



WILDLIFE ROADKILL IN THE SURROUNDINGS OF EMAS NATIONAL PARK, CERRADO BIOME, BRAZIL

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Abstract: Roadkill is a serious threat to biodiversity conservation especially when roads are near natural habitats of wildlife, such as the Emas National Park (ENP), a Protected Area in Mid-west Brazil in the Cerrado Biome. We aimed to identify the species killed on a stretch of the GO-341 highway that is tangent to the ENP. We investigated if roadkills were influenced by seasonality, sugar cane harvest and by the activity pattern of the animals (diurnal/nocturnal). We also analyzed if roadkills were aggregated in space, where the roadkill hotspots were located, if they were influenced by seasonality, and if they were different for the most abundant species. The highway was monitored with a vehicle, in the morning, at a speed of 40 to 60 km/hour, daily, with two observers. We covered a total of 4,230 km during the 90-day monitoring period, which included dry and rainy seasons. We recorded 132 wild animals' roadkills: 67 birds (51 %), 60 mammals (45 %) and 5 reptiles (4 %). We identified 22 vertebrate species, including 13 birds, six mammals and three reptiles. The roadkill rate was 0.03 animals/km/day. During the dry season, the blue-and-yellow macaw (*Ara ararauna*) had higher roadkill rates. The crab-eating fox (*Cerdocyon thous*) was the only species with higher roadkill rates during the sugarcane harvest. Most birds killed had diurnal habits, for mammals, no difference was observed concerning the activity pattern. Bird roadkills were aggregated considering the entire data set, as well as for the dry and rainy seasons separately. Mammal roadkills were aggregated only when considering the entire data set. The location of bird roadkill hotspots differed between seasons. The roadkill hotspot location of birds and mammals highly overlapped those observed for the two most abundant species of these taxa, respectively, *Ara ararauna* and *Euphractus sexcinctus*.

Keywords: Parque Nacional das Emas; Road Ecology; roadkill hotspots; sugarcane harvest; wildlife-vehicle collision.

INTRODUCTION

Although roads are extremely important to human beings, they have a series of effects on the environment (Laurance *et al.* 2009). The

majority of them are typically deleterious to biodiversity, as increased pollutants near these sites, habitat fragmentation, higher predation around the road, hunting facilitation, repulsion effect, barrier effect, invasion of exotic species,

roadkill, and others (Laurance *et al.* 2009, van der Ree *et al.* 2015). A range of factors is related to wildlife roadkill. Regarding road characteristics, roadkill rates increase with increasing vehicles flow, road density and road width. Paved roads and slope stretches also have higher roadkill rates (Laurance *et al.* 2009, Grilo *et al.* 2010, Gunson *et al.* 2011). Regarding landscape, we can mention the area of natural cover, the distance to the urban perimeter, to a river and/or to native vegetation as important variables (Gunson *et al.* 2011). Non-natural landscapes, such as pastures and monocultures, may also influence roadkill, since these areas still sustain a relatively high biodiversity of medium to large-sized mammals (Magioli *et al.* 2016) and their harvest operations may push animals into highways in search for new shelter. Some authors have suggested that harvest may increase roadkill rates due an increase in vehicles flow (Miranda 2017). Species specific characteristic also influence roadkill, such as home range size, body mass, sexual maturity, diet, reproduction-related characteristics, and behavior (Rytwinski & Fahrig 2015). Identifying the species that are most roadkilled can also help understand when those accidents happen more frequently. For instance, we expect to see more roadkill of mammals during the night as the driver and animals' sight is less effective. In contrast, bird roadkill may be more often during the day, because most of these animals are diurnal. Climate and seasonality also seem to influence roadkill. Reptiles and amphibians have higher roadkill rates during the rainy/hot season, which is the period they are more active (D'Amico *et al.* 2015, Garriga *et al.* 2017). For mammals and birds, some authors believe that their roadkill rates are higher during the dry season, when their movements increase for foraging (Bueno & Almeida 2010, Cunha *et al.* 2010).

Due to spatial, temporal, species-specific and road characteristics, roadkill is usually aggregated (Coelho *et al.* 2008, Cáceres *et al.* 2012, Teixeira *et al.* 2013). In order to decrease roadkill rates, road managers need to identify priority sites for road mitigation, as those with greater roadkill rates (Grilo *et al.* 2010). Previous research have identified that roadkill aggregations (also called roadkill hotspots) are not different between seasons, although it may be for some specific

mammal species (Cáceres *et al.* 2012). Roadkill hotspots may vary between taxa (Teixeira *et al.* 2013, Miranda *et al.* 2020), and through the years (Santos *et al.* 2017). The hotspots location may change when the most abundant species are removed from the analysis (Miranda *et al.* 2020). Nonetheless, the influence of seasonality on hotspots location has not been evaluated yet.

Areas that should be a refuge for wildlife are also under road effects. More than half (62 %) of the federal protected areas in Brazil are intersected by highways and 72 % are indirectly affected by roads, accounting for 5.6 % of the total protected areas (Lima 2013). In the Cerrado biome, a biodiversity hotspot for conservation (Myers *et al.* 2000), public protected areas cover only 7.5 % of the biome and the Brazilian government requires 20 % of private lands to be set aside for conservation (Brasil 2012). While Brazil was lowering Amazon deforestation rates annually, Cerrado has lost 46 % of its native vegetation coverage, and as little as 19.8 % remains undisturbed (Strassburg *et al.* 2017). Projections report that 31–34 % of the remaining Cerrado is likely to be cleared by 2050 (Strassburg *et al.* 2017). This represents a huge biodiversity loss, since Cerrado comprises 2,373 species of vertebrates, 433 (18.2 %) of them are endemic, and at least 199 species of vertebrates are in the Brazilian Red List of threatened species (Sawyer *et al.* 2016). Therefore, immediate measures are necessary to preserve what we still have left in this biome. Thus, we aimed to identify the species killed on a highway that is tangent to a Cerrado protected area, the Emas National Park (ENP), in Goiás state, Midwest Brazil. We investigated if roadkill was influenced by seasonality, sugarcane harvest, and by animals' activity pattern (diurnal/nocturnal). We also analyzed if roadkills were aggregated in space, where these aggregations were localized, if they were influenced by seasonality. Roadkill hotspots of the most abundant bird and mammal species were also investigated and tested if they were located at the same highway stretches as all other bird and mammal species, and if by removing these abundant species from the analysis, bird and mammal roadkill hotspots location would change.

MATERIAL AND METHODS

Study area

We monitored the stretch of GO-341 highway that is tangent to the limits of the ENP, a Protected Area listed as a UNESCO World Heritage Site (IBAMA & CEBRAC 2004), located in the Southeast of Goiás state, Brazil, on the borders of Mato Grosso do Sul and Mato Grosso states (Figure 1). The ENP covers 1,320 square kilometers (131,386 ha) of Cerrado savannah and protects 85, 353 and 88 species of mammals, birds and reptiles, respectively (Rodrigues *et al.* 2002, IBAMA & CEBRAC 2004) among the 251, 837 and 262 species of these taxa listed for the Cerrado biome (Silva 1995, Paglia *et al.* 2012, Sawyer *et al.* 2016).

We monitored 47 kilometers of the GO-341 highway from km 65 (17°45'12.41"S; 52°56'16.11"W) to km 112 (18°04'37.41"S; 53°8'2.25"W, datum WGS84). It is a paved two-lane highway without road shoulder in all its extension. The vehicles flow is approximately of 700 vehicles per day

(Carvalho-Roel *et al.* 2019). The study area is in one of the most productive agricultural areas in Brazil and its surrounding landscape is dominated by annual (soybean and corn) and perennial (sugarcane, mechanized harvest) crops (IBAMA & CEBRAC 2004). According to the Köppen classification, the region has a tropical savanna climate (Aw), with two well-defined seasons, dry during the winter, and rainy in the summer (IBAMA & CEBRAC 2004). In the rainy season, the average temperature of the hottest month ranges from 24 °C to 26 °C. Meanwhile, the average temperatures of the coldest month ranges between 15 °C and 24 °C. The total annual rainfall ranges between 1,200 mm and 3,000 mm.

Data collection

We monitored the GO-341 highway for 90 days. We covered 2,820 km in 60 days during the dry season (from May to August 2017) and 1,410 km in 30 days during the rainy season (from December 2017 to January 2018). The total sampling effort was 4,230

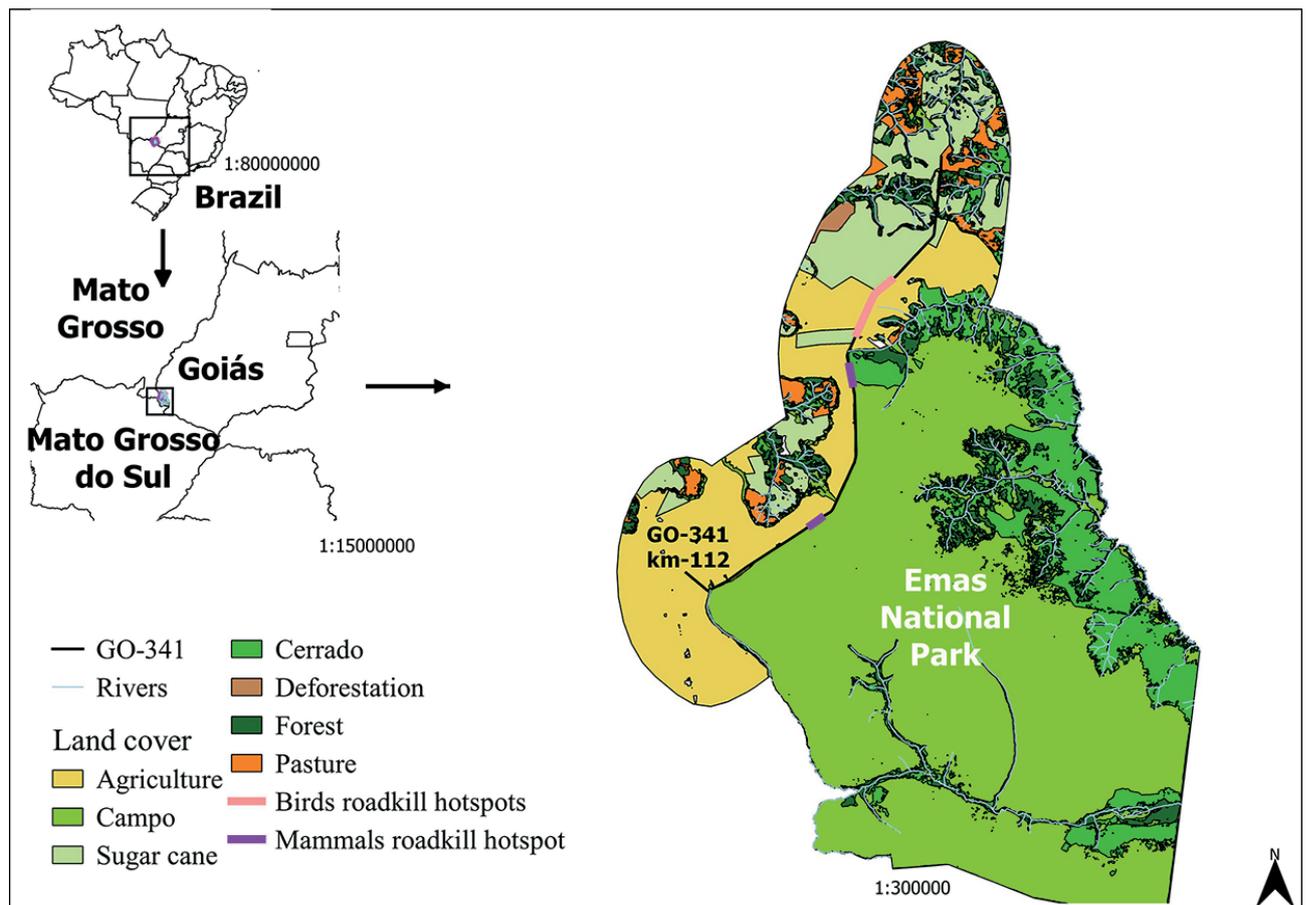


Figure 1. Location of the GO-341 highway in Goiás state, Brazil, indicating the stretch monitored (km 65 to km 112) and the surrounding landscape within a 10 km buffer, highlighting the Emas National Park and birds and mammals roadkill hotspots location. Source: NGO Oréades-Geoprocessing Nucleus (2017).

km. In the dry season, 34 sampling days coincided with the sugarcane harvest period. In this way, it was possible to compare the fauna roadkill rates with the sugarcane harvest in the study area (from July to August 2017) and without it (from May to June 2017). There was no harvest during the rainy season. The highway was monitored daily (consecutive days, skipping Sundays), from 07:00 h until approximately 09:00 h, in one direction (from km 65 to km 112). Two observers performed the survey with a vehicle moving at a speed of 40 to 60 km/h.

The locations in which we found wild animal roadkills were georeferenced. The animals were photographed, identified at species level (when possible), removed from the highway, and placed in its margin in order to avoid count replications or roadkills of opportunistic predators and scavengers. We identified mammals according to Reis *et al.* (2010), birds according to Sigrist (2014) and Piacentini *et al.* (2015), and reptiles based on Bérnils & Costa (2015). In order to classify the animals' activity pattern we used the same references. Such comparison was not possible for reptiles because there were few roadkill records.

Data analysis

The roadkill rate (RK) was calculated by dividing the total number of roadkills recorded by the total number of kilometers monitored. The Mann-Whitney test (U) (significance level of 95 %) was used to evaluate if RK differed between sugarcane harvest and non-harvest periods, rainy and dry season, and diurnal and nocturnal animals. We analyzed all taxa (species and classes) with at least ten records, separately. Regarding diurnal animals, only mammals and birds were analyzed. For these analysis, we used the software R 3.6.1 (R Core Team 2019).

We used the Siriema 2.0 software to verify whether there were stretches of the highway with higher roadkill incidence (Coelho *et al.* 2014). The 2D Ripley K-Statistics test was used to identify the radius in which there were roadkill aggregations. This test evaluates the occurrence of roadkill aggregation in different radius, starting with the initial radius defined by the user and increasing its size according to the values defined as radius increment, until the radius encompasses the entire highway. We used an initial radius of 500 m, with

a radius increment of 500 m, 1000 simulations and a confidence interval of 95 %. We performed the 2D HotSpot Identification in order to identify the stretches of the highway where roadkill hotspots were located. In this test, we used a radius of one km (the smallest significant radius according to the 2D Ripley K-Statistics results), 1000 simulations, a confidence interval of 95 %, and we split the road into 94 segments of 500 m each. The analyses were performed for birds and mammals separately. We investigated if roadkill hotspots differed between rainy and dry seasons by analyzing the data from both seasons together (the 90-days survey) and separately. We also tested if the roadkill hotspots of the most abundant bird and mammal species were located at the same highway stretches as all the other bird and mammal species, respectively, and if removing these species from the analysis, the roadkill hotspots would change.

RESULTS

During the monitoring, 132 wild animal roadkills were recorded, 67 birds (51 %), 60 mammals (45 %) and five reptiles (4 %). We identified 13 species of birds, six mammals and three reptiles (Table 1, Figure S1). The RK was 0.031 animals/km/day. The blue-and-yellow macaw (*Ara ararauna*) had the highest RK (0.008 animals/km/day), followed by the six-banded armadillo (*Euphractus sexcinctus*) (0.007 animals/km/day) and the crab-eating fox (*Cerdocoyon thous*) (0.003 animals/km/day).

There was no significant difference in RK between seasons, 0.029 and 0.034 animals/km/day for the dry and rainy season, respectively (U = 948, p = 0.67) (Table 1). The blue-and-yellow macaw suffered more roadkills in the dry season, 0.008 animals/km/day, and no roadkill during the rainy season (U = 555, p < 0.001). Sugarcane harvest (RK = 0.029 animals/km/day) and non-harvest periods (RK = 0.030 animals/km/day) did not show significant difference in roadkill numbers (U = 426, p = 0.81). The RK was significantly higher during the sugarcane harvest than non-harvest period only for the crab-eating fox (U = 529.5, p = 0.036) (Figure S2). Diurnal birds had a significant higher RK (0.013 animals/km/day) than nocturnal ones (0.002 animals/km/day) (U = 5289, p < 0.000). Mammals roadkills were not significantly different between nocturnal (RK = 0.007 animals/km/day)

Table 1. Identification and number of the roadkilled animals (N) during the dry (DS) and rainy (RS) seasons in 2017 and 2018, on GO-341 highway, km 65 to km 112, Goiás state, Brazil.

Species	Common name	N (90-days)	N (DS)	Roadkill rate (ind/km/ day) (DS)	N (RS)	Roadkill rate (ind/km/ day) (RS)
Birds						
<i>Tyto furcata</i> (Temminck, 1827)	american barn owl	3	1	0.0004	2	0.0014
<i>Athene cunicularia</i> (Molina, 1782)	burrowing owl	5	4	0.0014	1	0.0007
<i>Crotophaga ani</i> Linnaeus, 1758	smooth-billed ani	3	2	0.0007	1	0.0007
<i>Ara ararauna</i> (Linnaeus, 1758)	blue-and-yellow macaw	36	36	0.0128	0	0.0000
<i>Eupsittula aurea</i> (Gmelin, 1788)	peach-fronted parakeet	1	0	0.0000	1	0.0007
<i>Geranoaetus albicaudatus</i> (Vieillot, 1816)	white-tailed hawk	1	1	0.0004	0	0.0000
<i>Caracara plancus</i> (Miller, 1777)	southern caracará	1	0	0.0000	1	0.0007
<i>Cariama cristata</i> (Linnaeus, 1766)	red-legged seriema	2	1	0.0004	1	0.0007
<i>Rhea americana</i> (Linnaeus, 1758)	greater rhea	3	1	0.0004	2	0.0014
<i>Columbina talpacoti</i> (Temminck, 1810)	ruddy ground-dove	3	0	0.0000	3	0.0021
<i>Vanellus chilensis</i> (Molina, 1782)	southern lapwing	1	0	0.0000	1	0.0007
<i>Volatinia jacarina</i> (Linnaeus, 1766)	blue-black grassquit	1	0	0.0000	1	0.0007
<i>Coragyps atratus</i> (Bechstein, 1793)	black vulture	1	0	0.0000	1	0.0007
Non-identified birds		6	2	0.0007	4	0.0028
Total number of bird roadkills		67	48	0.0170	19	0.0135
Reptiles						
<i>Ameiva ameiva</i> (Linnaeus, 1758)	giant ameiva	1	0	0.0000	1	0.0007
<i>Crotalus durissus</i> Linnaeus, 1758	rattlesnake	1	1	0.0004	0	0.0000
<i>Boa constrictor</i> Linnaeus, 1758	red-tailed boa	1	0	0.0000	1	0.0007
Non-identified snakes		2	0	0.0000	2	0.0014
Total number of reptile roadkills		5	1	0.0004	4	0.0028

Table 1. Continues on next page...

Table 1. ...continued

Species	Common name	N (90-days)	N (DS)	Roadkill rate (ind/km/ day) (DS)	N (RS)	Roadkill rate (ind/km/ day) (RS)
Mammals						
<i>Cerdocyon thous</i> (Linnaeus, 1766)	crab-eating fox	15	10	0.0035	5	0.0035
<i>Lycalopex vetulus</i> Lund, 1842	hoary fox	3	1	0.0004	2	0.0014
<i>Conepatus semistriatus</i> (Boddaert, 1785)	striped hog-nosed skunk	6	5	0.0018	1	0.0007
<i>Didelphis albiventris</i> Lund, 1840	white-eared opossum	4	1	0.0004	3	0.0021
<i>Euphractus sexcinctus</i> (Linnaeus, 1758)	six-banded armadillo	30	17	0.0060	13	0.0092
<i>Tamandua tetradactyla</i> (Linnaeus, 1758)	southern tamandua	2	0	0.0000	2	0.0014
Total number of mammal roadkills		60	34	0.0121	26	0.0184
Number of roadkill records		132	83	0.0294	49	0.0348
Number of species found roadkilled		32	13		19	

and diurnal animals (RK = 0.007 animals/km/day) (U = 4141.5, p = 0.74).

Roadkill aggregation was observed for birds either for the whole data set (Figure 2a), or for dry and wet seasons separately (Figures 2b and c). Conversely, mammal roadkills were aggregated in small radius when we analyzed the entire data set (Figure 2f), but not within seasons (Figures 2g and h). We also observed roadkill aggregation for *A. ararauna* and *E. sexcinctus* (Figures 2d and i). Roadkill of birds excluding *A. ararauna* counts and mammals excluding *E. sexcinctus* counts were not aggregated (Figures 2e and j).

DISCUSSION

The RK observed in this study of 0.03 animals/km/day should not be considered low when compared to other studies. Carvalho *et al.* (2015) and Veloso (2015), on a Cerrado highway whose vehicle flow was approximately 17 times greater than that of the highway in this study, obtained a RK of 0.051 animals/km/day, during a weekly monitoring. In

a transition area between Amazon and Cerrado, Brum *et al.* (2017) found 0.036 animals/km/day on a highway with nearly three times greater vehicle flow than that of this study, also during a weekly monitoring. The vehicles flow of approximately 700 vehicles per day on the studied highway (Carvalho-Roel *et al.* 2019) should also contribute for the observed RK, since, in theory, RK are positively correlated to vehicles flow (Jacobson *et al.* 2016). Another fact that may have contributed to the observed RK is the applied methodology. Daily monitoring would result in lower RK for medium to large-sized animals when compared to weekly surveys. Carcass removal can explain this difference because medium to large-sized animals are the groups less affected by carcass removal in roadkill monitoring. In the Atlantic Forest, 65 % of the carcasses of small body-sized animals and 21 % of large body-sized animals disappear within 24 hours (Teixeira *et al.* 2013). In the Cerrado biome, 27 % of the carcasses seems to persist after four days (Santos *et al.* 2016). Nevertheless, some carcasses of medium to large-sized animals

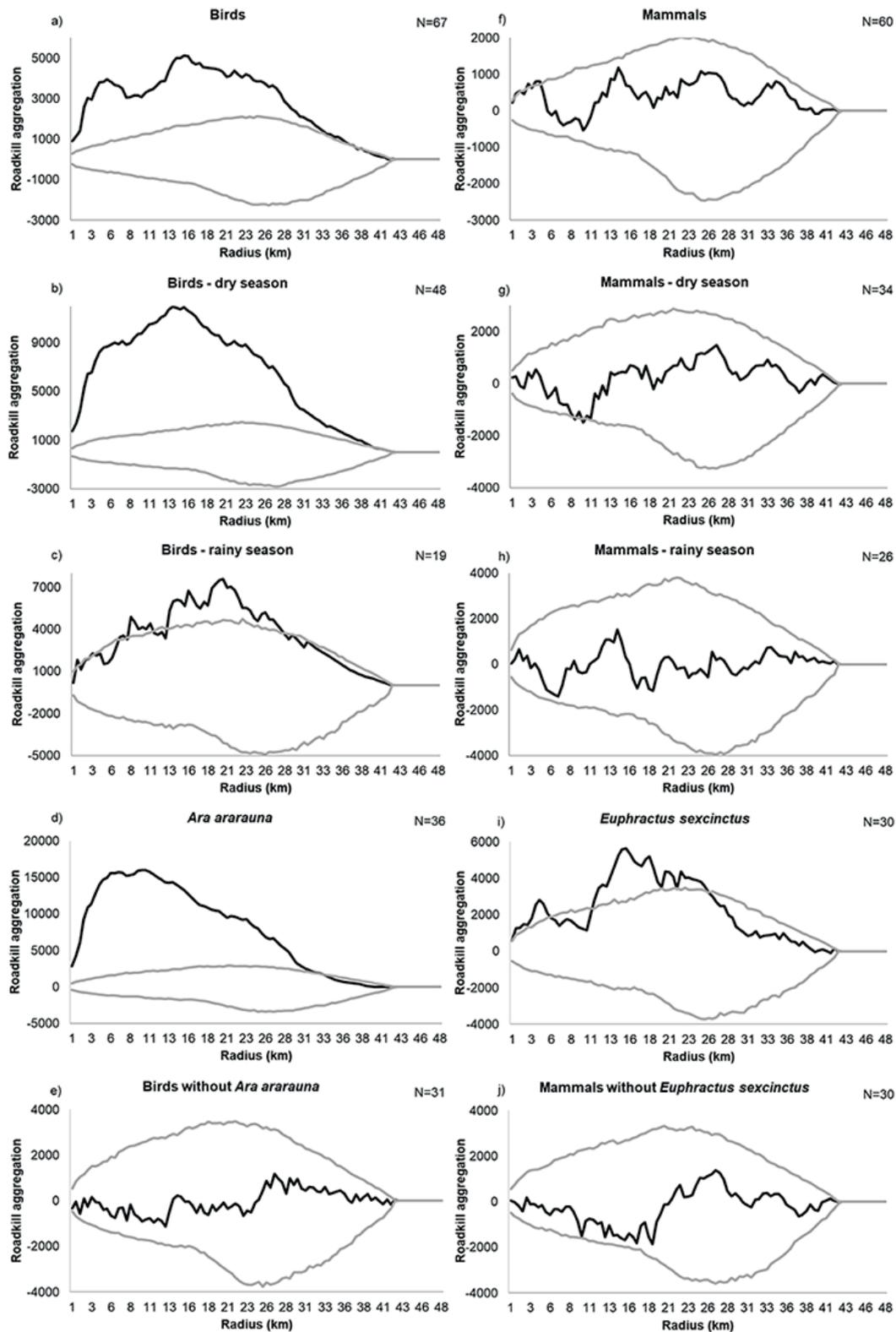


Figure 2. Roadkill aggregation (black line) as a function of scale distance (radius) and confidence limits of 95 % (light-gray lines) for the roadkill distribution on GO-341 highway, from km 65 to 112, Goiás state, Brazil (2017-2018): a) birds - dry and rainy seasons; b) birds - dry season; c) birds - rainy season; d) *Ara ararauna*; e) birds without *Ara ararauna*; f) mammals - dry and rainy seasons; g) mammals - dry season; h) mammals - rainy season; i) *Euphractus sexcinctus*; j) mammals excluding *Euphractus sexcinctus*. Values of roadkill aggregation above the confidence limits indicate significant aggregation and values below these thresholds indicate significant dispersion. Analysis carried out in Siriema software v. 2.0 (Coelho *et al.* 2014).

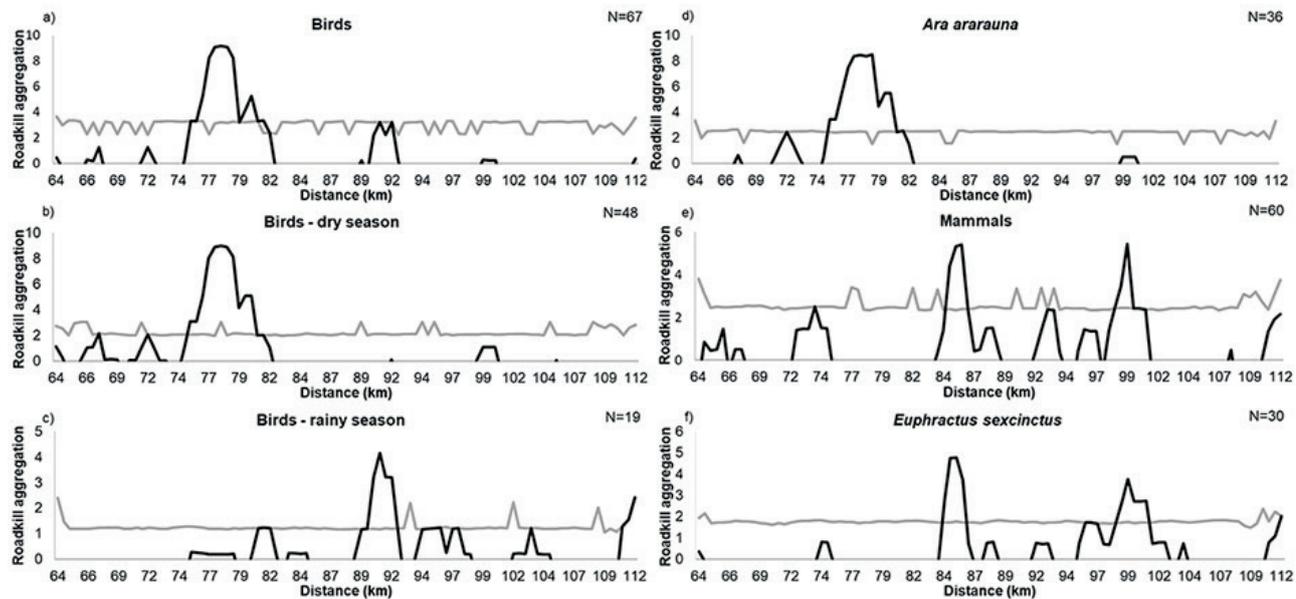


Figure 3. Wild animals' hotspot location on GO-341 highway, from km 65 to 112, Goiás state, Brazil (2017-2018). Black line – aggregation values; gray line - upper confidence limits; roadkill aggregation values above confidence limits indicate stretches with significant roadkill aggregation intensity a) birds - dry and rainy seasons; b) birds - dry season; c) birds, rainy season; d) *Ara ararauna*; e) mammals - dry and rainy seasons; f) *Euphractus sexcinctus*. Analysis carried out in Siriema software v. 2.0 (Coelho *et al.* 2014).

remain on the road during a week, which would result in higher RK observed for weekly surveys when compared to daily ones.

Among the 33 medium to large-sized mammal species listed for the ENP and region (Giozza *et al.* 2017), 12 are considered as threatened in the Brazilian Red List (Instituto Chico Mendes de Conservação da Biodiversidade - ICMBio 2018) and two of them have been found killed in this survey. The hoary fox is listed as endangered in Brazil in the “vulnerable” category (Instituto Chico Mendes de Conservação da Biodiversidade - ICMBio 2016). Giozza *et al.* (2017) observed that the abundance of this species decreased in ENP in a ten-year interval, thus, even the loss of a few individuals due to roadkill can be a great threat to the population survival. The greater rhea is another species in risk of extinction, classified as near threatened (IUCN 2020), in spite of the park had received its name because of the great abundance of this species in the area (IBAMA & CEBRAC 2004). Besides, species such as the greater grison, *Galictis vitata*, (Schreber, 1776), the pampas deer, *Ozotoceros bezoarticus* (Linnaeus, 1758) and the Brazilian tapir, *Tapirus terrestris* (Linnaeus, 1758) were visualized crossing or feeding on the plantations adjacent to the highway. It is worth mentioning that some other species registered in

the study region are victims of roadkill. In previous or subsequent periods to the roadkill monitoring, we found carcasses of Brazilian tapir, marsh deer, *Blastocerus dichotomus* (Illiger, 1815) and ocelot, *Leopardus pardalis* (Linnaeus, 1758). Neto *et al.* (2015) identified marsh deer, maned wolf, *Chrysocyon brachyurus* (Illiger, 1815) and giant anteater, *Myrmecophaga tridactyla* (Linnaeus, 1758), roadkilled on roads near the ENP. Therefore, the absence of a certain species on the roadkill monitoring does not mean that this species is not a victim of roadkill, and more extensive monitoring is necessary to make such assumption.

In the South-western region of Brazil the crab-eating fox, the six-banded armadillo, the giant anteater, the southern tamandua and the nine-banded armadillo, *Dasybus novemcinctus* (Linnaeus, 1758), are being roadkilled more frequently than expected based on their abundance, according to Caceres (2011). This may indicate that, in our study area, despite being a common species, the six-banded armadillo and the crab-eating fox need to receive special attention as they can be in risk of local extinction due to roadkill, demanding future studies.

The blue-and-yellow macaws were the animals that presented the highest RK in the study. These birds are large and slow to fly (Sick 1997), they

have difficulties to escape from the road quickly and end up being hit by high-speed vehicles. On some occasions, more than one individual was killed at once. Macaws were killed only during the dry season. This fact is possibly related to lower food supply (fruits) in natural environments at this time of the year (Batalha & Martins 2004) and to the greater movement of the macaws in search for food (Gwynne *et al.* 2010). Sexual behavior may also contribute to higher RK during the dry season as the nesting period starts in the end of this season (from August to December) (Bianchi 1998). During this period, blue-and-yellow macaws may be attracted to the road due to the presence of grains and pieces of sugarcane that fall from the trucks during the transportation from the agricultural area to the companies responsible for the processing of the raw material. Still, it is precisely during the dry season that the sugarcane harvest occurs and, consequently, the road may have a greater vehicles flow. The greater intensity of vehicular traffic during the day (Carvalho-Roel *et al.* 2019) increases the likelihood of roadkill of animals that have diurnal habits, for instance, we observed that diurnal birds had higher RK than nocturnal ones. Brum *et al.* (2017) also found diurnal animals to be the most affected by roadkill and explained this because of the greater vehicles flow in the morning and a lower one in the early hours. The same observation about vehicles flow was made for the study area (Carvalho-Roel *et al.* 2019). Vehicles flow is an important variable that influences roadkill. The number of roadkilled animals usually increases with increasing vehicle flow (Coelho *et al.* 2008, Gunson *et al.* 2011). This is probably the reason why we found a higher number of crab-eating fox killed during the sugarcane harvest period. Another hypothesis is that during harvest, the animals that use these areas are pushed onto the highway in search for shelter and food.

Although other authors have also found roadkill aggregations for mammals (Coelho *et al.* 2008, Cáceres *et al.* 2012, Miranda *et al.* 2020) and birds (Coelho *et al.* 2008), this is the first research article to suggest that roadkill hotspots are located in different places regarding the season (for birds). This result reinforces the necessity to survey both seasons. Roadkill hotspots for the whole year encompassed just those ones of the

dry season. Nonetheless, these results must be interpreted with caution. Firstly, because the sampling effort was different for each season, as the dry season was monitored twice the rainy one. Secondly, because increasing the sampling effort in the rainy season, the roadkill hotspots of the whole data set would probably encompass the hotspots of this season. Therefore, it is necessary to understand how large the sampling effort must be so that roadkill hotspots can be correctly identified. We recommend that other research should also compare roadkill hotspots between seasons. Another important factor to highlight is that bird and mammal roadkill hotspots may reflect the most abundant species hotspots and some innovative approaches need to be implemented to avoid this bias. In addition, without considering the most abundant species, birds and mammals roadkill were not aggregated in our study.

Measures aiming to decrease the number of roadkills in the ENP need to be taken. Generally, measures to be taken in the short term are: install speed reducers in the stretches of roadkill hotspots (km 76 to 81, 85 to 86, 90 to 92 and 99) to force the vehicles to reduce their speed, regardless of their category; install speed control and surveillance equipment on cargo vehicles; and adapt the bridges and culverts so that they can work as animal passages with fences that direct the animals to these places and prevent them from crossing the highway (Glista *et al.* 2009, Grilo *et al.* 2010). Although less expensive, the first two measures need more studies to prove its efficiency. Long-term measures are the installation of passages along with fences in the stretches pointed as critical (Rytwinski *et al.* 2016). Though typically conceived for mammals, reptiles and amphibians, birds also benefit from wildlife passages. The construction of structural elements can encourage birds to fly above traffic or below the road through bridges or culverts (Kociolek *et al.* 2015).

The ENP is of crucial importance in the conservation of Cerrado species (IBAMA & CEBRAC 2004) and this research was a first step in order to understand how roadkill could be affecting this protected area. Even performing consecutive surveys, we were not able to identify small species as amphibians, therefore, we advise that some

stretches of the highway should be monitored on foot. We also suggest the implementation of carcass removal experiments in order to calculate a correction rate and identify these small species. A correction rate would enable to estimate how many individuals die on GO-341 and calculate how roadkill is affecting the ENP wildlife populations. Finally, it is necessary to continue the roadkill monitoring of the highway and also implement a monitoring of mitigating measures in order to verify their effectiveness (Lesbarrères & Fahrig 2012). The fauna roadkill is a problematic issue for species conservation and for the ENP, as it is one of the few refuges of the Cerrado fauna, needing to be urgently protected from this threat.

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REFERENCES

- Batalha, M. A., & Martins, F. R. 2004. Reproductive phenology of the cerrado plant community in Emas National Park (Central Brazil). *Australian Journal of Botany*, 52(2), 149–161. DOI: 10.1071/BT03098
- Bérnils, R. S., & Costa, H. C. 2015. Répteis brasileiros: lista de espécies. *Herpetologia Brasileira*, 4(3), 75–93.
- Bianchi, C. A. C. 1998. Biologia reprodutiva da arara canindé (*Ara ararauna*, Psittacidae) no Parque Nacional das Emas, GO. Universidade de Brasília. p. 75.
- Brasil. Lei nº 12.727 de 17 de outubro de 2012.
- Brum, T. R., Santos-Filho, M., Canale, G. R., Ignácio, A. R. A., Brum, T. R., Santos-Filho, M., Canale, G. R., & Ignácio, A. R. A. 2018. Effects of roads on the vertebrates diversity of the Indigenous Territory Paresi and its surrounding. *Brazilian Journal of Biology*, 78(1), 125–132. DOI: 10.1590/1519-6984.08116
- Bueno, C., & Almeida, P. J. A. L. 2010. Sazonalidade de atropelamentos e os padrões de movimentos em mamíferos na BR-040 (Rio de Janeiro-Juiz de Fora). *Revista Brasileira de Zoociências*, 12(3), 219–226.
- Caceres, N. C. 2011. Biological characteristics influence mammal road kill in an Atlantic Forest–Cerrado interface in south-western Brazil. *Italian Journal of Zoology*, 78(3), 379–389. DOI: 10.1080/11250003.2011.566226
- Cáceres, N. C., Casella, J., & Goulart, C. S. 2012. Variação espacial e sazonal de atropelamentos e mamíferos no bioma cerrado, rodovia BR 262, Sudoeste do Brasil. *Mastozoología Neotropical*, 19(1), 21–33.
- Carvalho-Roel, C. F., Alves, G. B., Jácomo, A. T. D. A., Moreira, R. A., Tôrres, N. M., & Silveira, L. 2019. Vehicles flow on GO-341 highway, tangent to the Emas National Park. *Figshare*, 1–3. DOI: 10.6084/m9.figshare.8984291.v1
- Carvalho, C. F., Iannini Custodio, A. E., & Marçal Junior, O. 2015. Wild vertebrates roadkill aggregations on the BR-050 highway, state of Minas Gerais, Brazil. *Bioscience Journal*, 31(3), 951–959. DOI: 10.14393/BJ-v31n3a2015-27468
- Coelho, A. V. P., Coelho, I. P., Teixeira, F. Z., & Kindel, A. 2014. Siriema: road mortality software. V. 2.0. Version 2.0. Porto Alegre, Brasil: NERF, UFRGS.
- Coelho, I. P., Kindel, A., & Coelho, A. V. P. 2008. Roadkills of vertebrate species on two highways through the Atlantic Forest Biosphere Reserve, southern Brazil. *European Journal of Wildlife Research*, 54(4), 689–699. DOI: 10.1007/s10344-008-0197-4
- Cunha, H. F. Da, Moreira, F. G. A., & Silva, S. D. S. 2010. Roadkill of wild vertebrates along the GO-060 road between Goiânia and Iporá, Goiás State, Brazil. *Acta Scientiarum. Biological Sciences*, 32(3), 257–263. DOI: 10.4025/actascibiolsci.v32i3.4752
- D'Amico, M., Román, J., de los Reyes, L., & Revilla, E. 2015. Vertebrate road-kill patterns in Mediterranean habitats: Who, when and where. *Biological Conservation*, 191(November), 234–242. DOI: 10.1016/j.biocon.2015.06.010
- Garriga, N., Franch, M., Santos, X., Montori, A., & Llorente, G. A. 2017. Seasonal variation in vertebrate traffic casualties and its implications for mitigation measures. *Landscape and*

- Urban Planning, 157, 36–44. DOI: 10.1016/j.landurbplan.2016.05.029
- Giozza, T. P., Jácomo, A. T. D. A., Silveira, L., & Tôrres, N. M. 2017. Riqueza e abundância relativa de mamíferos de médio e grande porte na região do Parque Nacional das Emas-GO. *Revista Brasileira de Zoociências*, 18(3), 71–87.
- Glista, D. J., DeVault, T. L., & DeWoody, J. A. 2009. A review of mitigation measures for reducing wildlife mortality on roadways. *Landscape and Urban Planning*, 91(1), 1–7. DOI: 10.1016/j.landurbplan.2008.11.001
- Grilo, C., Bissonette, J. A., & Cramer, P. C. 2010. Mitigation measures to reduce impacts on biodiversity. In: S. R. Jones (Ed.), *Highways: Construction, Management, and Maintenance*. pp. 73–114. Nova Science Publishers, Inc.
- Gunson, K. E., Mountrakis, G., & Quackenbush, L. J. 2011. Spatial wildlife-vehicle collision models: a review of current work and its application to transportation mitigation projects. *Journal of Environmental Management*, 92(4), 1074–82. DOI: 10.1016/j.jenvman.2010.11.027
- Gwynne, J. a., Ridgely, R. S., Argel, M., & Tudor, G. 2010. *Aves do Brasil. Pantanal & Cerrado*. São Paulo: Editora Horizonte: p. 322.
- IBAMA & CEBRAC. 2004. *Plano de Manejo - Parque Nacional das Emas*. IBAMA: p. 893.
- Instituto Chico Mendes de Conservação da Biodiversidade - ICMBio. 2018. *Brazil red book of threatened species of fauna*. Brasília, Brazil: ICMBio: p. 4162.
- IUCN. 2020. *The IUCN Red List of Threatened Species*. (Retrieved on from www.iucnredlist.org/).
- Jacobson, S. L., Bliss-ketchum, L. L., Rivera, C. E. De, & Smith, W. P. 2016. A behavior-based framework for assessing barrier effects to wildlife from vehicle traffic volume. *Ecosphere*, 7(4), 1–15.
- Kociolek, A., Grilo, C., & Jacobson, S. 2015. Flight doesn't solve everything: mitigation of road impacts on birds. In: R. van der Ree, D. J. Smith, & C. Grilo (Eds.), *Handbook of Road Ecology*, First Edit, pp. 281–289. First Edit ed. John Wiley & Sons.
- Laurance, W. F., Goosem, M., & Laurance, S. G. W. 2009. Impacts of roads and linear clearings on tropical forests. *Trends in Ecology & Evolution*, 24(12), 659–69. DOI: 10.1016/j.tree.2009.06.009
- Lesbarrères, D., & Fahrig, L. 2012. Measures to reduce population fragmentation by roads: what has worked and how do we know? *Trends in Ecology & Evolution*, 27(7), 374–80. DOI: 10.1016/j.tree.2012.01.015
- Lima, K. C. B. 2013. *Impacto de estradas em unidades de conservação do Brasil*. Federal University of Lavras. p. 94.
- Magioli, M., Maria, K., Micchi, P., Ferraz, D. B., Zulnara, E., Setz, F., Percequillo, A. R., Viviane, M., Santos, D. S., Kuhnen, V. V., Cristina, M., Evelyn, K., Kanda, C. Z., Fregonezi, G. D. L., & Alves, H. 2016. Connectivity maintain mammal assemblages functional diversity within agricultural and fragmented landscapes. *European Journal of Wildlife Research*, 62(4), 431–446. DOI: 10.1007/s10344-016-1017-x
- Miranda, H. 2017. Transporte fluvial de cargas e passageiros cresce no Amazonas. (Retrieved on November 7th, 2018, from <https://www.marinha.mil.br/content/transporte-fluvial-de-cargas-e-passageiros-cresce-no-amazonas>).
- Miranda, J. E. S., Melo, F. R. de, & Umetsu, R. K. 2020. Are Roadkill Hotspots in the Cerrado Equal Among Groups of Vertebrates? *Environmental Management*, 65(4), 565–573. DOI: 10.1007/s00267-020-01263-y
- Myers, N., Fonseca, G. A. B., Mittermeier, R. a, Fonseca, G. A. B., & Kent, J. 2000. Biodiversity hotspots for conservation priorities. *Nature*, 403(6772), 853–858. DOI: 10.1038/35002501
- Neto, C. de M. S., Carneiro, V. A., Gonçalves, B. B., & Ribeiro, F. J. da C. 2015. *Fauna Atropelada Nas Estradas Do Município De Chapadão Do Céu (Goiás, Brasil)*. *Revista Percurso -NEMO Maringá*, 7(1), 97–114. DOI: 10.4025/percurso.v7i1.24915
- Paglia, A. P., Fonseca, G. A. B. da, Rylands, A. B., Herrmann, G., Aguiar, L. M. S., Chiarello, A. G., Leite, Y. L. R., Costa, L. P., Siciliano, S., Kierulff, M. C. M., Mendes, S. L., Mittermeier, R. A., & Patton, J. L. 2012. Annotated checklist of Brazilian mammals 2° Edição. *Occasional Papers in Conservation Biology*. Vol. 6p. 1–76.
- Piacentini, V. de Q., Aleixo, A., Agne, C. E., Maurício, G. N., Pacheco, J. E., Bravo, G. A., Brito, G. R. R., Naka, L. N., Olmos, F., Posso, S., Silveira, L. F., Betini, G. S., Carraro, E., Franz, I., Less, A. C., Lima, L. M., Schunk, F., Amaral, F.

- R., Bencke, G. A., Cohn-Haft, M., Figueiredo, L. F. A., Straube, F. C., & Cesari, E. 2015. Annotated checklist of the birds of Brazil by the Brazilian Ornithological Records Committee / Lista comentada das aves do Brasil pelo Comitê Brasileiro de Registros Ornitológicos. *Revista Brasileira de Ornitologia*, 23(2), 91–298.
- R Core Team. 2019. R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing.
- Reis, N. R., Peracchi, A. L., Fragonezi, M. N., & Rossaneis, B. K. 2010. Mamíferos do Brasil - Guia de Identificação. First Edition. Technical Books: p. 557.
- Rodrigues, F. H. G., Silveira, L., Jácomo, A. T. A., Carmignotto, A. P., Bezerra, A. M. R., Coelho, D. C., Garbogini, H., Pagnozzi, J., & Hass, A. 2002. Composição e caracterização da fauna de mamíferos do Parque Nacional das Emas, Goiás, Brasil. *Revista Brasileira de Zoologia*, 19(2), 589–600. DOI: 10.1590/S0101-81752002000200015
- Rytwinski, T., & Fahrig, L. 2015. The Impacts of Roads and Traffic on Terrestrial Animal Populations. In: R. van der Ree, D. J. Smith, & C. Grilo (Eds.), *Handbook of Road Ecology*. pp. 237–246. John Wiley & Sons, Ltd.
- Rytwinski, T., Soanes, K., Jaeger, J. A. G., Fahrig, L., Findlay, C. S., Houlahan, J., Van Ree, R. Der, & Van Der Grift, E. A. 2016. How effective is road mitigation at reducing road-kill? A meta-analysis. *PLoS ONE*, 1–25. DOI: 10.1371/journal.pone.0166941
- Santos, R. A. L., Santos, S. M., Santos-Reis, M., Figueiredo, A. P., de Bager, A., Aguiar, L. M. S., & Ascensão, F. 2016. Carcass Persistence and Detectability: Reducing the Uncertainty Surrounding Wildlife-Vehicle Collision Surveys. *PloS One*, 11(11), 1–15. DOI: 10.1371/journal.pone.0165608
- Santos, R. A. L., Ascensão, F., Ribeiro, M. L., Bager, A., Santos-Reis, M., & Aguiar, L. M. S. 2017. Assessing the consistency of hotspot and hot-moment patterns of wildlife road mortality over time. *Perspectives in Ecology and Conservation*, 15(1), 56–60. DOI: 10.1016/j.pecon.2017.03.003
- Sawyer, D., Mesquita, B., Coutinho, B., Almeida, F. V. de, Figueiredo, I., Lamas, I., Pereira, L. E., Pinto, L. P., & Navega, R. 2016. Perfil do Ecossistema Hotspot de Biodiversidade do Cerrado. *Critical Ecosystem Partnership Found*: p. 495.
- Sick, H. 1997. *Ornitologia brasileira*. Rio de Janeiro: Nova Fronteira: p. 912p.
- Sigrist, T. 2014. *Guia de Campo: Avifauna Brasileira*. 4a ed. 4a ed. ed. São Paulo: Avis Brasilis: p. 600.
- Silva, J. M. C. 1995. Birds of the Cerrado Region, South America. *Steenstrupia*, 21(March), 69–92.
- Strassburg, B. B. N., Brooks, T., Feltran-Barbieri, R., Iribarrem, A., Crouzeilles, R., Loyola, R., Latawiec, A. E., Oliveira Filho, F. J. B., Scaramuzza, C. A. de M., Scarano, F. R., Soares-Filho, B., & Balmford, A. 2017. Moment of truth for the Cerrado hotspot. *Nature Ecology & Evolution*, 1(4), 1–3. DOI: 10.1038/s41559-017-0099
- Teixeira, F. Z., Coelho, I. P., Esperandio, I. B., Oliveira, N. R., Porto, F., Dornelles, S. S., Delazeri, N. R., Tavares, M., Martins, M. B., & Kindel, A. 2013. Are road-kill hotspots coincident among different vertebrate groups? *Oecologia Australis*, 17(1), 36–47. DOI: 10.4257/oeco.2013.1701.04
- Teixeira, F. Z., Coelho, A. V. P., Esperandio, I. B., & Kindel, A. 2013. Vertebrate road mortality estimates: Effects of sampling methods and carcass removal. *Biological Conservation*, 157, 317–323. DOI: 10.1016/j.biocon.2012.09.006
- van der Ree, R., Smith, D. J., & Grilo, C. 2015. The Ecological Effects of Linear Infrastructure and Traffic: Challenges and Opportunities of Rapid Global Growth. In: R. van der Ree, D. J. Smith, & C. Grilo (Eds.), *Handbook of Road Ecology*. pp. 1–9. John Wiley & Sons, Ltd.
- Veloso, A. C. 2015. Atropelamentos de vertebrados no trecho Uberlândia-Uberaba da rodovia BR 050, Minas Gerais. Universidade Federal de Uberlândia, Uberlândia, Brasil. p. 38.

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