitigation structures not included in this study. Synthesizing this research will be crucial to understanding the broader picture that is the ecology of SH 100, which will help to inform future construction of mitigation structures in South Texas and consequently aid in the conservation of ocelots and other threatened or endangered species in the region such as the Texas tortoise and Texas indigo snake.

The results of this study show that differential wildlife use of WCS was affected by one or more factors (e.g. structural dimensions, distance to nearby vegetation, water presence), and as such, it is recommended that these factors be considered in species-specific approaches prior to the construction of mitigations structures. Additionally, future monitoring of these structures could address wildlife habituation to WCS and other structures. This work will add to a global effort to understand the efficacy of a wide variety of road mitigation measures and successfully manage for the negative impacts that roads have on wildlife. Furthermore, this research and its implications may be applied in similar ecological systems in order to conserve wildlife and mitigate for roads.

Management Implications

Given individual species' differing responses to openness ratio and distance to nearby native vegetation, selecting a variety of underpass WCS designs for construction and placing WCS at the highest frequency possible is advised in order to increase connectivity for a variety of species. Additionally, managing the influence of water within WCS by providing a dry ledge or draining excessive pooled water to maintain a footpath for wildlife is recommended for maintaining functionality of underpass WCS. Allowing some water to remain as an attractant or resource for wildlife, as well as a corridor for semi-aquatic species, would also be beneficial. Lastly, in order for WG to be effective, WG of any design should not allow sufficient footing for wildlife. Narrower beams and crossbars with widest possible spacing are recommended to minimize wildlife crossing, and fencing adjacent to WG should extend all the way to beams or crossbars so as to obstruct potential pathways for wildlife to cross along the WG edge.

TABLES

Table 1. Type and dimensions (width, height, and length in meters) of wildlife crossing structures (WCS) constructed by the Texas Department of Transportation to address ocelot (*Leopardus pardalis abescens*) mortality along State Highway 100 in Cameron County, Texas, USA. All structures were monitored using remote cameras, and data were collected at WCS from January 2017 to May 2019. Openness ratio was calculated as width × height / length.

Attribute	WCS1	WCS2	WCS3	WCS3A	WCS4
Underpass type	box culvert	box culvert	bridge	box culvert	box culvert
Dimensions	3.0 x 2.1 x 48.8	3.0 x 2.1 x 54.9	6.1 x 2.0 x 22.6	1.8 x 1.2 x 35.1	3.0 x 1.5 x 24.4
Openness ratio	0.13	0.11	0.54	0.06	0.18

Table 2. List of species and species group recorded interacting with wildlife crossing structures and wildlife guards constructed by the Texas Department of Transportation to address ocelot (*Leopardus pardalis albescens*) mortality on State Highway 100 in Cameron County, Texas, USA between January 2017 and May 2019.

Common name	Scientific name
Virginia opossum	*Didelphis virginianus*
Nine-banded armadillo	*Dasypus novemcinctus*
American beaver	*Castor canadensis*
Nutria	*Myocastor coypus*
Mexican ground squirrel	*Ictidomys mexicanus*
Eastern cottontail	*Sylvilagus floridanus*
Black-tailed jackrabbit	*Lepus californicus*
White-tailed deer	*Odocoileus virginianus*
Javelina	*Tayassu tajacu*
Feral hog	*Sus scrofa*
Domestic cattle	*Bos taurus*
Nilgai	*Boselaphus tragocamelus*
Domestic sheep	*Ovis aries*
Bobcat	*Lynx rufus*
Ocelot	*Leopardus pardalis*
Domestic cat	*Felis catus*
Coyote	*Canis latrans*
Domestic dog	*Canis lupus familiarus*
Northern raccoon	*Procyon lotor*
Striped skunk	*Mephitis mephitis*
Long-tailed weasel	*Mustela frenata*
Texas tortoise	*Gopherus berlandieri*
Texas indigo snake	*Drymarchon melanurus erebennus*
Grouped herpetofauna	

Table 3. Number of wildlife interactions and count of different species detected by cameras at three wildlife crossing structures (WCS) constructed by the Texas Department of Transportation to address ocelot (*Leopardus pardalis albescens*) mortality on State Highway 100 in Cameron County, Texas, USA. Monitoring was broken into two time periods; during construction (January 2017-May 2018) and post construction (May 2018-May 2019).

	Interactions						Species Count			
	During			Post			During		Post	
WCS	Crossing	Refusal	Total	Crossing	Refusal	Total	Crossing	Total	Crossing	Total
1	394	236	630	182	159	341	4	9	6	11
2	651	600	1251	648	395	1043	3	12	5	15
3A	1526	1105	2631	1108	877	1985	11	17	11	18

Table 4**.** Most abundant species crossing at each wildlife crossing structure (WCS), during and post construction periods, based on similarity percentage analysis of data collected from January 2017 to May 2019 in Cameron County, Texas, USA. Average abundance denotes average number of individuals crossing per month, and percent (%) contributed shows the extent to which each species contributed to the clustering of the species observed within each group.

Site	During			Post		
	Species	Average Abundance	% Contributed	Species	Average Abundance	% Contributed
WCS1	raccoon	35.1	99.9	raccoon	11.7	91.8
				bobcat	1.2	4.4
				opossum	0.7	3.8
WCS2	raccoon	58.6	99.9	raccoon	10.9	55.6
				opossum	36.2	35.6
				bobcat	2.6	8.8
WCS3A	opossum	56.8	65.3	opossum	42.9	54.6
	coyote	14.2	12.5	bobcat	10.0	12.2
	raccoon	6.3	10.2	nine-banded armadillo	9.5	11.3
	nine-banded armadillo	8.2	9.7	coyote	8.2	7.7
	bobcat	2.1	2.1	raccoon	4.1	7.6
				javelina	7.5	4.5
				Texas indigo snake	0.8	0.9

Table 5. Number of wildlife interactions and count of different species detected by cameras at two wildlife guard (WG) types constructed by the Texas Department of Transportation to address ocelot (*Leopardus pardalis albescens*) mortality on State Highway 100 in Cameron County, Texas, USA. Monitoring was broken into two time periods; during construction (April 2017-May 2018) and post construction (May 2018-May 2019).

	Interactions						Species Count					
	During			Post			During			Post		
WG	Repel	Crossing	Total	Repel	Crossing	Total	Repel	Crossing	Total	Repel	Crossing	Total
Bridge grating	1145	385	1530	1295	701	1996	17	11	17	18	14	18
Pipe	788	252	1040	939	433	1372	15	9	15	18	11	18

Table 6. Most abundant species being repelled at pipe wildlife guards (PWG) and bridge grating wildlife guards (BGWG), during and post construction, as a result of similarity percentage analysis of data collected from April 2017 to May 2019 on State Highway 100 in Cameron County, Texas, USA. Average abundance denotes average number of individuals being repelled per month, and percent (%) contributed shows the extent to which each species contributed to the clustering of the species observed within each group.

Type	During			Post		
	Species	Average Abundance	% contribution	Species	Average Abundance	% contribution
PWG	raccoon	2.0	55.9	opossum	4.0	40.3
	coyote	1.1	20.3	raccoon	0.6	16.0
	black-tailed jackrabbit	0.6	11.0	coyote	0.8	13.0
	opossum	0.7	3.9	black-tailed jackrabbit	0.9	10.6
	eastern cottontail	0.4	2.8	striped skunk	0.6	9.7
	white-tailed deer	0.3	1.8	eastern cottontail	1.9	4.8
	striped skunk	0.2	1.8	domestic cat	0.3	2.0
	domestic cat	0.3	1.3	white-tailed deer	0.3	1.5
	bobcat	0.1	0.8	bobcat	0.2	0.9
				nine-banded armadillo	0.2	0.1
BGWG	opossum	2.7	61.9	opossum	5.3	68.6
	coyote	0.6	14.3	domestic cat	2.1	10.7
	raccoon	0.4	8.6	coyote	0.7	8.8
	domestic cat	0.5	4.7	striped skunk	0.6	5.5
	domestic sheep	1.1	4.3	white-tailed deer	0.4	2.2
	eastern cottontail	1.1	2.8	raccoon	0.3	1.5
	white-tailed deer	0.2	1.4	domestic sheep	0.6	1.1
	striped skunk	0.3	1.2	eastern cottontail	0.2	0.9

Table 7. Descriptive statistics, including mean, standard deviation, and range, of quantitative factors being tested in the generalized linear model for data collected May 2018-May 2019 on State Highway 100 in Cameron County, Texas, USA.

	Mean ± SD	Range
Daily Low Temperature (°C)	18.4±6.8	2.2-27.2
Openness ratio	0.22±0.19	0.06-0.54
WCS Height (m)	1.63±0.37	1.2-2.1
WCS Length (m)	32.7±11.0	22.6-54.9
Precipitation (cm)	0.16±0.78	0.0-11.9
Distance to Vegetation (m)	12.4±17.7	0.0-83.9

Table 8. Results of pairwise permutational analysis of variance showing significant differences in the wildlife communities crossing between each pair wildlife crossing structures (df=24), from data collected post construction (May 2018-May 2019) on State Highway 100 in Cameron County, Texas, USA.

WCS Pair	*t*	*P*
WCS1, WCS2	1.60	0.047
WCS1, WCS3	4.90	≤0.001
WCS1, WCS3A	5.55	≤0.001
WCS1, WCS4	5.68	≤0.001
WCS2, WCS3	3.81	≤0.001
WCS2, WCS3A	4.35	≤0.001
WCS2, WCS4	5.08	≤0.001
WCS3, WCS3A	3.82	≤0.001
WCS3, WCS4	4.21	≤0.001
WCS3A, WCS4	4.69	≤0.001

Table 9. Results of global generalized linear model testing counts of all species crossings against structural characteristics and landscape variables in the post construction period (May 2018-May 2019) in Cameron County, Texas, USA.

	Estimate ± SE	*Z*	P
* Intercept	1.33 ± 0.13	10.15	≤0.001
* Openness	-5.29 ± 0.30	-17.36	≤0.001
* WCS Height	1.19 ± 0.12	9.67	≤0.001
* WCS Length	-0.05 ± 0.003	-15.23	≤0.001
* Precipitation	-0.06 ± 0.03	-2.16	0.031
* Daily Low Temperature	-0.02 ± 0.002	-8.36	≤0.001
* Distance to Vegetation	0.01 ± 0.001	9.60	≤0.001

* statistical significance within 95% confidence intervals.

Table 10. Results of best-fitting generalized linear models resulting from the dredge function in R, testing binomial counts of crossings of individual species against environmental factors, from data collected post construction (May2018-May 2019) in Cameron County, Texas, USA.

	Estimate ± SE	*Z*-value	*P*
Bobcat			
* Intercept	2.26 ± 0.70	3.22	≤0.001
Precipitation	-0.27 ± 0.15	-1.73	0.083
* Daily Low Temperature	-0.04 ± 0.02	-2.03	0.042
Coyote			
* Intercept	7.99 ± 1.0	7.93	≤0.001
* WCS Height	-5.04 ± 0.69	-7.26	≤0.001
Raccoon			
* Intercept	1.86 ± 0.23	8.01	≤0.001
* Openness ratio	-3.28 ± 0.88	-3.73	≤0.001
* Distance to Vegetation	-0.01 ± 0.004	-2.54	0.011
Opossum			
* Intercept	7.30 ± 0.63	11.54	≤0.001
* Openness ratio	-4.78 ± 1.55	-3.08	0.002
WCS Height	-0.99 ± 0.70	-1.42	0.154
* WCS Length	-0.07 ± 0.02	-3.12	0.002
* Precipitation	-0.27 ± 0.12	-2.31	0.021
* Daily Low Temperature	-0.04 ± 0.01	-3.78	≤0.001
* Vegetation Distance	0.05 ± 0.01	5.42	≤0.001
Nine-banded Armadillo			
* Intercept	4.27 ± 0.67	6.38	≤0.001
* WCS Height	-3.49 ± 0.51	-6.89	≤0.001

* statistical significance within 95% confidence intervals.

FIGURES

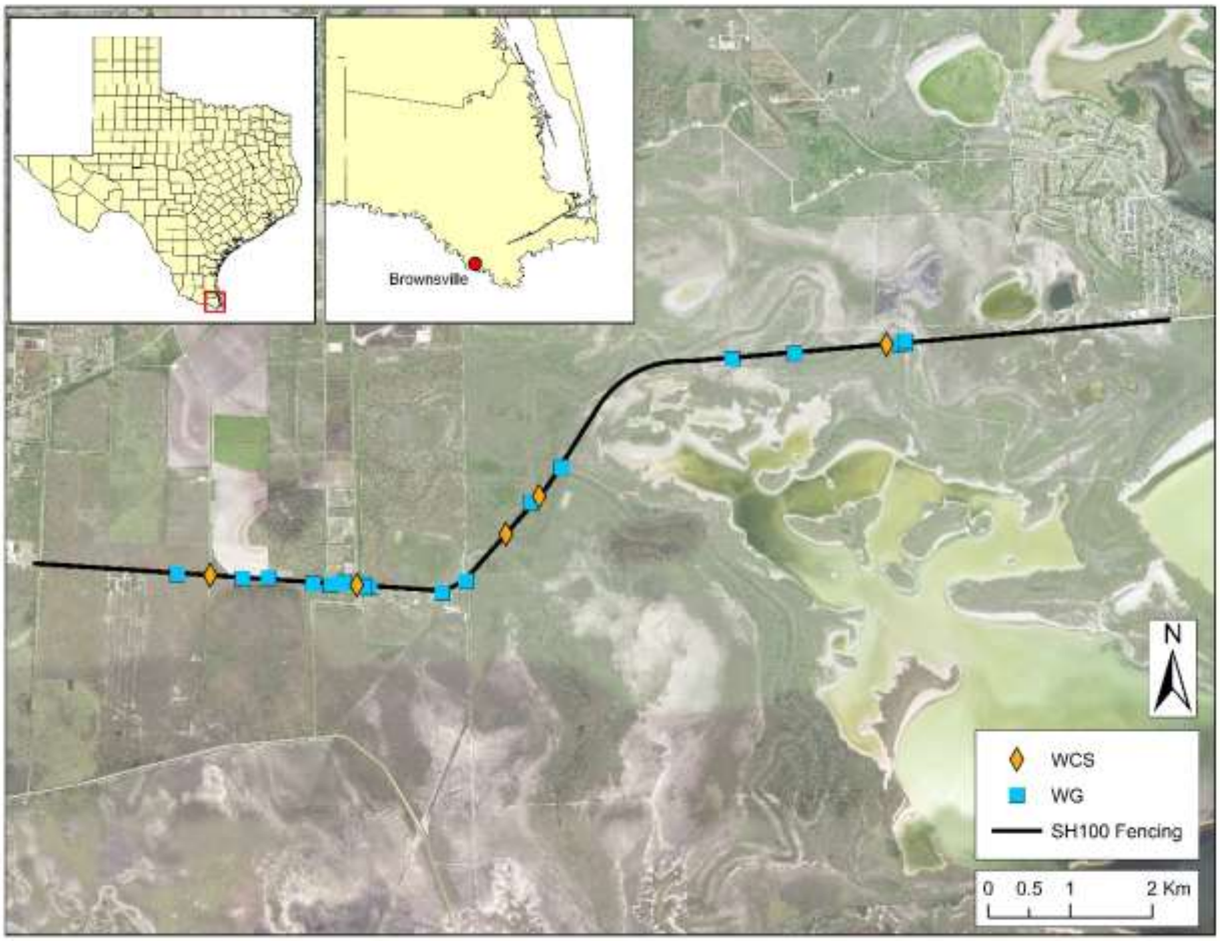

Figure 1. Wildlife crossing structures (WCS; n = 5), 11.9 km of fencing, and wildlife guards (WG; n = 18) constructed by the Texas Department of Transportation to address ocelot (*Leopardus pardalis albescens*) mortality along State Highway 100 in Cameron County, Texas, USA. All structures were monitored using remote cameras, and data were collected at WCS from January 2017 to May 2019 and at WG from April 2017 to May 2019.

Figure 2. Four of the five wildlife crossing structures (WCS) constructed by the Texas Department of Transportation to address ocelot (*Leopardus pardalis albescens*) mortality along State Highway 100 in Cameron County, Texas, USA. From top left: WCS2 (a), WCS3 (b), WCS4 (c), and WC3A (d). WCS1 is not pictured because it has the same configuration as WCS2.

Figure 3. The two wildlife guard (WG) types constructed by the Texas Department of Transportation to address ocelot (*Leopardus pardalis albescens*) mortality along State Highway 100 in Cameron County, Texas, USA: pipe (left) and bridge grating (right). WG were monitored using remote cameras, and data were collected at these sites from April 2017 to May 2019.

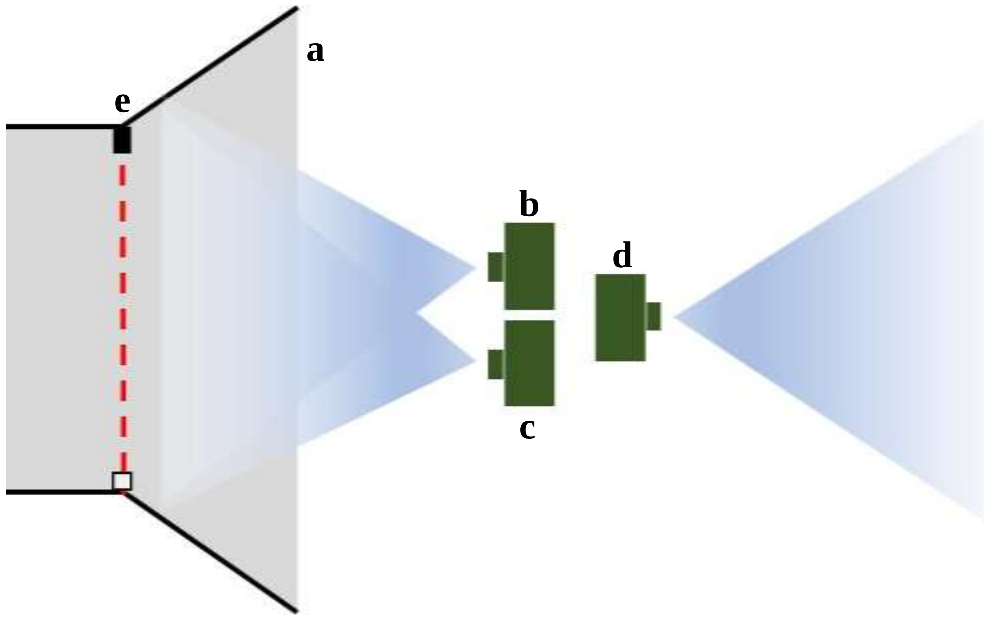

Figure 4. Example of camera trap setup at the opening of a wildlife crossing structure (WCS; a) constructed by the Texas Department of Transportation to address ocelot (*Leopardus pardalis albescens*) along State Highway 100 in Cameron County, Texas, USA from January 2017 to May 2019. One active infrared camera (b) and one video camera (c) face toward the WCS opening, while one passive infrared camera (d) faces away. The external sensing system is represented by the black and white boxes connected with the red dashed line (e).

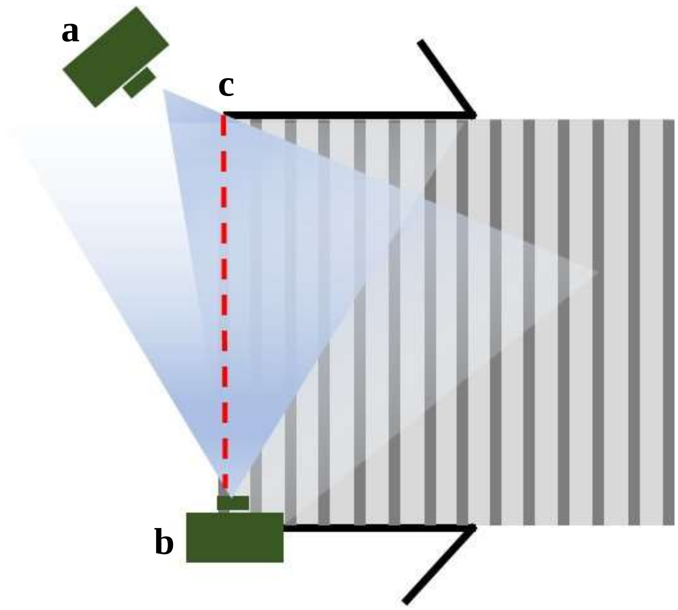

Figure 5. Example of camera trap setup at a wildlife guard (WG) constructed by the Texas Department of Transportation to address ocelot (*Leopardus pardalis albescens*) mortality along State Highway 100 in Cameron County, Texas, USA from April 2017 to May 2019. One active infrared camera (a) faces toward the WG and road, while one passive infrared camera (b) faces toward the habitat side. The external sensing system is represented by the red dashed line (c).

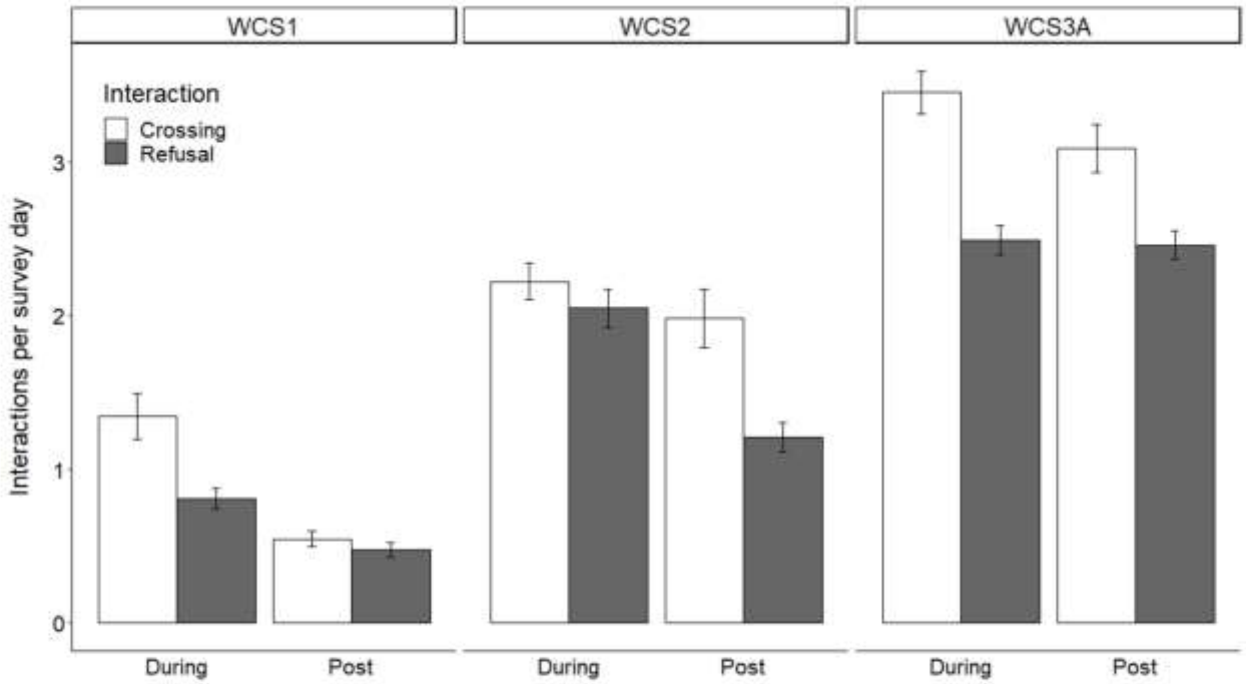

Figure 6. Bar graph showing mean number of crossings and refusals per survey day ± standard error at each wildlife crossing structure (WCS) during construction (January 2017-May 2018) and post construction (May 2018-May 2019). Crossings and refusals were significantly different between WCS1, WCS2, and WCS3A (crossings: *P*=0.001; refusals: *P*=0.001) and significantly higher during construction than post construction (crossings: *P*=0.001; refusals: *P*=0.001) along State Highway 100 in Cameron County, Texas, USA.

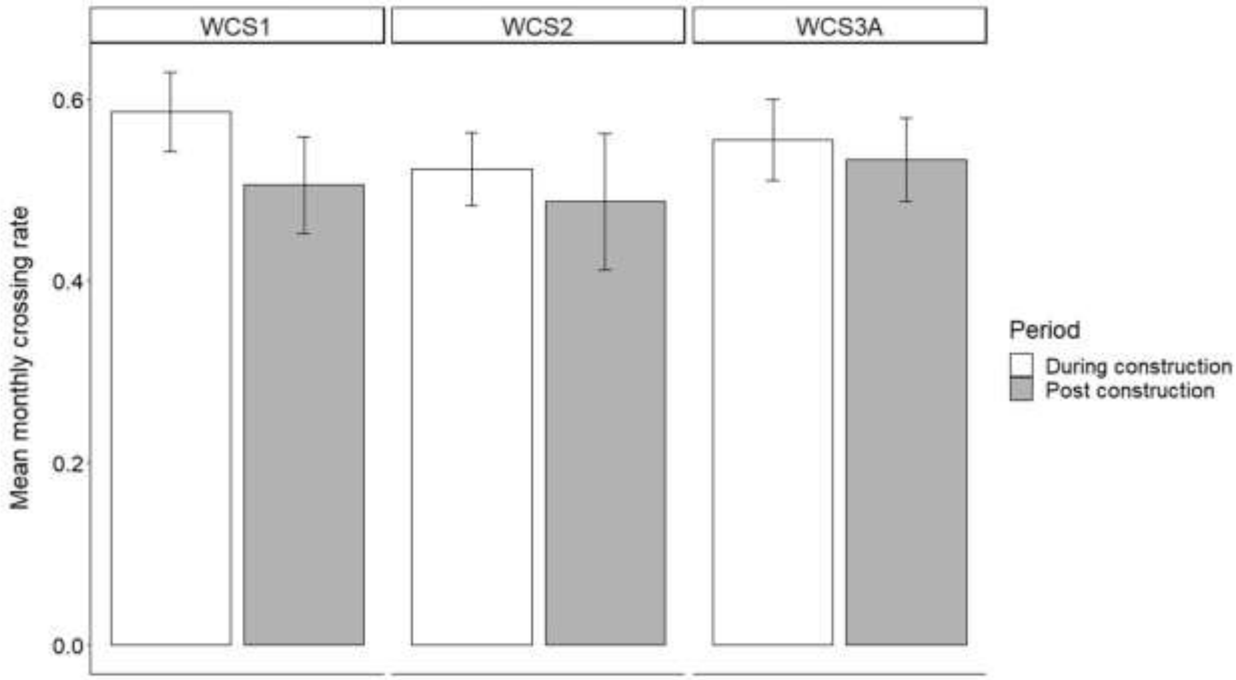

Figure 7. Bar graph showing mean monthly crossing rates ± standard error of all species combined at each wildlife crossing structure (WCS) during construction (January 2017-May 2018) and post construction (May 2018-May 2019) along State Highway 100 in Cameron County, Texas, USA. Crossing rates were not significantly different between WCS1, WCS2, and WCS3A (*P*=0.714) or between during construction and post construction (*P*=0.282).

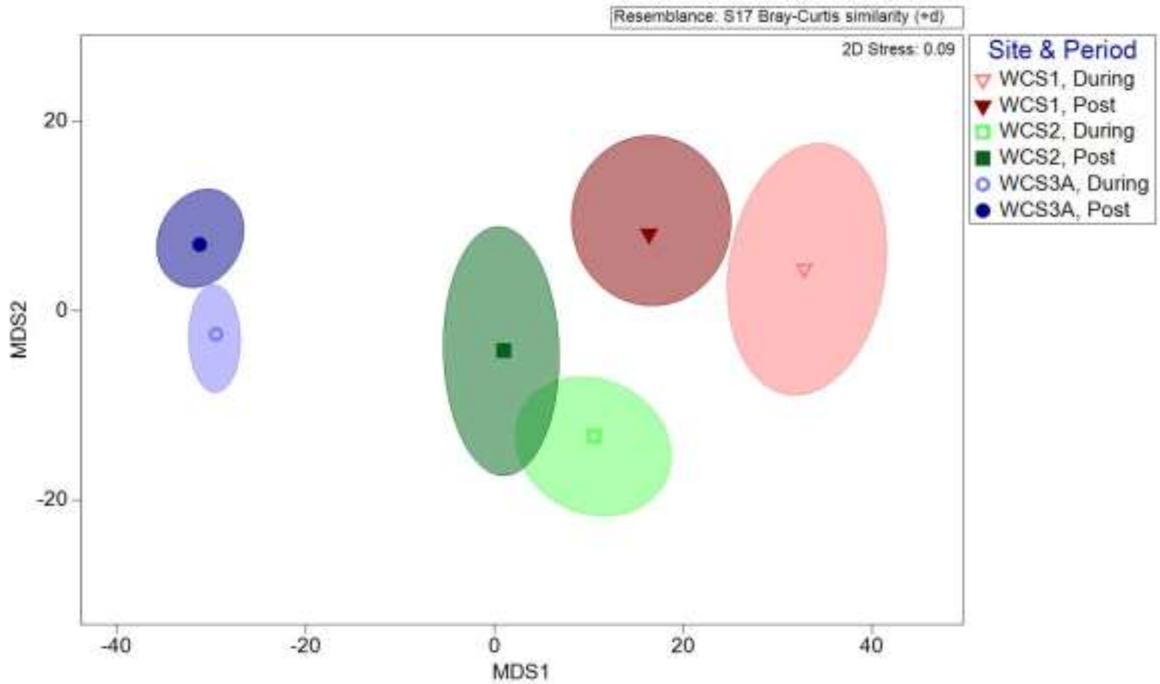

Figure 8. Bootstrapped metric MDS plot showing differences in monthly crossing rates between communities observed at three wildlife crossing structures ($P \leq 0.001$) and between during construction (Jan 2017-May 2018) and post construction (May 2018-May 2019; $P \leq 0.001$) along State Highway 100 in Cameron County, Texas, USA. Differences are indicated by physical distance between bootstrapped averages surrounded by 95% confidence areas.

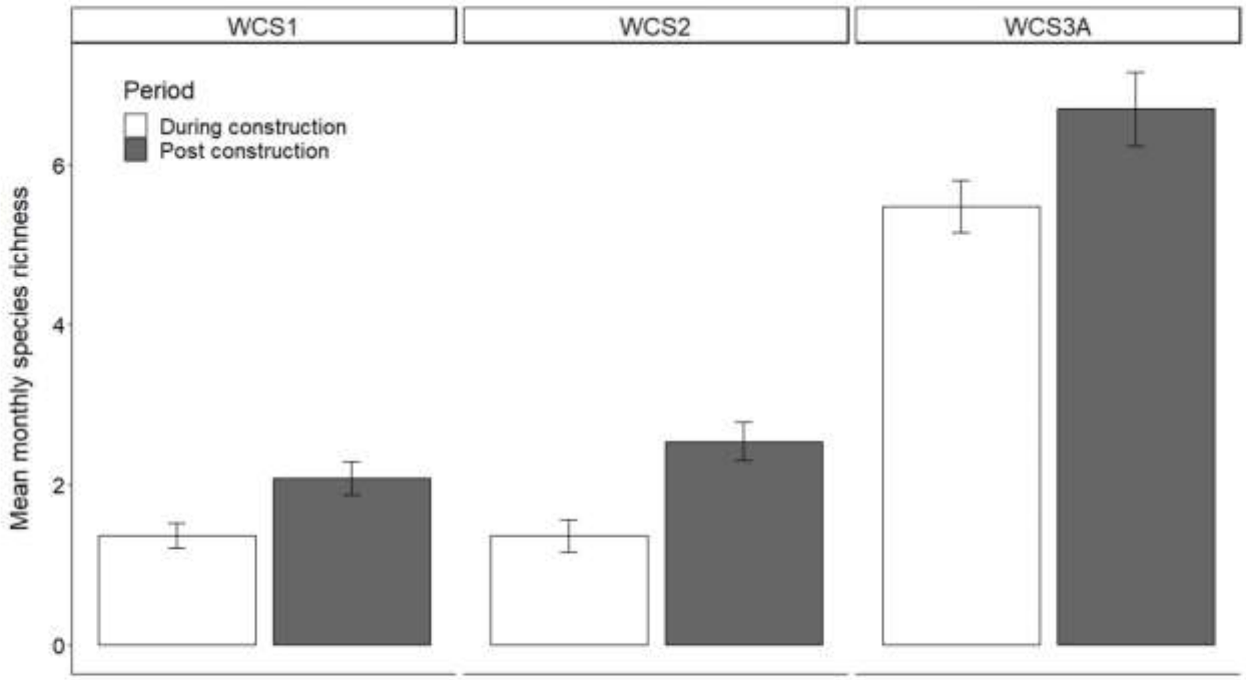

Figure 9. Bar graph showing mean monthly species richness ± standard error for each wildlife crossing structure (WCS) during construction (Jan 2017-May 2018) and post construction (May 2018-May 2019) along State Highway 100 in Cameron County, Texas, USA. Species richness was significantly higher at WCS3A ($P \leq 0.001$) and significantly higher post construction ($P \leq 0.001$).

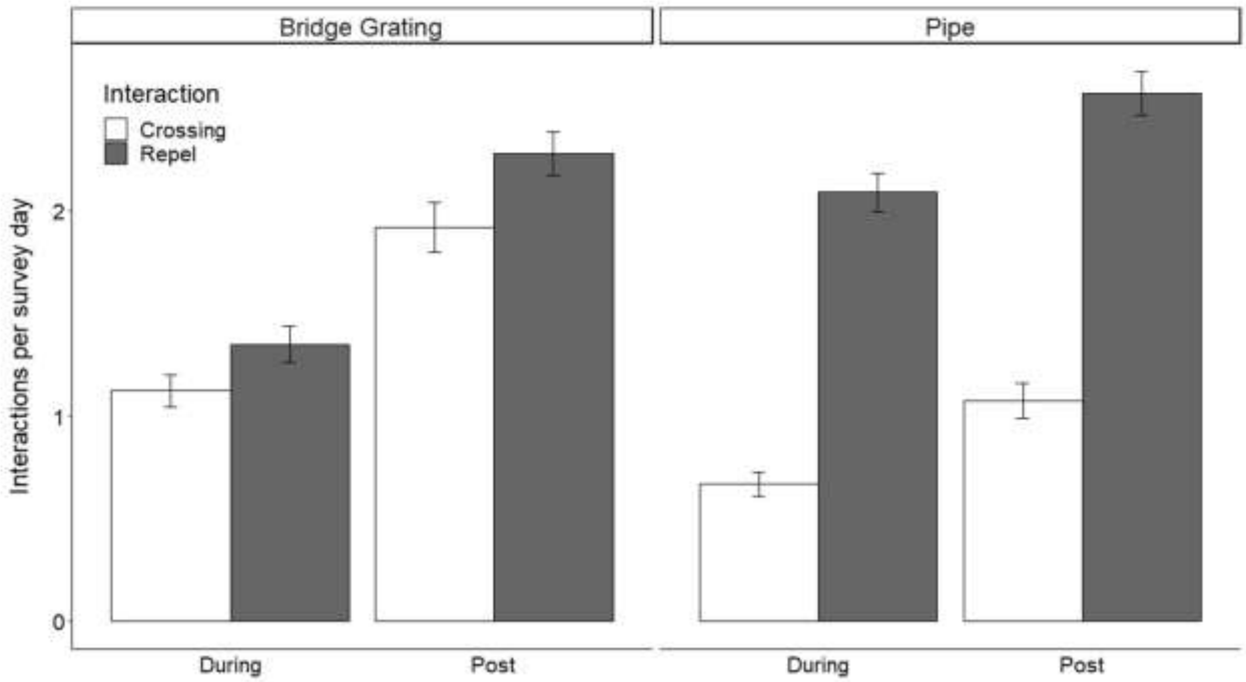

Figure 10. Bar graph showing mean number of repels and crossings per survey day ± standard error at pipe and bridge grating wildlife guards during construction (Apr 2017-May 2018) and post construction (May 2018-May 2019) along State Highway 100 in Cameron County, Texas, USA. Repels and crossings per survey day were significantly different between pipe and bridge grating wildlife guards (P=0.001) and significantly higher post construction (P=0.002).

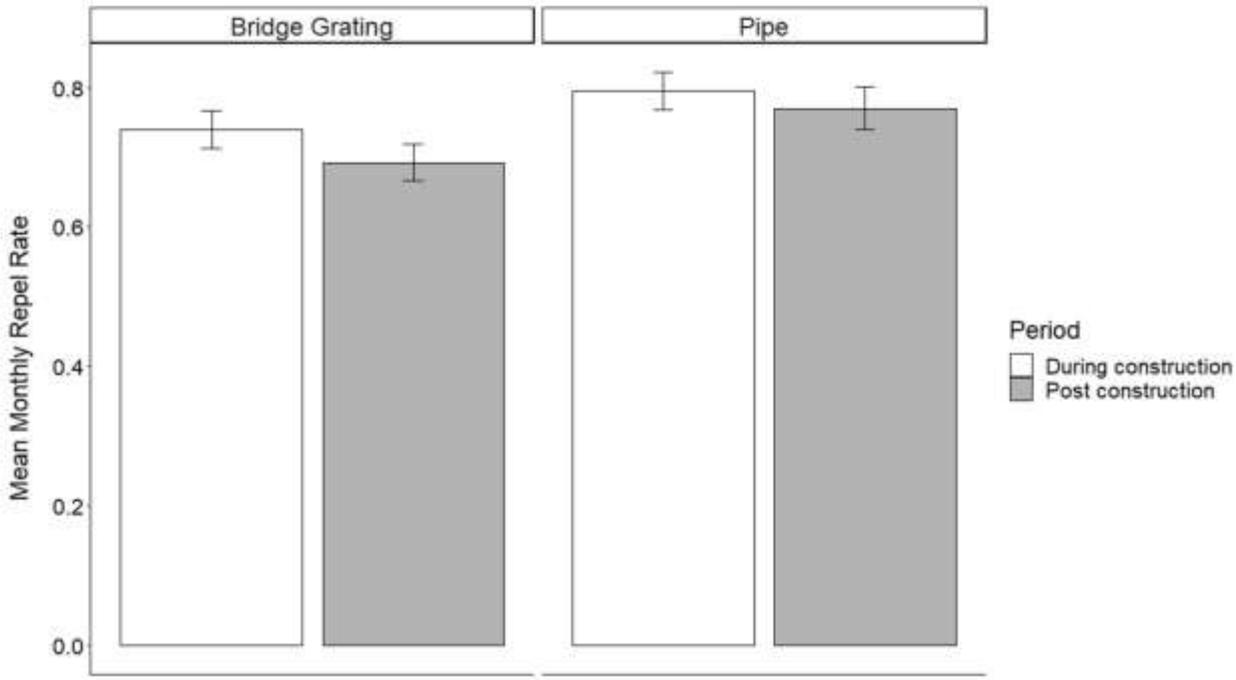

Figure 11. Bar graph showing mean monthly repel rates ± standard error of all species combined at pipe and bridge grating wildlife guards during construction (Apr 2017-May 2018) and post construction (May 2018-May 2019) along State Highway 100 in Cameron County, Texas, USA. Repel rates were significantly higher at pipe wildlife guards (*P*=0.004) but were not significantly different between during construction and post construction (*P*=0.195).

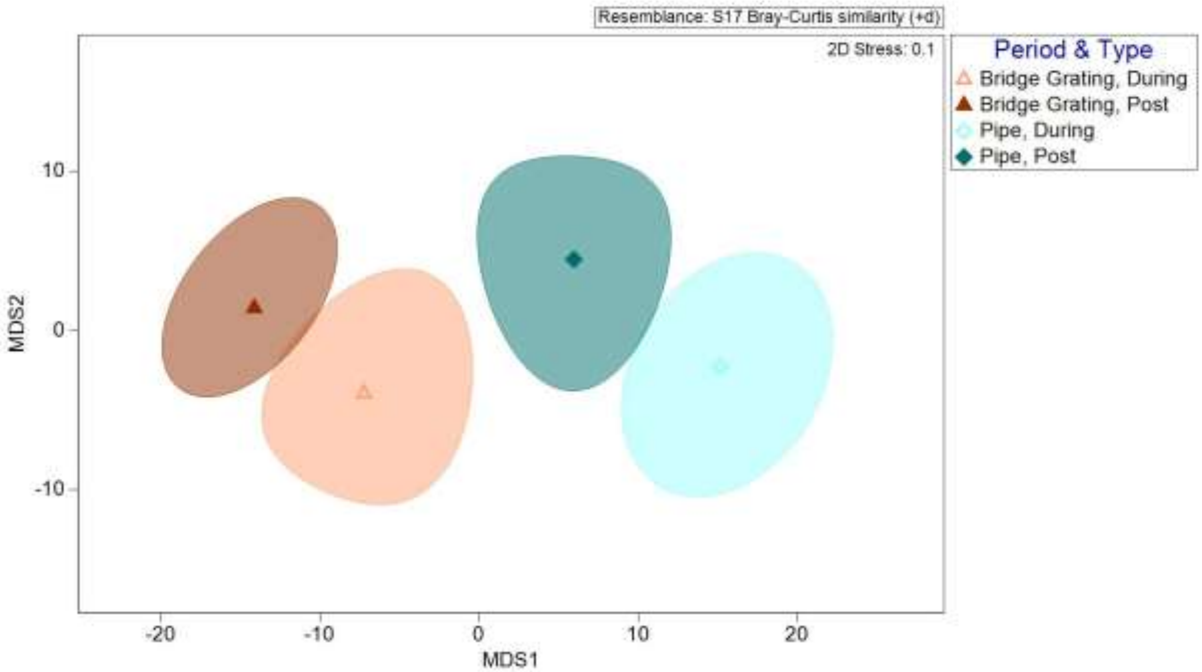

Figure 12. Bootstrapped metric MDS plot showing differences in monthly repel rates between communities observed at each wildlife guard type (*P*=0.012) and between during construction (Apr 2017-May 2018) and post construction (May 2018-May 2019; *P*=0.010) along State Highway 100 in Cameron County, Texas, USA. Differences are indicated by physical distance between bootstrapped averages surrounded by 95% confidence areas.

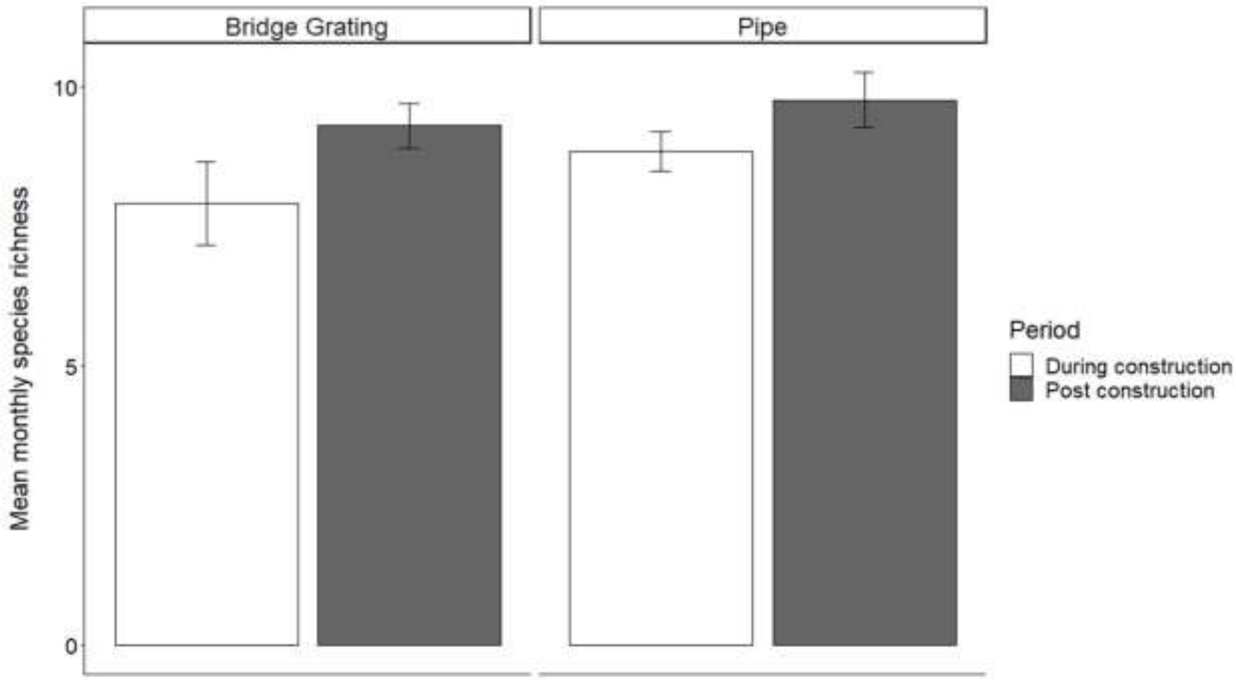

Figure 13. Bar graph showing mean monthly species richness ± standard error at pipe and bridge grating wildlife guards during construction (Apr 2017- May 2018) and post construction (May 2018-May 2019) along State Highway 100 in Cameron County, Texas, USA. Species richness was not significantly different between pipe and bridge grating wildlife guards (*P*=0.197) but were significantly higher post construction (*P*=0.029).

Figure 14. Water inundation at one wildlife crossing structure, located on State Highway 100 in Cameron County, Texas, USA, occurring between January 2018 and May 2019: little to no water (0); intermediate pooling of water (1); full of water (2).

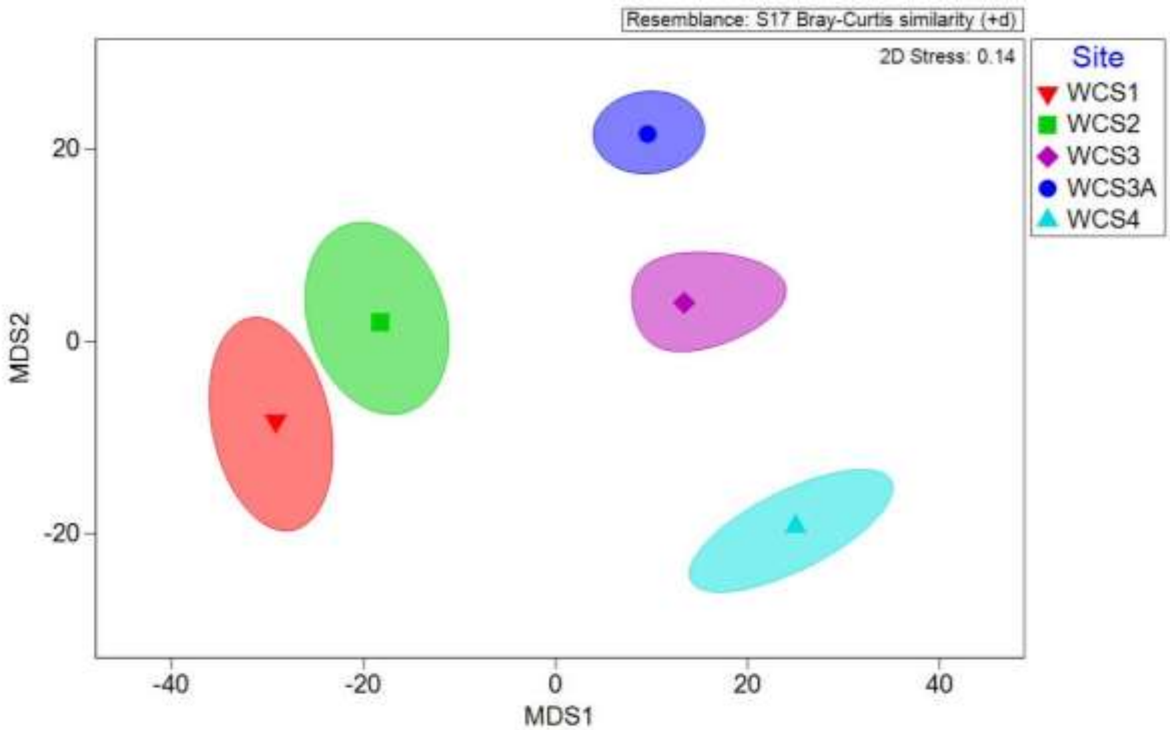

Figure 15. Bootstrapped metric MDS plot showing differences in wildlife communities crossing at all wildlife crossing structures (*P*≤0.001) post construction (May 2018-May 2019) along State Highway 100 in Cameron County, Texas, USA. Differences are indicated by physical distance between bootstrapped averages surrounded by 95% confidence areas.

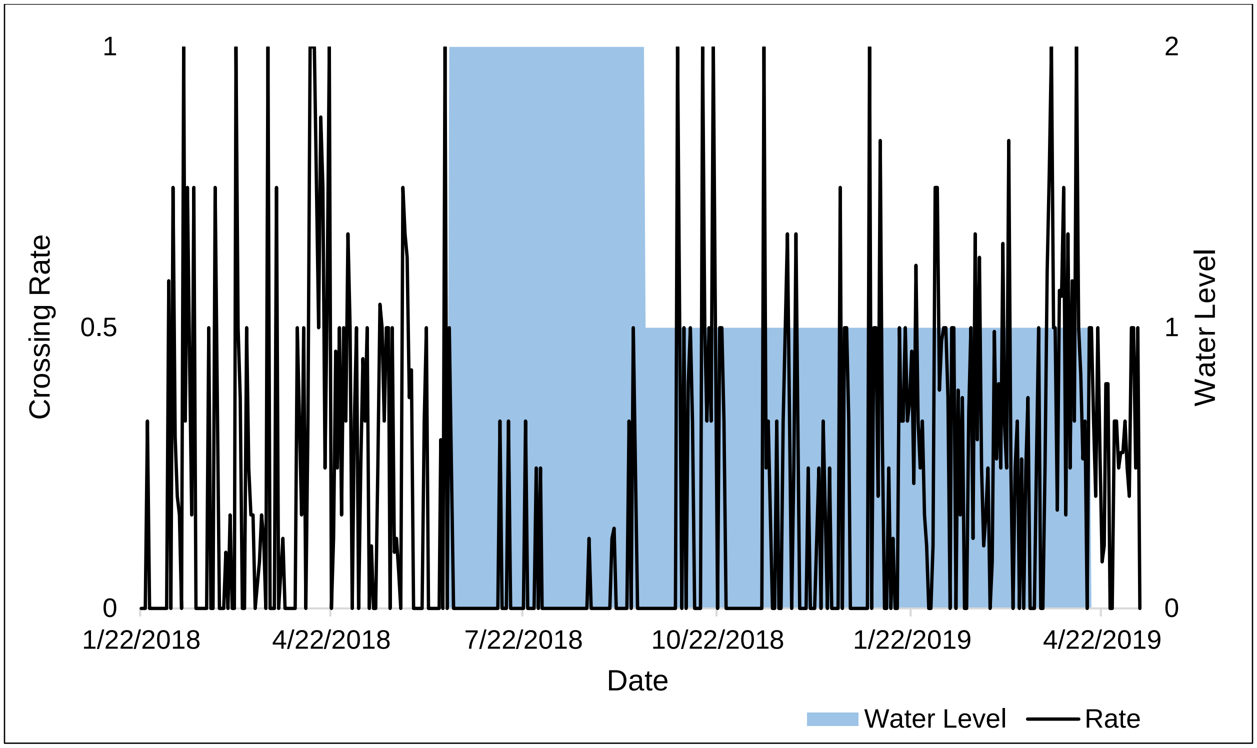

Figure 16. Daily crossing rates (Σ crossings/Σ total occurrences) of all wildlife compared to three assigned water levels at WCS3 for data collected from January 2018 to May2019 in Cameron County, Texas, USA.

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BIOGRAPHICAL SKETCH

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