1	Preventing wildlife roadkill can offset mitigation investments in
2	short-medium term
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Preventing wildlife roadkill can offset mitigation investments in
 short-medium term

34

35 Abstract

36 Wildlife vehicle collisions (WVC) are a threat to wildlife and humans, killing millions of 37 animals of numerous species, as well as causing significant damage to vehicles, drivers and 38 passengers. Road fencing is a highly effective mitigation measure at reducing WVC, however 39 its large-scale implementation requires a high investment. We questioned how long it would 40 take for savings from avoided collisions to offset the investments in road fencing mitigation, 41 focusing on vehicle damage costs. Using the information of a 3-year systematic roadkill 42 monitoring of 1,158 km in Mato Grosso do Sul, Brazil, we estimated the real number of 43 casualties accounting for bias in roadkill counting. We obtained information on the material 44 costs on cars and trucks due to WVC and, considering the road traffic volume characteristics, 45 estimated the total material costs resulting from collisions with larger animals. Cost-benefit 46 analyzes allowed estimating the time required to amortize the investment in fencing, 47 considering its application along the full surveyed roads or only in hotspots of mortality. We 48 recorded over 10,000 WVC, 40% of which involved animals that can cause significant 49 material damage to vehicles, namely the endangered lowland tapir (*Tapirus terrestris*, n=267) 50 and giant anteater (Myrmecophaga tridactyla, n=608). The average material cost per accident 51 was US\$ 885±1,575 (mean±SD). We show that investments are likely to pay off in 16-40 52 years for the mitigation of the full roads, and in 9-25 years for hotspots of mortality. Thus, road mitigation is a win-win solution for increasing traffic safety for humans and reduce 53 54 road-related negative effects on biodiversity.

- 55 Keywords: Road fencing, road-mortality, endangered species, material damages, carcass
- 56 persistence, cost-benefit analysis.

57 1 Introduction

58 Roads are the most common human-made features on the globe, spreading across nearly all 59 of its surface (Ibisch et al. 2016). An additional 25 million km of new roads are expected to 60 be built by 2050, 90% of which in developing countries (Dulac 2013), particularly in tropical 61 regions, which have support exceptional biodiversity and vital ecosystem services (Laurance 62 et al. 2001, 2015, Alamgir et al. 2017). These infrastructures allow human population 63 development by facilitating the movement of people and goods, as well their accessibility to 64 different services, e.g. schools and markets (Ali et al. 2015). However, particularly outside 65 urban areas, roads can also become areas of conflict with biodiversity, notably due to the high number of wildlife-vehicle collisions (WVC). For example, over two million mammals are 66 67 estimated to be road-killed every year solely in Brazil (González-Suárez et al. 2018).

68 Collisions with wild animals, mainly large ones, can also be a serious threat to human 69 safety, causing significant injuries to drivers and passengers, in addition to considerable 70 economic impact (Bissonette et al. 2008, Huijser et al. 2009, 2013, Abra et al. 2019). In USA, 71 it is estimated that 1-2 million WVC occur involving large mammals, of which ca. 5% cause 72 human injury with costs surpassing US\$ 8bn (Huijser et al. 2008). In California, in particular, 73 it was estimated a material damage cost of US\$ 76 million due to large wildlife-vehicle 74 collision, for 2018 alone (Shilling et al. 2019). The annual cost of WVC involving ungulates 75 in Sweden was estimated at US\$ 406 million (Gren and Jägerbrand 2019), and in the São Paulo State, Brazil, the total annual cost to society was estimated at US\$ 25 million (Abra et 76 77 al. 2019). Yet, there is a great lack of knowledge about the material costs that WVC entail in 78 most regions, even though this information is particularly relevant for making decisions about 79 the implementation of mitigation measures on roads.

Roadkill data is usually acquired through systematic monitoring of roads. However,
roadkill surveys generally fail to fully record all casualties due to imperfect detection issues,

82 and yet accurate estimation of the total mortality is important for precise estimates of the 83 material costs associated with WVC. Imperfect detection may be due to one or more 84 detection biases, including: i) researchers failing to find carcasses along the road or verges 85 while surveying the road; *ii*) traffic volume, scavengers or management staff removing carcasses before searches are conducted; and iii) carcasses ending up outside the searched 86 87 area (Santos et al. 2011, 2016, Teixeira et al. 2013). To account for such biases, researchers 88 typically conduct field trials to estimate the effects of the first two components (Barrientos et 89 al. 2018). For the third component, while generally less considered in mortality estimates, 90 one can perform extra surveys outside the road corridor to count the number of carcasses 91 therein or search for signs of injured animals.

92 Once a detailed knowledge of roadkill patterns is obtained, different mitigation 93 measures can be considered to reduce WVC. Fencing is one of most common mitigation 94 measures, and one with higher effectiveness at reducing road mortality, particularly when 95 connecting to existing road passages (Seiler et al. 2016, Rytwinski et al. 2016, Spanowicz et 96 al. 2020). Other measures, such as raising the road on pillars or placing the road underground 97 are potentially more effective than fencing but are also cost prohibitive and unrealistic for 98 most regions. Conversely, less expensive measures than fencing, such as wildlife warning 99 signs and reflectors, are apparently ineffective (Rytwinski et al. 2016, Brieger et al. 2017, 100 Benten et al. 2018). Hence, while also requiring regular maintenance, wildlife fencing seems 101 to be an important cost-effective strategy for road mortality mitigation of most species (Seiler 102 et al. 2016, Rytwinski et al. 2016, Spanowicz et al. 2020).

Due to financial constraints, it is rarely realistic to fence an entire road network simultaneously, and thus mitigating particular road sections may be more cost-effective than fencing all roads (Ascensão et al. 2013, Spanowicz et al. 2020). One common approach is to focus on road sections with significant aggregation of collisions (Spanowicz et al. 2020).

Given that collisions with large body mass species represent a higher threat for humans, with
corresponding higher material costs, fencing may be more beneficial in sections where these
species tend to be more road-killed, as where roads cross landscape connectivity corridors
(Grilo et al. 2011, Santos et al. 2017), in opposition to fencing where higher overall roadkill
occurs.

112 Knowledge of the patterns of mortality, namely how it varies along time, its spatial 113 aggregation, and their costs allow for better management of existing roads as well as better 114 planning for future road networks. This information is even more necessary in developing 115 regions where road networks are expected to have greater expansion and impact in near 116 future (Meijer et al. 2018, Ascensão et al. 2018, Ascensão 2020). In this study, our main goal 117 was to estimate the material costs due to WVC in a region of fast development as is the state 118 of Mato Grosso do Sul, Brazil. The reduction of roadkill is essential therein due to the large 119 number of WVC that has been evidenced in several studies, but also due to the threat to 120 human safety that collisions with species of large body mass represent, including the lowland 121 tapir (*Tapirus terrestris*), giant anteater (*Myrmecophaga trydactyla*) or capybara 122 (Hydrochoerus hydrochaeris) (Ascensão et al. 2017, 2019).

123 We used the information from a 3-year systematic roadkill monitoring program over 124 of 1,158 km, for which we estimated the real number of casualties accounting for bias in 125 roadkill counting. We obtained information on the material costs on vehicles (cars and trucks) 126 due to WVC from car mechanics and online surveys and estimated the total material costs 127 resulting from collisions with larger animals considering the traffic volume characteristics 128 (proportion of cars and trucks). We then performed a cost-benefit analysis comparing the 129 costs of material damage with the necessary investment to implement fences (e.g. Huijser et 130 al. 2009), either along the full road corridor or only in sections with significant roadkill 131 aggregation (i.e. hotspots of mortality). With this information we estimated how long it

would take to obtain a return on the investment in fencing that prevent collisions, consideringthe avoided costs due to WVCs.

134

135 2 Methods

136 **2.1** Study area

The study was conducted in the state of Mato Grosso do Sul (MS), located in center-west
region of Brazil, throughout six different transects on four roads (Fig.1), namely the federal
roads BR-262 (R1A, R1B and R4) and BR-267 (R3B), and the state roads MS-040 (R2) and
MS-338 (R3A), totaling 1,158 km (Fig. 1, Table 1). All roads have two-lanes and are paved.
[FIG. 1]

145 The land use bordering these roads was dominated by grassland/pasture and agriculture, 146 followed by remnant native vegetation and riparian areas. The westernmost transect (R4) runs 147 through part of the Pantanal biome (Brazilian wetland, see Fig. 1) and has higher cover of 148 native vegetation in its surroundings when compared to the other transects that cross the 149 Cerrado biome (Brazilian savannah). Urban areas had relatively low representation in the 150 study area and were not surveyed in road transects. Traffic volume was similar across 151 transects in the two federal roads (BR) surveyed, ca. 3,600 vehicles per day, of which 33-152 48% were trucks (DNIT 2020; Table 1). In roads R2 and R3A, there were no official traffic counts, so we recreated the methodology used by governmental authorities (DNIT 2020), 153 154 namely vehicle counting throughout a full week (24 h per day) (Table 1). The climate 155 throughout MS is wet from October to March and dry from April to September (Koppen's As

156	or Aw), with mild year-round temperatures (range 21-32°C). Average annual rainfall ranges
157	between 1000-1500 mm.
158	

[TABLE 1]

160

161 2.2 Roadkill systematic surveys

162 Roadkill surveys were carried out by car (40-60 km/h), searching for road-killed animals on 163 both lanes and shoulders. The choice to carry out research by car and at this speed was due to 164 the aim to cover a large territory and to record wildlife carcasses with body mass >1 kg. 165 Monitoring lasted three years on a fortnightly basis (average interval of 14.5 days) (Table 1). 166 The monitoring on R1B was interrupted after one year of survey for safety reasons, given the 167 lack of safe places to park the car when an animal was detected. A total of 420 survey events 168 were performed, totaling 84,673 km of survey effort. Each observation was classified to 169 species level (when possible) and its location recorded with hand-held GPS before carcasses 170 were removed from the road to avoid recounting or road accidents.

171

172 **2.3** Carcass persistence time

In parallel with road surveys, we carried out experiments to assess the effect of carcass removal, in order to more accurately estimate the real number of WVC. Experiments were performed both in wet and dry seasons, on 337 km of three of the surveyed transects, one with low traffic volume (R2) and two with higher traffic volume (R3B and R4). These sections were surveyed on a daily basis for 15-30 days (mean 25.9 days) (Table 2). All animals found on the first day were discarded and in the following days all road-killed carcasses were left intact, where detected, recording the species identification (or lower taxonomic level), its approximate weight and their condition (detectable or not). In total, 423
carcasses with body mass >1kg were followed, 268 with less than 5 kg (hereafter <5 kg), 124
between 5-30 kg (hereafter <30 kg) and 31 with more than 30 kg (hereafter >30 kg); 207 in
wet season and 216 in dry season (Table 2).

- 184
- 185

[TABLE 2]

186 **2.4 Material costs of collisions**

187 We obtained wildlife-vehicle collisions data from Mato Grosso do Sul State containing 188 information about the accident, including the species involved, vehicle type (car, truck, 189 other), and damage to the vehicle. Photos of the accident taken by the road company staff 190 allowed to confirm the species involved in WVC. An estimate of the cost to repair each 191 vehicle was then requested to three different vehicle repair shops in Campo Grande and then 192 averaged. In complement, we created an online questionnaire with the Google forms platform 193 (Supplementary material S1) and disseminated in social networks. There, we asked about the 194 date, place and period of the day (day, night, sunset, sunrise) of the collision events; the type 195 of vehicle (car, truck, other); name or description of the species involved in collisions; and 196 the cost of the material damage, if any. A section for adding observations was also included 197 in which citizens could provide details about the accident (e.g. whether the vehicle has been 198 irreparably damaged).

199

200 2.5 Data analysis

201 **2.5.1** Species potentially causing vehicle damage

All species detected in collisions (systematic surveys) were first characterized according to
 their likelihood of causing material damage to vehicles when involved in collisions. Based on

204 information from material damage (interviews and questionnaires, see Results), we classified 205 as potentially causing damage those species with body mass >1kg, excluding all reptiles such 206 as caimans, and short stature mammals as armadillos. Flying birds were also not considered 207 as fencing (the mitigation method considered here) is unlikely to reduce collisions with these 208 species. Overall, we considered mammals with >1kg and the non-flying bird greater rhea 209 (Rhea americana). Those species likely to cause material damage were further classified in 210 three body mass classes (kg): <5, <30 and >30 (see Table S2.1 in Supplementary material S2 211 for full classification).

212

213 **2.5.2 Estimating total number of roadkill**

214 We estimated the real number of roadkills that occurred during the survey period, for each 215 road transect, using the GenEst framework and corresponding R package (Simonis et al. 216 2018, Dalthorp et al. 2020). This software integrates data collected during carcass searches 217 together with estimates of carcass persistence, search area and searcher efficiency, to 218 accurately estimate the real number of collisions and to provide a measure of precision 219 associated with the estimate. GenEst was designed to address the general problem of 220 estimating the size of a population when not all animals are present on all survey occasions 221 and the probability of detection is less than one (Simonis et al. 2018). Following Dalthorp 222 (2018), GenEst is an elaboration of a binomial probability model $X \sim \text{binomial } (M, g)$, where 223 X is the observed number of carcasses and g is the detection probability. If g is known, then 224 M' = X/g is an unbiased estimator for M and the sampling variance of X is the only source of uncertainty about M. Splitting the total mortality into groups, e.g. A and B, then $M' = M'_A + M'_A$ 225 $M'_B = X_A / g_A + X_B / g_B$ is unbiased for *M* (Dalthorp et al. 2018). GenEst makes extensive use 226 227 of this simple idea of splitting the carcass observation data into distinct subgroups, estimating 228 mortality in each subgroup, and combining subgroups into larger groups by summing. Further details are extensively documented in GenEst supporting documents (Dalthorp et al. 2018,
Simonis et al. 2018). We used GenEst to model carcass persistence time and, together with
estimates on searcher efficiency and the area searched along the road, to estimate the total
mortality for the different body mass groups per road transect.

233 Carcass persistence was modelled using the data from our experiments, to estimate 234 the amount of time a carcass would persist for, given the conditions under which it arrived. 235 For each body mass class, four classes of parametric models of carcass persistence were 236 tested: exponential, Weibull, logistic, and lognormal. In each model we further tested the 237 effect of traffic volume (high/low) and season (wet/dry) on the persistence probability, by 238 comparing models including all combinations of these two predictors together with the null 239 model, according to the Akaike Information Criterion (AIC). Models were selected based on 240 AIC values, considering that $\Delta AIC \le 2$ indicate similar models (Burnham and Anderson 241 2004).

As for searcher efficiency (SE), estimating this component requires a different set of 242 243 experiments, where the carcasses are disposed by other people in known locations, and 244 roadkill surveyors have to detect them. The detection error rate provides an estimate of the 245 detectability (Santos et al. 2016). Due to logistic constrains we did not carry out these experiments. However, researchers were highly experienced in roadkill surveys in our study 246 247 area (Ascensão et al. 2017), and based on this experience we estimated that we could detect at 248 least 85% of carcasses with >1kg on the road. Given the uncertainty of the real value, we 249 used different values of SE for estimating total mortality: 0.85, 0.90, and 0.95. For carcasses 250 with lower body masses, we expected a much lower detection probability (Boves and 251 Belthoff 2012, Teixeira et al. 2013, Santos et al. 2016, Barrientos et al. 2018) and therefore 252 were not considered in mortality estimations.

253 Regarding the search area (SA) along the road corridor, it is known that many 254 carcasses are projected off the road during the collision, and that injured animals may still be 255 able to move away from road lanes and shoulders. It follows that a proportion of the 256 carcasses can never be detected. From our experience in the study area, whose investigation includes occasional surveys on the verges, we know that some larger animals die away from 257 258 the road, while smaller species are projected far from the road lanes. Again, to tackle the 259 uncertainty on this parameter, we also used different values of SA for estimating total 260 mortality: 0.80, 0.85, 0.90, and 0.95, i.e. between 5-20% of all carcasses never fall into a 261 detectable area, the road pavement and verges.

Using GenEst, we estimated the total mortality for the six different transects, per body mass classes, and using the different combinations of SE and SA (12 models) considering *i*) the dataset of records >1kg and *ii*) only those species likely to cause material damage to vehicles. For each model, the accuracy of estimates was measured through 2000 replicates.

267 **2.5.3 Determining spatial aggregations of collisions**

268 The records pertaining to species likely to cause material damage were aggregated by body 269 mass class (<5, <30 and >30 kg) and by road sections of five km length. This segment length 270 was chosen to minimize the chance of spatial variation of hotspot location with time (Santos 271 et al. 2017), while also being a reasonable distance to apply mitigation measures such as 272 fencing (Seiler et al. 2016). For each transect and body mass class, we assumed that the 273 observed number of roadkill per road section would follow a random Poisson 274 distribution with a mean (λ) equal to the total number of roadkill divided by the total number 275 of road sections (Malo et al. 2004, Santos et al. 2017). The probability of any road segment 276 having *x* number of collisions was therefore:

$$p(x) = \frac{\lambda^x}{x! e^{\lambda}}$$

278 A mean value (λ) for each body mass class and road transect was calculated. We 279 considered a road section to be an aggregation of collisions, i.e. hotspot of mortality, 280 when p(x) > 0.95. We preferred using this conservative value of p to reduce the likelihood of 281 highlighting spurious aggregations, resulting from multiple comparisons (Borda-de-Água et al. 2019). We further assumed that the carcass persistence, searcher efficiency and search 282 283 area were equal within and outside the road sections classified as hotspots, and therefore the 284 proportion of animals not recorded in roadkill surveys was constant across each road transect. 285 For example, if a transect had 100 records and a total mortality estimated in 120 records (i.e. 286 20% increase), a given road section of that transect having a significant aggregation of 287 records, e.g. n=40, was assumed to have had a total mortality of 48 records (increase in 20%). 288

289 2.5.4 Cost-benefit analyses of road fencing

290 To estimate the material cost of collisions, we used the estimated total collisions with species 291 potentially causing vehicle damage (see 2.5.1). For each body mass class, we calculated the 292 mean of reported cost in interviews and online questionnaire, per vehicle type (car or truck). 293 We assumed that the probability of an animal being hit by a car or truck was proportional to 294 the traffic volume of each type of vehicle. Hence, the cost calculation considered the mean 295 traffic volume in each road transect, per vehicle type. For example, the estimated cost of 296 WVC in the R4 transect (with 33% of traffic of trucks and 66% of cars) was calculated as 297 follows:

298

299 0.33 * cost with trucks $(N_{<5} + N_{<30} + N_{>30}) + 0.66$ * cost with cars $(N_{<5} + N_{<30} + N_{>30})$, 300

301 where *N* are the number of individuals from total mortality estimates, of each body mass302 class.

303 The material costs were estimated separately for each full transect (considering all 304 WVC along the road) and for the sections were aggregations of collisions were detected 305 (WVC in hotspots). We then performed simple cost-benefit analyses for each road transect, 306 by estimating the return period in years, i.e. the time needed to pay off the investment made 307 in mitigation measures (fencing) if it avoided 100% of collisions with those larger species. 308 We also assumed a similar roadkill rate along time (i.e. without significant changes in the 309 number of collisions along time). The return period was calculated as the ratio between the 310 initial investment per km and the estimated cost per km and per year. We used as investment 311 value US\$ 34,056 per km of fencing (on both roadsides), obtained from the average budgets 312 of three companies that apply fences on MS roads (see Supplementary material S3).

313

314 3 Results

315 3.1 Roadkill systematic surveys

316 We recorded 10,942 carcasses of road-killed animals of which 9,431 had a body mass >1 kg, 317 including 4,208 larger animals likely to cause material damage with monetary cost to vehicles 318 (see Table S2.1). The roadkill rate was generally higher in the transect R4 for species 319 pertaining to body mass classes <5 and >30; and had similar values across transects for the 320 intermediate class <30 (Fig. 2). A considerable proportion of animals pertaining to weight 321 classes <30 and >30, mainly reptile species such as snakes and caimans, as well short stature 322 mammals such as six-banded armadillo (Euphractus sexcinctus) and nine-banded armadillo 323 (Dasypus novemcinctus), were also found in transect R4 (Fig. 2). The number of WVC seems 324 to be increasing with time in some transects, namely R2, R3A and R3B (Fig. 2).

325

326 [FIG. 2]

328 **3.2 Total mortality estimates**

329 Carcass persistence models for the three body mass classes had a higher fit using Weibull 330 distribution (Fig. S4.1 in Supplementary material S4). Overall, carcasses with >1kg were 331 detectable for 4.3 ± 3.7 days, being the probability of a carcass persisting 15 days < 60%, 332 with higher values for large body mass species (Fig. S4.1). Models of carcass persistence 333 including the effects of season and traffic were similar (AIC < 1) to those not including these 334 variables, and therefore we choose the simpler models (without those variables) to estimate 335 total mortality. Using the carcass persistence models and the different combinations of search 336 efficiency and search area, we estimated that total mortality of animals >1kg ranged between 337 (median and 90% CI) 11,983 (11,424 – 12,661) and 15,108 (14,192 – 16,214), of which 338 between 5,581 (5,245 – 5,965) and 7,072 (6,533–7,705) were with animals likely to cause 339 material damage with monetary cost to vehicles (Fig. 3). Overall, there was an 340 underestimation of mortality in roadkill surveys of ca. 22-38%. Hereafter, we will use two scenarios combining the extreme values of search efficiency (SE) and search area (SA), i.e. 341 342 the one having higher mortality estimates (SE: 0.85, SA: 0.80) and the lower mortality 343 estimates (SE: 0.95, SA: 0.95).

344

345

[FIG. 3]

346

347 **3.3** Spatial aggregations of collisions

We identified 43 road sections (ca. 19% of total road length) having a significantly higher
aggregation of collisions. We recorded in these hotspots 1,481 collisions with animals likely
to cause material damage with monetary cost to vehicles, i.e. ca. 34% of the mortality with

351	these species. The majority of these hotspots (n=18; 90 km) were in the transect R4 (Fig. 4).
352	Using the two scenarios of SE and SA combinations, we estimated that a total of $1,376 -$
353	1,744 (median values of each scenario) collisions were likely to have occurred in the hotspot
354	sections, which we used in vehicle costs estimates. Noteworthy, hotspots covered a lower
355	proportion of other smaller but highly road-killed species, including yacare caiman (Caiman
356	yacare; 9% of its mortality), black-and-white tegu (Salvator merianae; 18%), or six-banded
357	armadillo (29%) (see Table S2.1).
358	
359	[FIG. 4]
360	
361	3.4 Material costs in vehicles
362	We obtained information on material costs for 185 animal-vehicle collisions, of which 94%
363	referred to animals with body mass >1kg. None of these involved collisions with reptiles or
364	with short stature mammals, supporting our choice to focus on the greater rhea and larger
365	mammals as potentially causing material damages (Table S2.1). The average material cost
366	with animals >1 kg was US 885 \pm 1,575 (mean \pm SD). The mean cost per vehicle type (car and
367	truck) and body mass ranged between US\$ 65-2,133. Nine responses (5%) referred the total
368	loss of the vehicle, with costs averaging US\$ 6,505 \pm 1,626 (Fig. 5). The cost of hitting a >30
369	kg animal was considerably higher for trucks.
370	
371	[FIG. 5]
372	
373	The total costs resulting from the estimated mortality throughout the study period was
374	estimated to range between US\$ 4,455,730 and 5,645,009, with a cost value per road
375	kilometer and per year averaging between US\$ 1,171 and US\$ 1,492 across the road

transects, respectively for the lower and higher mortality scenarios. The hotspots of WVC
represented 19% of the roads but aggregated ca. 36% of the potential costs with vehicle
damage, i.e. US\$ 1,593,485 – 2,018,779.

379

380 **3.5 Cost-benefit analyses of road fencing**

381 Considering an initial investment of US\$ 39,436,848 to fence the whole surveyed transects 382 (1,158 km; US\$ 34,056 per km of fencing), we calculated the return period of investment 383 considering the estimated mortality from lower and higher mortality scenarios. Accordingly, 384 the return period would range between 16 and 40 years across the transects, if fencing the full 385 length of transects (Table 4). On the other hand, the investment required to mitigate the 386 hotspot sections would be significantly lower, US\$ 7,322,040, and would have a return 387 period ranging between 9 and 25 years. Transects with higher number of WVC with larger 388 animals would have a shorter return period, namely the transect R4 (Table 3).

- 389
- 390

[TABLE 3]

391 4 Discussion

392 We performed over three years of roadkill monitoring in which we recorded >10,000 WVC 393 along the 1,158 km. About 40% of collisions involved large animals that cause significant 394 material damage to vehicles, some of which represent a real threat to human lives. Moreover, 395 some of these larger species are of conservation concern and had considerable roadkill rates, 396 such as the lowland tapir (267 records) and giant anteater (608 records), probably threatening 397 their population persistence in the long term in this region (Medici et al. 2012, Desbiez et al. 398 2020). Furthermore, according to our estimates, the total mortality of species weighting >1 kg 399 was 22-38% higher than the one recorded and therefore these numbers are likely to be highly

conservative. Additionally, the number of WVC seems to be increasing, at least on some
surveyed roads, probably reflecting the increasing traffic volumes in Mato Grosso do Sul
(DNIT 2020). Overall, such high roadkill rates may have serious negative effects on wildlife
population viability (Ceia-Hasse et al. 2018, González-Suárez et al. 2018, Desbiez et al.
2020), as well on human safety (Shilling et al. 2019, Abra et al. 2019, Gren and Jägerbrand
2019), and therefore the implementation of mitigation measures at least in key areas of the
highway is highly necessary.

407 Fencing has been acknowledged as one of the most effective measure to reduce WVC, 408 particularly when combined with faunal passages (Jaeger and Fahrig 2004, Seiler et al. 2016, 409 Rytwinski et al. 2016, Spanowicz et al. 2020). There is evidence that several medium and 410 large mammals that inhabit the region use existing passages, including tapir, capybara and 411 giant anteaters (Abra et al. 2020). Hence, channel these species into passages through fences 412 would likely result in greater road mitigation effectiveness. However, the large-scale 413 implementation of fencing can be hindered due to the high initial investment required. Yet, 414 our study shows that when compared to the cost of material damage of vehicles, such 415 investments are likely to pay off in the medium term. According to our data, the initial 416 investment necessary to fence the whole surveyed transects would have a full return in an 417 average of 27 years. This period is shorter than most concession contracts, ca. 20-30 years. 418 Also, to put these investments in perspective, the World Bank estimates that new bituminous 419 two-lane highway costs ca. one million dollar per km (World Bank 2006), therefore 420 suggesting that fencing investment represents about 1% of the presumed construction cost. 421 If surrounding the entire road network simultaneously is unviable due to the high 422 budget required, focusing on road sections concentrating higher number of WVC involving 423 larger species - at least in a first phase - would provide a much higher cost-benefit relation. 424 We identified 43 sections that concentrated ca. 34% of WVC with larger animals. Fencing

425 these sections would require 19% of the initial investment with a full return in much shorter 426 periods. This is particularly evident in transect R4 which add most hotspots with larger 427 animals, and for which about 9-11 years would suffice to compensate the initial investment. 428 It should be noted that focusing on hotspots of mortality, while greatly reducing the investment required in mitigation, potentially fails to reduce significantly the mortality of 429 430 other species. Particularly in transect R4, we recorded a high number of reptiles as caiman 431 and short stature mammals as armadillos that would not be protected by exclusionary fencing, 432 if it was applied only in hotspots. Likewise, fencing is not effective in reducing collisions for 433 most birds. Yet, these species can be heavily impacted by roadkill (Loss et al. 2014, 434 González-Suárez et al. 2018), and may also represent a danger for traffic. For example, 435 although not considered in our estimates, our dataset contains records of accidents with 436 smaller birds, such as the red-legged seriema (Cariama cristata), causing material damages 437 averaging US\$ 520 (n=7). Therefore, road fencing in the sections with the highest mortality 438 should be considered an important step, but not an isolated one in the road mitigation process. 439 The average material cost per accident (US\$ 885±1,575) was considerably lower than 440 the one reported for other regions in Brazil such as in the São Paulo state US\$ 5,000 (Abra et 441 al. 2019), or in other studies from developed countries, namely US\$ 7,000 in Spain (Sáenz-442 de-Santa-María and Tellería 2015) and US\$ 11,900 (with large animals) in California 443 (Shilling et al. 2019). Such disparity of values probably stems from differences in the cost of 444 living across countries and regions of Brazil. Accordingly, the investment required to install 445 fences that we have obtained is considerably lower than the one reported by Seiler (2016) for 446 fences targeting ungulates in Europe, budgeted at US\$ 44,000 – 65,000 per km road. Hence, 447 for regions or countries with higher material costs, the return periods are probably similar to the ones estimated here. 448

449 The material damage in cars was in average higher when compared to trucks. This 450 was expected given the higher shaft height of trucks, that allows them to run over animals 451 without major damages. The exception was for trucks colliding with larger mammals (>30 452 kg), which had considerable higher costs, over US\$ 2,000, relatively to cars. When a truck 453 hits a lowland tapir, it collides against a 150-250 kg body mass animal, with a shoulder height 454 of 77 to 108 cm (Padilla and Dowler 1994). That is, it hits a large and heavy object, 455 potentially damaging different parts of the vehicle. Conversely, car passengers can be 456 seriously injured or killed when colliding with these animals. For example, between April 457 2013 and March 2020, 77 people were injured and 28 died in collisions with tapirs on 458 different roads of Mato Grosso do Sul (Patricia Medici pers. comm.). Moreover, in tropical 459 countries, roads have mostly two lanes, often without wide shoulders, and therefore the entry 460 of animals on the road may not be perceived by drivers, especially at night. Therefore, 461 preventing animals from accessing roads by diverting them to under or over passages is 462 imperative in regions where these large animals roam, both for their conservation and human 463 safety.

464 It should be emphasized that our estimates only considered the material damages in 465 vehicles and are therefore highly conservative figures. We did not include values on human lives and injuries, due to the difficulty in assigning them monetary value. Yet, when 466 467 available, these values would significantly lower the return period of investments (Wijnen 468 and Stipdonk 2016). For example, Sáenz-de-Santa-María and Tellería (2015) mentions 469 compensations of US\$ 1,655,750 for fatally injured people; US\$ 259,000 seriously injured 470 people; and US\$ 7,210 for minor injuries; and in São Paulo state, the average cost for a crash 471 with human injuries or fatalities is US\$ 29,813 (Abra et al. 2019). In addition, there are other 472 costs associated with WVC not counted here, namely those associated with traffic police and 473 medical assistance, road repair, traffic congestion and resulting delays, removal of carcasses,

474 searches for injured or dead animals and the value of animals lost. As an example, jaguar 475 (Panthera onca) ecotourism in Pantanal represents a gross annual income of US\$ 6,827,392 476 (Tortato et al. 2017), and therefore roadkill of these animals has a serious impact on both 477 biodiversity and local economy. Also, collisions often imply delays in traffic, with consequent costs, which can be even more problematic in Brazil, a country where most 478 479 transportation of goods often relies on trucks. On the other hand, to guarantee its 480 effectiveness, fences must be carefully planned and properly installed. For example, for 481 species capable of climbing it is required to use smaller mesh size or gravity walls, which are 482 known to provide an effective and long-lasting barrier impeding smaller animals to access the 483 road (Kenneth Dodd et al. 2004, Baxter-Gilbert et al. 2015) (see Supplementary material S3). 484 Furthermore, fences require regular inspection, maintenance and repair, which will raise the 485 associated costs.

486 Overall, accounting for all the different costs related with WVC, from material 487 damage to insurance payments and general road maintenance, would provide a more precise 488 evaluation of the economic savings of road fencing. However, using the costs associated only 489 to material damages in this simple cost-benefit analyses already provided valuable 490 information to help decision makers to evaluate the feasibility and need of implementing road 491 mitigation structures. Both the number of injured people and roadkill rates may worsen in the 492 future, with the increasing development of road networks and traffic volume worldwide 493 (Meijer et al. 2018). This is particularly true in countries of great diversity where new 494 development corridors are being planned, cutting into areas hitherto pristine (Laurance et al. 495 2015, Alamgir et al. 2017, Ascensão et al. 2018).

In conclusion, this study shows that investments in road mitigation using properlyinstalled fencing and connected to safe crossing passages, are likely to be paid off in medium

498	term. Investing in mitigation measures provides a win-win solution for increasing traffic
499	safety for humans and reduce road-related negative effects on biodiversity.
-	

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509

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- 656





660 Figure 1 – Location of the surveyed roads in Mato Grosso do Sul, Brazil. Surveyed

transects are shown in white. The study was conducted on federal roads - R1A, R1B
and R4 in BR262, and R3B in BR267; and on state roads - R2 (MS040) and R3A

663 (MS338); totaling 1,158 km. Main road network is depicted as black lines. Main land

664 use classes are forest (darker areas) and open areas, including agriculture and pastures

665 (lighter areas). Land use was reclassified from MapBiomas (Azevedo Sr et al. 2018).

666 The city Campo Grande is in the middle of the State. The Pantanal is the upper left667 region of the map.



Figure 2 – Roadkill rate (individuals per year and per 5 km) recorded along the 3-year
study period for the six road transects, per body mass class. Values are aggregated

671 according to the likelihood of a species causing or not material damage in vehicles.



673 Figure 3 – Mortality estimates (median and 90% CI) for all species weighting >1 kg

674 (top), and for the subset of species likely to cause damages in vehicles in collisions

(bottom), according to different levels of Search efficiency (x-axis) and Search area
(colors). Points are slightly scattered to allow a better reading, due to overlapping CI.



679 Figure 4 – Spatial patterns of roadkill (individuals per 5 km road section, per year),

680 involving species likely to cause material damage with monetary cost to vehicles, per
681 body mass class and road transect. Dots on top of each panel indicate hotspots of

682 mortality with probabilities of 0.95. Transects are oriented west-east (see Fig.1).





684 Figure 5 – Material costs from collisions with animals, by type of vehicle (truck and

685 car) and animal body mass class (<5, <30, >30 kg). Big circles represent the median

values of data collected (represented by the smaller dots, jittered to enable reading).

687 Bars indicate 5-95% quantile range.

688 Tables

690	Table 1 - Summary information of transects surveyed searching for roadkill, indicating
691	the length of transects in km, first date of survey, number of surveys, daily traffic
692	volume, and the proportion (%) of cars and trucks in traffic volume. Traffic volume
693	was obtained from official counts (DNIT 2020), except for MS040 and MS338 which
694	was assessed by the team using the same methodology of DNIT (2020). For R3A we
695	assumed the same traffic characteristics as R2 (contiguous and similar roads). BR262
696	and BR267 are federal roads, and MS040 and MS338 are state roads.

Transect ID	Road name	Length (km)	First systematic survey	Number of Surveys	Daily Traffic Volume (median)	Trucks (%)	Cars (%)
R1A	BR262	182	06/02/2017	78	3,633	39	59
R1B	BR262	129	07/02/2017	25	3,773	48	52
R2	MS040	224	07/02/2017	79	653*	37*	60*
R3A	MS338	66	07/02/2017	79	653*	37*	60*
R3B	BR267	218	27/02/2017	79	3,973	46	49
R4	BR262	339	29/03/2017	80	3,098	33	77

698	Table 2 – Survey	effort for carcass	persistence ex	periments: leng	th of transects,
	5		1	1 0	

number of daily surveys per season, and number of carcasses surveyed per body mass
class and season. Body mass classes: between 1-5 kg (<5), between 5-30 kg (<30) and

701 more than 30 kg (>30).

Transect ID	Length (km)	Surveys Wet / Dry	Body mass Class (kg)	Carcasses Wet / Dry	Total carcasses
			<5	66 / 13	79
R2	139	29 / 15	<30	24 / 20	44
			>30	8 / 3	11
			<5	- / 5	5
R3B	96	0 / 15	<30	- / 9	9
			>30	- / 1	1
			<5	72 / 112	184
R4	107	21 / 30	<30	26 / 45	71
			>30	11 / 8	19
Total				207 / 216	423

702

704 Table 3 – Estimated total costs associated with animal-vehicle collisions with large 705 animals (>1kg), and per km and year (includes costs from collisions of animals with 706 cars and trucks). All costs in US\$. Return period is the expected time in years for the 707 accumulated costs from material damage to match up the initial investment. Estimates 708 are given for the full transect length in top table and for the hotspots of mortality in 709 bottom table. Range values result from the different scenarios using Search Efficiency 710 and Search Area of 0.95 (lower costs, higher period values) and Search Efficiency of 711 0.85 and Search Area of 0.80 (higher costs, smaller period values).

Transect			Return
ID	Total cost (range)	Cost per km per year (range)	period
Fencing the	full transects		
R1A	664,222 - 857,192	1,170 - 1,510	29 - 23
R1B	122,275 - 158,151	948 - 1,226	36 - 28
R2	876,854 - 1,117,907	1,239 - 1,579	27 - 22
R3A	178,492 - 232,135	856 - 1,113	40 - 31
R3B	765,291 - 950,688	1,111 - 1,380	31 - 25
R4	1,848,596 - 2,328,935	1,704 - 2,147	20 - 16

Fencing the hotspot sections

0	I		
R1A	211,375 - 272,806	1,936 - 2,498	18 - 14
R1B	19,350 - 25,037S	1,935 - 2,504	18 - 14
R2	267,980 - 341,693	2,120 - 2,703	16 - 13
R3A	63,612 - 82,632	1,342 - 1,743	25 - 20
R3B	150,295 - 186,859	1,902 - 2,365	18 - 14
R4	880,873 - 1,109,752	3,059 - 3,853	11 - 9

713 Supplementary material

- 714
- 715 **S1 Google form**
- 716
- 717 The form can be found in the following URL:
- 718 <u>https://docs.google.com/forms/d/1fZ57Y8IJcARRKKzm7uOqDPAJN69VkQFF5cJmuxz41x</u>
- 719 <u>k/edit</u>

511	vestres
Este (<u>ww</u> Que colis med	questionário é liderado pelo ICAS - Instituto de Conservação de Animais Silvestres <u>w.icasconservation.org.br</u>). É muito breve, mas a sua informação é muito importante. remos recolher informações sobre custos materiais nos veículos resultantes de iões com animais silvestres. O objetivo é fazer uma avaliação custo-benefício de idas mitigadoras de colisões (por exemplo, instalação de cercas).
Se ti por a	ver informação para mais do que um acidente, por favor responda a um questionário acidente. Muito obrigado!
*Rec	juired
0	la accuración de sidente 2
Qual	a estrada e localização aproximada? Por exemplo, cidade mais próxima ou Município.
Your	answer
Em	que dete (annuime demente)
⊏rn Pode	colocar o ano, se não souber mais detalhe
V ~ · · ·	000000

720

Fig. S1.1 - Snapshot of the first questions in Portuguese of the Google form asking
about the date, place and period of the day (day, night, sunset, sunrise) of the collision
events; the type of vehicle (car, truck, other); name or description of the species
involved in collisions; and the cost of the material damage, if any.

- 725
- 726

727 S2 – Roadkill summary information

728 Table S2.1 – Summary of roadkill recorded by number (N), body mass class, and the

729 likelihood of causing material damage in vehicles when involved in collisions

730 (Damage). Taxa are sorted according to BM and alphabetic order. Body mass classes:

between 1-5 kg (<5), between 5-30 kg (<30) and more than 30 kg (>30).

Class	Species / taxa	Ν	Body mass	Damage
Amphibians	Amphibians	203	< 0.1	no
Reptiles	Reptiles <1kg	68	<1	no
	Boiruna maculata	1	<5	no
	Bothrops moojeni	2	<5	no
	Dracaena paraguayensis	18	<5	no
	Drymarchon corais	1	<5	no
	Iguana iguana	13	<5	no
	Palusophis bifossatus	1	<5	no
	Spilotes pullatus	2	<5	no
	Other reptiles <5kg	204	<5	no
	Boa constrictor	34	<30	no
	Crotalus durissus	13	<30	no
	Eunectes notaeus	72	<30	no
	Hydrodynastes gigas	25	<30	no
	Paleosuchus palpebrosus	1	<30	no
	Salvator merianae	246	<30	no
	Other reptiles <30kg	10	<30	no
	Caiman latirostris	35	>30	no
	Caiman yacare	849	>30	no
	Eunectes murinus	16	>30	no
	Other reptiles >30kg	10	>30	no
Birds	Ammodramus humeralis	1	< 0.1	no
	Brotogeris chiriri	6	< 0.1	no
	Campylorhamphus trochilirostris	1	< 0.1	no
	Columbina talpacoti	1	< 0.1	no
	Crotophaga ani	26	< 0.1	no
	Eupsittula aurea	47	< 0.1	no
	Icterus croconotus	1	< 0.1	no
	Myiozetetes similis	1	< 0.1	no
	Nyctidromus albicollis	6	< 0.1	no
	Nystalus chacuru	1	< 0.1	no

Class	Species / taxa	Ν	Body mass	Damage
	Paroaria capitata	6	< 0.1	no
Birds (cont.)	Passer domesticus	2	< 0.1	no
	Patagioenas picazuro	1	< 0.1	no
	Pitangus sulphuratus	17	< 0.1	no
	Polytmus guainumbi	1	< 0.1	no
	Sicalis flaveola	1	< 0.1	no
	Taraba major	1	< 0.1	no
	Thamnophilus doliatus	4	< 0.1	no
	Trogon curucui	1	< 0.1	no
	Turdus rufiventris	1	< 0.1	no
	Tyrannus melancholicus	2	< 0.1	no
	Xolmis velatus	2	< 0.1	no
	Other birds <0.1kg	158	< 0.1	no
	Alipiopsitta xanthops	2	<1	no
	Amazona aestiva	15	<1	no
	Amazona amazonica	1	<1	no
	Aramides cajanea	7	<1	no
	Aratinga nenday	2	<1	no
	Athene cunicularia	59	<1	no
	Campephilus melanoleucos	1	<1	no
	Colaptes campestris	12	<1	no
	Columba livia	1	<1	no
	Crotophaga ani	26	<1	no
	Crotophaga major	2	<1	no
	Crypturellus parvirostris	1	<1	no
	Cyanocorax chrysops	4	<1	no
	Cyanocorax cyanomelas	1	<1	no
	Dendrocygna viduata	1	<1	no
	Falco sparverius	7	<1	no
	Guira guira	49	<1	no
	Heterospizias meridionalis	3	<1	no
	Jacana jacana	1	<1	no
	Megaceryle torquata	2	<1	no
	Megascops choliba	5	<1	no
	Melanerpes candidus	3	<1	no
	Myiopsitta monachus	1	<1	no
	Nothura maculosa	4	<1	no

Class	Species / taxa	Ν	Body mass	Damage
	Nyctibius griseus	2	<1	no
Birds (cont.)	Ortalis canicollis	15	<1	no
	Piaya cayana	12	<1	no
	Primolius auricollis	1	<1	no
	Psittacara leucophthalmus	2	<1	no
	Pteroglossus castanotis	5	<1	no
	Ramphastos toco	32	<1	no
	Rhynchotus rufescens	24	<1	no
	Rupornis magnirostris	10	<1	no
	Tigrisoma lineatum	1	<1	no
	Tyto alba	12	<1	no
	Zenaida auriculata	1	<1	no
	Other birds <1kg	583	<1	no
	Aburria cumanensis	1	<5	no
	Ara ararauna	4	<5	no
	Ara chloropterus	1	<5	no
	Aramus guarauna	6	<5	no
	Cairina moschata	1	<5	no
	Caracara plancus	303	<5	no
	Cariama cristata	273	<5	no
	Cathartes aura	2	<5	no
	Cathartes burrovianus	2	<5	no
	Coragyps atratus	63	<5	no
	Crax fasciolata	2	<5	no
	Geranoaetus albicaudatus	2	<5	no
	Other birds <5kg	42	<5	no
	Rhea americana	20	<30	yes
Mammals	Mammals <1kg	42	<1	no
	Alouatta caraya	6	<5	yes
	Cabassous unicinctus	67	<5	no
	Chironectes minimus	2	<5	yes
	Dasyprocta azarae	44	<5	yes
	Dasypus novemcinctus	723	<5	no
	Dasypus septemcinctus	1	<5	no
	Didelphis albiventris	37	<5	yes
	Eira barbara	29	<5	yes
	Euphractus sexcinctus	1831	<5	no

Class	Species / taxa	Ν	Body mass	Damage
	Herpailurus yagouaroundi	8	<5	yes
Mammals (cont.)	Leopardus braccatus	2	<5	yes
	Leopardus wiedii	1	<5	yes
	Lycalopex vetulus	52	<5	yes
	Nasua nasua	100	<5	yes
	Sapajus cay	4	<5	yes
	Sylvilagus brasiliensis	16	<5	yes
	Tamandua tetradactyla	549	<5	yes
	Other mammals <5kg	251	<5	no
	Cerdocyon thous	1614	<30	yes
	Chrysocyon brachyurus	16	<30	yes
	Cuniculus paca	3	<30	yes
	Leopardus pardalis	14	<30	yes
	Lontra longicaudis	9	<30	yes
	Mazama americana	6	<30	yes
	Mazama gouazoubira	33	<30	yes
	Myrmecophaga tridactyla	608	<30	yes
	Pecari tajacu	5	<30	yes
	Procyon cancrivorus	135	<30	yes
	Speothos venaticus	3	<30	yes
	Other mammals <30kg	95	<30	no
	Blastocerus dichotomus	7	>30	yes
	Hydrochoerus hydrochaeris	578	>30	yes
	Panthera onca	1	>30	yes
	Priodontes maximus	3	>30	yes
	Puma concolor	6	>30	yes
	Sus scrofa	11	>30	yes
	Tapirus terrestris	267	>30	yes
	Tayassu pecari	19	>30	yes

734 S3 - Suggested fence characteristics

In this study, the fences are the main measures discussed to be adopted, properly linked to existing road passages. In the studied transects there are several underpasses, including cattle boxes, drainage culverts and bridges. In R2 alone, there are 47 underpasses (39 cattle boxes and 8 bridges). Livestock management between roadsides is performed under the road, in large concrete structures with dimensions $\geq 2 \times 2$ meters. Such structures allow the crossing of wild animals, including large ones such as tapir, giant anteater and deer species.

741 Fences must prevent medium and large wild animals from entering the road. They 742 should be at least 1.70 meters high and 30 cm buried in the ground, have a concrete wall or 743 fiber cement screen in the lower part of the fence in contact with the ground and metallic wire 744 mesh up to 10 x 10 cm and metal or concrete posts (Fig. S3.1). This design is recommended 745 to bar the entry of animals that try to pass between the fence lines (e.g Leopardus sp.), jump 746 over the fence (e.g. Mazama gouazoubira, Mazama americana, Ozotoceros bezoarticus), dig 747 under the fence (e.g. Dasypus sp., Euphractus sexcinctus, Priodontes maximus) or that have 748 enough strength to trying to push into the fence (e.g. *Hydrochoerus hidrochoerus*, *Tapirus* 749 terrestris). Yet, the recommended design may only discourage climbing animals from 750 overcoming the fence, but not completely bar their entrance to the highway (e.g. Nasua 751 nasua, Didelphis sp.).

The cost of this fence type averages US\$ 34,056 per km (in both sides), a value that was obtained from three companies that apply fences on MS roads (values in US\$: 44,280, 31,680 and 26,208).



756



- Figure S3.1 The design of fence projected as a mitigation to decrease WVC (A), and in
- 760 combination with wildlife underpasses, water culverts or cattle boxes (B). Source:
- 761 ViaFAUNA Environmental Studies.



Fig S4.1 - Carcass persistence probability with time (days), per body mass class. Stairshape line is the Kaplan Meier estimator, continuous line is the Weibull distribution
fitted to data. Respective confidence intervals are plotted in dotted line. Dotted blue
lines indicate the median persistence probability for the time interval of systematic
surveys.