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**Mammal use of underpasses to cross Route 606 in  
Guacimal, Costa Rica**

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## Mammal use of underpasses to cross Route 606 in Guacimal, Costa Rica

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### ABSTRACT

Roads severely affect the health of ecosystems across the globe by fragmenting habitats, reducing population connectivity, and increasing animal mortality. Wildlife underpasses allow for increased road permeability—the ability for animals to safely cross the road. Despite growing success in other regions, underpass usage in Central America is critically under-researched. In this study, I monitored two dry circular culverts and two unfenced tunnels on Route 606 in Guacimal, Costa Rica, from 22 November to 6 December 2021 using 14 camera traps to assess which species used them to cross. Twelve species used the culverts and tunnels for a total of 108 individual crossings. The tunnels were used, in descending order, by agouti (*Dasyprocta punctata*), opossum (*Didelphis marsupialis*), dog (*Canis familiaris*), armadillo (*Dasyurus novemcinctus*), cat (*Felis catus*), Norway rat (*Dasyopus novemcinctus*), ocelot (*Leopardus pardalis*), squirrel (*Sciurus variegatoides*), tamandua (*Tamandua mexicana*), and coati (*Nasua narica*). The circular tunnel, Tunnel 1, was used more frequently and by a greater diversity of species than observed in the square tunnel, Tunnel 2. The two smaller culverts were used by opossum (*Didelphis marsupialis*), cat (*Felis catus*), rat opossum (*Micoureus alstoni*), and Watson's climbing rat (*Tylomus watsoni*). Culvert 2 was used more frequently; however, Culvert 1 was used by a greater diversity of species. Species such as coyote (*Canis latrans*) and gray fox (*Urocyon cinereoargenteus*) were captured in the surrounding fragments but not using any underpass. This study highlights wildlife underpasses as a critical strategy for biological conservation in Costa Rica though improved road safety and habitat connectivity.

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### KEYWORDS

Camera Trapping, Central America, Habitat Fragmentation, Road Ecology, Wildlife Crossing Structures.

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### INTRODUCTION

Habitat space is essential for each species to survive and maintain a reproducing population. Habitat fragmentation is the process by which a large and continuous area of habitat is divided into two or more smaller fragments that are surrounded by areas inhospitable to the species that reside there (Didham, 2010; Wilson et. al 2015). This fragmentation often occurs when human activity transforms natural areas into roadways, agricultural plots, human settlements, and more, that results in a disconnect between the remaining fragments and often a net loss of habitat (Mullu 2016). The species remaining during and after fragmentation are often at greater risk of inbreeding, reduced diversity, genetic drift, and subsequent extinction due to their isolation (Didham 2010; Dixo et. al. 2009; Haddad et. al., 2015; Wilson et. al 2015). In addition, when individuals attempt to access isolated fragments divided by dangerous roadways, they inevitably increase wildlife-vehicle collisions, which further decreases genetic diversity and endangers motorists (Barbosa et. al. 2020).

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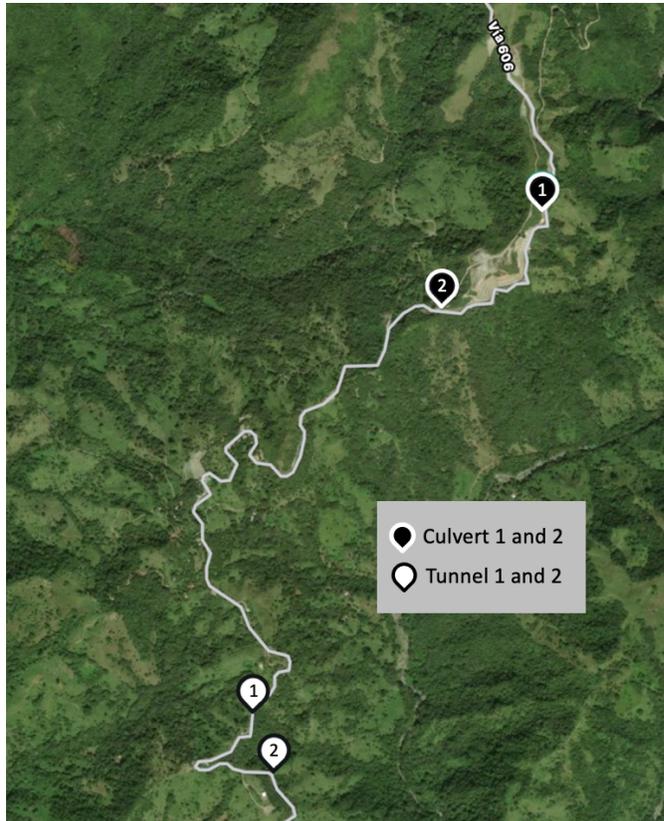
Wildlife crossing structures present one possible mitigation to habitat fragmentation by increasing road permeability—the ability for animals to safely cross the road. An effective crossing

structure allows safe and continuous passage through an inhospitable environment that separates two habitats (Bennet 2003). Wildlife underpasses are structures that allow animals to pass under a road and will generally include both underpass tunnels and culverts in this study, although they both have distinct attributes. An assumption is made when animal crossing structures are implemented that animals will prefer to use them to move between fragmented habitats rather than cross through the inhospitable environment. Despite being in one of the most biologically diverse regions in the world, the use of these underpasses as effective tools for conservation is critically under-studied in Central America (Venegas 2018; Villalobos-Hoffman et. al 2022).

Two unfenced subterranean animal crossing tunnels, one circular and one square, were built in 2016 to mitigate the fragmentation caused by the Route 606 roadway from Guacimal to Monteverde, Costa Rica, based on locals' observations of roadkill and animal crossings (Camacho and Chinchilla 2013). This roadway is particularly treacherous for species moving between fragmented habitats because tourism and travel in the area creates a higher-than-average traffic density (Naranjo-Ureña et. al. 2019). Subterranean tunnels built for animal passage and culverts built for water diversion under roads have been shown to facilitate travel between fragments for various small, medium, and large mammals in South, North, and Central America (Beier 2008; Venegas 2018; Abra et. al 2020; Villalobos-Hoffman et. al. 2022). The two tunnels and two culverts along Route 606, however, had not been previously monitored to find out what animals use them to cross. In this study I investigated differential usage of these wildlife underpasses by local mammal species. I analyzed the effectiveness of these types of underpasses as a tool to mitigate fragmentation and conserve the health of local ecosystems in these locations. Knowing which species use these corridors to cross the road can help determine which species' populations are positively affected by the underpasses and inform future conservation efforts.

## MATERIALS AND METHODS

This study was conducted at the section of Route 606 through la Guaria on the road leading from Monteverde to Guacimal, a section of road that was first paved in 2017 and has frequent traffic (Naranjo-Ureña et. al. 2019). The land fragments bisected by the road are a mixture of pasturelands with secondary and primary growth forests. Culvert 1 and 2 locations: (10.250, -84.839) and (10.246, -84.844). Tunnel 1 and 2 locations: (10.229, -84.851) and, (10.226, -84.851) (Fig. 1).



**Figure 1:** Underpass locations. Satellite view of the underpass locations between Guacimal and Monteverde along Route 606. Black points indicate Culvert 1 and 2. White points indicate circular Tunnel 1 and square Tunnel 2. Map Data: Google (C) 2023 CNES / Airbus, Landsat / Copernicus, Maxar Technologies, U.S. Geological Survey.

Bushnell HD camera traps and mud track stations were used to identify which mammal species are using the wildlife underpasses to cross under Route 606. The camera traps were set on camera mode to 12 megapixels, 3 photo bursts, auto sensitivity, medium shutter speed, and 3-second exposure intervals. At each site, machetes were used to remove vegetation such as tall grass or vines from the two-meter area in front of the camera to reduce misfiring.

Camera traps were installed on 22 November 2021, at one end of two circular culverts (radius 86 and 88cm), and removed on 6 December 2021. Three cameras were positioned facing the entrances of the two larger subterranean tunnels—Tunnel 1 circular (radius 1m) and Tunnel 2 square (height 1m 77cm, width 2m 1cm). One camera was placed at each tunnel on 14 November, to collect a week of preliminary data. In the tunnels, mud made with water and surrounding dirt was smoothed approximately 0.5 by 2 meters at each entrance to record animal tracks as supporting data for the cameras. Six cameras were placed from 22 November to 6 December throughout the land fragments at least 50 meters apart to monitor roadside and fragment species diversity. Memory cards and track stations were replaced every other weekday to ensure they were operational. Any additional evidence of animal presence between site visits was noted, such as roadkill, insect, or other bird presence. The photo data was reliably coded to assess species type and number of crossings by using photo references for identification. A “crossing” by a species

was based on whether that animal was captured going into or out of the underpass. The presence of any species recorded in the track station was corroborated with camera trap footage.

**RESULTS**

I observed twelve mammal species using the culverts and subterranean tunnels to cross under Route 606. Each species photographed using the tunnels or culverts to cross is listed in the tables below. The common opossum (*Didelphis marsupialis*), agouti (*Dasyprocta punctata*), and armadillo (*Dasypus novemcinctus*) were the native species that used the underpasses most frequently (Table 1).

**Table 1:** Sum of total detected crossings by species across all underpasses.

Species	Crossing #
Common Opossum ( <i>Didelphis marsupialis</i> )	30
Agouti ( <i>Dasyprocta punctata</i> )	21
Domestic Dog ( <i>Canis familiaris</i> )	15
Armadillo ( <i>Dasypus novemcinctus</i> )	13
Domestic Cat ( <i>Felis catus</i> )	12
Norway Rat ( <i>Rattus norvegicus</i> )	5
Ocelot ( <i>Leopardus pardalis</i> )	4
Squirrel ( <i>Sciurus variegatoides</i> )	3
Tamandua ( <i>Tamandua mexicana</i> )	2
Rat Opossum ( <i>Micoureus alston</i> )	1
Coati ( <i>Nasua narica</i> )	1
Watson's Climbing Rat ( <i>Tylomys watsoni</i> )	1
Total	108

Tunnel 1 was crossed more frequently and by a greater diversity of species than Tunnel 2. Tunnel 1 was crossed a total of 67 times by ten species, while Tunnel 2 was crossed a total of 24 times by four species (Table 2 and 3).

**Table 2:** Number of detected crossings by species through Tunnel 1.

Tunnel 1	
Species	Crossing #
Agouti ( <i>Dasyprocta punctata</i> )	21
Domestic Dog ( <i>Canis familiaris</i> )	15
Common Opossum ( <i>Didelphis marsupialis</i> )	8
Armadillo ( <i>Dasypus novemcinctus</i> )	7
Ocelot ( <i>Leopardus pardalis</i> )	4
Norway Rat ( <i>Rattus norvegicus</i> )	4
Squirrel ( <i>Sciurus variegatoides</i> )	3
Domestic Cat ( <i>Felis catus</i> )	2
Tamandua ( <i>Tamandua mexicana</i> )	2
Coati ( <i>Nasua narica</i> )	1
Total:	67

**Table 3:** Number of detected crossings by species through Tunnel 2.

Tunnel 2	
Species	Crossing #
Common Opossum ( <i>Didelphis marsupialis</i> )	11
Armadillo ( <i>Dasypus novemcinctus</i> )	6
Domestic Cat ( <i>Felis catus</i> )	6
Norway Rat ( <i>Rattus norvegicus</i> )	1
Total:	24

Culvert 2 was crossed more frequently than Culvert 1; however, Culvert 1 was used by a greater diversity of species. Culvert 1 was crossed a total of five times by three species, while culvert 2 was crossed a total of 12 times by two species (Table 4 and 5).

**Table 4:** Number of detected crossings by species through Culvert 1.

Culvert 1	
Species	Crossing #
Common Opossum ( <i>Didelphis marsupialis</i> )	3
Watson's Climbing Rat ( <i>Tylomys watsoni</i> )	1
Rat Opossum ( <i>Micoureus alstoni</i> )	1
Total:	5

**Table 5:** Number of detected crossings by species through Culvert 2.

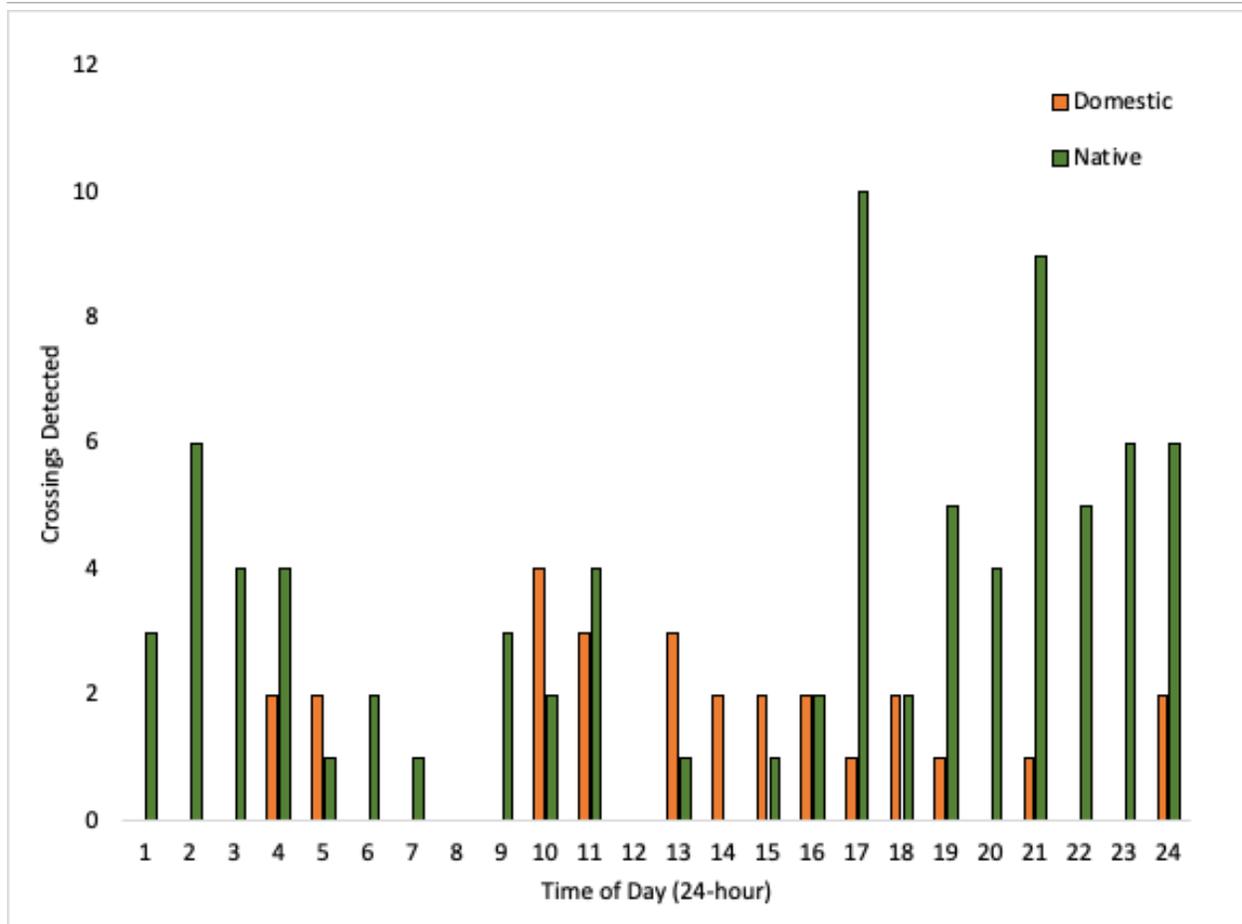
Culvert 2	
Species	Crossing #
Common Opossum ( <i>Didelphis marsupialis</i> )	8
Domestic Cat ( <i>Felis catus</i> )	4
Total:	12

Some animals were documented in the surrounding land fragments but were not observed using one or more of the underpasses. Species such as the Olive Sparrow (*Arremonops rufivirgatus*), cow (*Bos taurus*), Black Iguana (*Ctenosaura similis*), Swainson’s Thrush (*Catharus ustulatus*), spiny pocket mouse (*Heteromyidae*), rice rat (*Oryzomys*), Watson’s climbing rat (*Tylomis watsoni*), coyote (*Canis latrans*), chicken (*Gallus Domesticus*), and gray fox (*Urocyon cinereoargenteus*) were captured by camera trap in the land fragments but not observed using any of the underpasses to cross. Some species, for instance, agouti (*Dasyprocta punctata*) and tamandua (*Tamandua Mexicana*) were documented using only Tunnel 1 and not Tunnel 2 to cross (Table 6).

**Table 6:** Presence of species by detected location. “X” indicates that a species was photographed using the underpass or in the land fragment. “E” indicates that a species was observed using the underpass and is expected to be in the land fragment.

Species:	Culvert 1	Culvert 2	Tunnel 1	Tunnel 2	Fragment
Agouti ( <i>Dasyprocta punctata</i> )			X		X
Armadillo ( <i>Dasypus novemcinctus</i> )			X	X	X
Black Iguana ( <i>Ctenosaura similis</i> )					X
Chicken ( <i>Gallus domesticus</i> )					X
Coati ( <i>Nasua narica</i> )			X		X
Common Opossum ( <i>Didelphis marsupialis</i> )	X	X	X	X	X
Cow ( <i>Bos taurus</i> )					X
Coyote ( <i>Canis latrans</i> )					X
Domestic Cat ( <i>Felis catus</i> )		X	X	X	E
Domestic Dog ( <i>Canis familiaris</i> )			X		X
Gray Fox ( <i>Urocyon cinereoargenteus</i> )					X
Norway Rat ( <i>Rattus norvegicus</i> )			X	X	X
Ocelot ( <i>Leopardus pardalis</i> )			X		E
Olive Sparrow ( <i>Arremonops rufivirgatus</i> )					X
Rat Opossum ( <i>Micoureus alstoni</i> )	X				E
Rice Rat ( <i>Oryzomys</i> )					X
Spiny Pocket Mouse ( <i>Heteromyidae</i> )					X
Squirrel ( <i>Sciurus variegatoides</i> )			X		X
Swainson's Thrush ( <i>Catharus ustulatus</i> )					X
Tamandua ( <i>Tamandua mexicana</i> )			X		X
Watson's Climbing Rat ( <i>Tylomis watsoni</i> )	X				E

Activity of native species using the corridors was most common at night, while activity of domesticated species using the corridors was most common during the day (Fig. 2).



**Figure 2:** Animal Activity Across the 24-Hour Day. Compares the combined number of crossings by native and domesticated species of mammals across the day. Green represents crossings by native mammals and orange represents crossings by domestic dogs and cats.

## DISCUSSION

A total of twelve species used the underpasses to cross Route 606 a combined 108 times over three weeks of observation (Table 1). The use of the culverts by smaller-sized mammals aligns with one study in Alberta, Canada, that found underpasses such as culverts increase road permeability (Clevenger 2002). The use of the tunnels by small, medium, and large mammals aligns with similar studies in North, South, and Central America (McDonald and St Clair 2004; Clevenger and Waltho 2005). It was predicted that black iguanas (*Ctenosaura similis*) and other reptiles present in the fragments would not use the corridors, since the warmer road pavement would be more attractive for sunbathing than the shaded underpasses (Table 6). As most of the native species to use the corridors are nocturnal, it follows that they are most frequented during the night compared to domesticated species (Fig. 2). These four underpasses are effective in the sense that animals used them to cross under Route 606 rather than across the road itself. Although

all of the underpasses were successfully utilized, some were frequented more often and by different species than others.

Although less than 400 meters apart, Tunnel 1 has noticeably more activity and diversity of species compared to Tunnel 2 (Table 2 and 3). This difference could be due to the vegetation in the fragments directly surrounding the tunnels. Tunnel 1 has considerably more tree cover surrounding the entrances of the corridor compared to Tunnel 2 (Appendix 1 and 2). Culvert 2 also had more vegetation and was used more frequently compared to Culvert 1. As a general principle, wildlife crossing structures will only be as effective as the land and resources surrounding them (Clevenger et al. 2005). Although this study was not designed for comparative analysis, one way to increase the efficacy of wildlife underpasses could be to increase the proximity of vegetation in the surrounding land.

Structural shape is another possibility to account for the difference in diversity and abundance between the two tunnels. For instance, in this study, Tamandua (*Tamandua mexicana*) only crossed using the circular Tunnel 1 (Appendix 3), despite being photographed less than 5 meters from the entrance of the square Tunnel 2 (Appendix 4) (Table 2 and 3). This species might prefer the more circular structure and felt more comfortable crossing through it, however, others of the same species in Costa Rica have been recorded using square underpasses to cross roadways (Venegas 2018 and Villalobos-Hoffman et al. 2022). In addition to unequal species usage between underpasses, some vertebrates observed in the fragments surrounding the underpasses were not observed using them during the period of the study. Generally, no single underpass will allow all species to cross a road, because each species has a preference for crossing structure shape and design (Clevenger et al. 2001; McDonald and St Clair 2004; Clevenger and Waltho 2005; Mata et al. 2005). This study's length and design do not provide enough data to explain why tamandua (*Tamandua mexicana*) and other species such as coyote (*Canis latrans*) and gray fox (*Urocyon cinereoargenteus*) did not use any of the underpasses even though similar species have used them in other regions (Clevenger et al. 2001).

The presence of barbed wire fences across both entrances of the underpasses may also be influencing which species cross the tunnels (Appendix 5 and 6). Larger organisms such as peccary (*Pecari tajacu*) and white-tailed deer (*Odocoileus virginianus*) that are present in the region would have difficulty passing through the fences to access the corridors (Timm and LaVal 2018). The fences were established by the landowners following the tunnels' construction to prevent cows (*Bos taurus*) from crossing pastures. The fences at Tunnel 2 have fewer barbed lines and are more degraded than those at Tunnel 1 which may have been another reason Tunnel 2 saw less activity. In areas where underpasses connect fragments used for agriculture, alternative methods for livestock diversion that allow for other larger mammals to access the tunnels should be explored and recommended.

Vertebrates are not the only organisms using the underpasses. Primarily two species of insects, army ants (*Eciton burchellii*) and termites (*Atta cephalotes*), were recorded using the underpasses to cross under Route 606 in foraging trails. These observations occurred during in-person visitations to the underpasses; however, they were not included in the results because this study was not designed to quantify use by insects. Insect roadkill and decreasing insect abundance

are threats to the health of global ecosystems, and a study surveying the use of these underpasses to conserve insect populations would be beneficial for conservation efforts (New 2015)

Because road infrastructure inevitably grows with the human population, it is vital for transportation agencies and urban planners to consider the ways this growth could harm local ecosystems. Continued investment in wildlife crossing structures will likely help conserve species threatened by the habitat fragmentation caused by roadways. Future studies would benefit from surveying a greater number of underpasses with a variety of different attributes over longer time periods. This research is necessary to assess what features would make animal crossing structures more effective for a greater variety of species. The data from this study support the claim that the underpasses built under Route 606 have helped reduce the impacts of fragmentation by allowing mammal species to travel between fragments. Further studies will help inform the optimal design of future crossing structures.

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## APENDICES



**Appendix 1:** Aerial view of Tunnel 1. Underpass is indicated with an arrow. Photo taken by Randy Chinchilla on 30 November 2021.



**Appendix 2:** Aerial view of Tunnel 2. Underpass is indicated with an arrow. Photo taken by Randy Chinchilla on 30 November 2021.



**Appendix 3:** Entrance of Tunnel 1. Frank Joyce and Orlando Méndez pictured at the exit.



**Appendix 4:** Entrance of Tunnel 2.



**Appendix 5:** Fence at Tunnel 1.



**Appendix 6:** Fence at Tunnel 2.



**Appendix 7:** Entrance of culvert 1. Wooden debris from cliff ladder shown.

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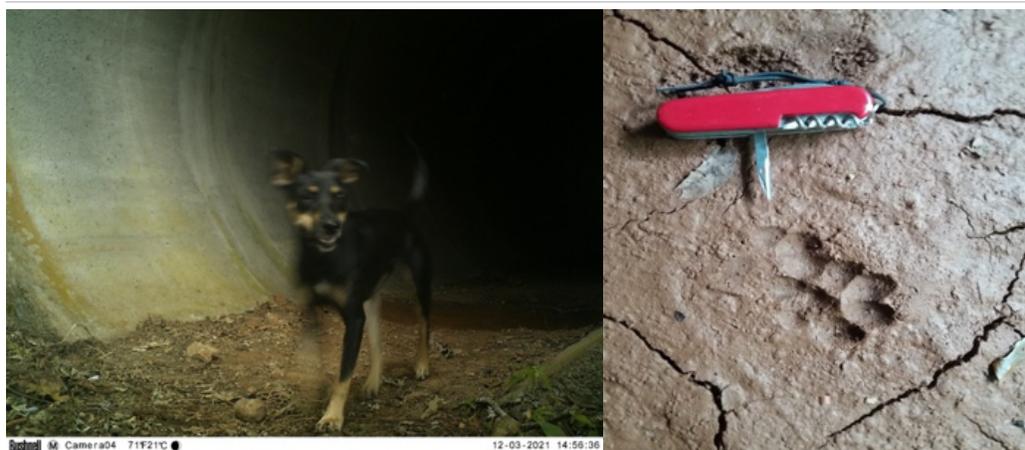
**Appendix 8:** Entrance of culvert 2.

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The following are examples of species that were identified with the corresponding camera trap and (if applicable) footprint observation.



**Appendix 9:** Coyote (*Canis latrans*) pictured in fragment



**Appendix 10:** Domestic Dog (*Canis familiaris*) pictured in Tunnel 1



**Appendix 11:** Black Iguana (*Ctenosaura similis*) pictured in fragment



**Appendix 12:** Agouti (*Dasyprocta punctata*) pictured in Tunnel 1



**Appendix 13:** Armadillo (*Dasypus novemcinctus*) pictured in Tunnel 2



**Appendix 14:** Common Opossum (*Didelphis marsupialis*) pictured in Tunnel 2



**Appendix 15:** Domestic Cat (*Felis catus*) pictured in Tunnel 2



**Appendix 16:** Ocelot (*Leopardus pardalis*) pictured in Tunnel 1



**Appendix 17:** Coati (*Nasua narica*) pictured in Tunnel 1



**Appendix 18:** Squirrel (*Sciurus variegatoides*) pictured in Tunnel 1



**Appendix 19:** Tamandua (*Tamandua mexicana*) pictured in Tunnel 1



**Appendix 20:** Gray Fox (*Urocyon cinereoargenteus*) pictured in fragment.