Short title: Wildlife is imperiled in peri-urban landscapes

Wildlife is imperiled in peri-urban landscapes: threats to arboreal mammals

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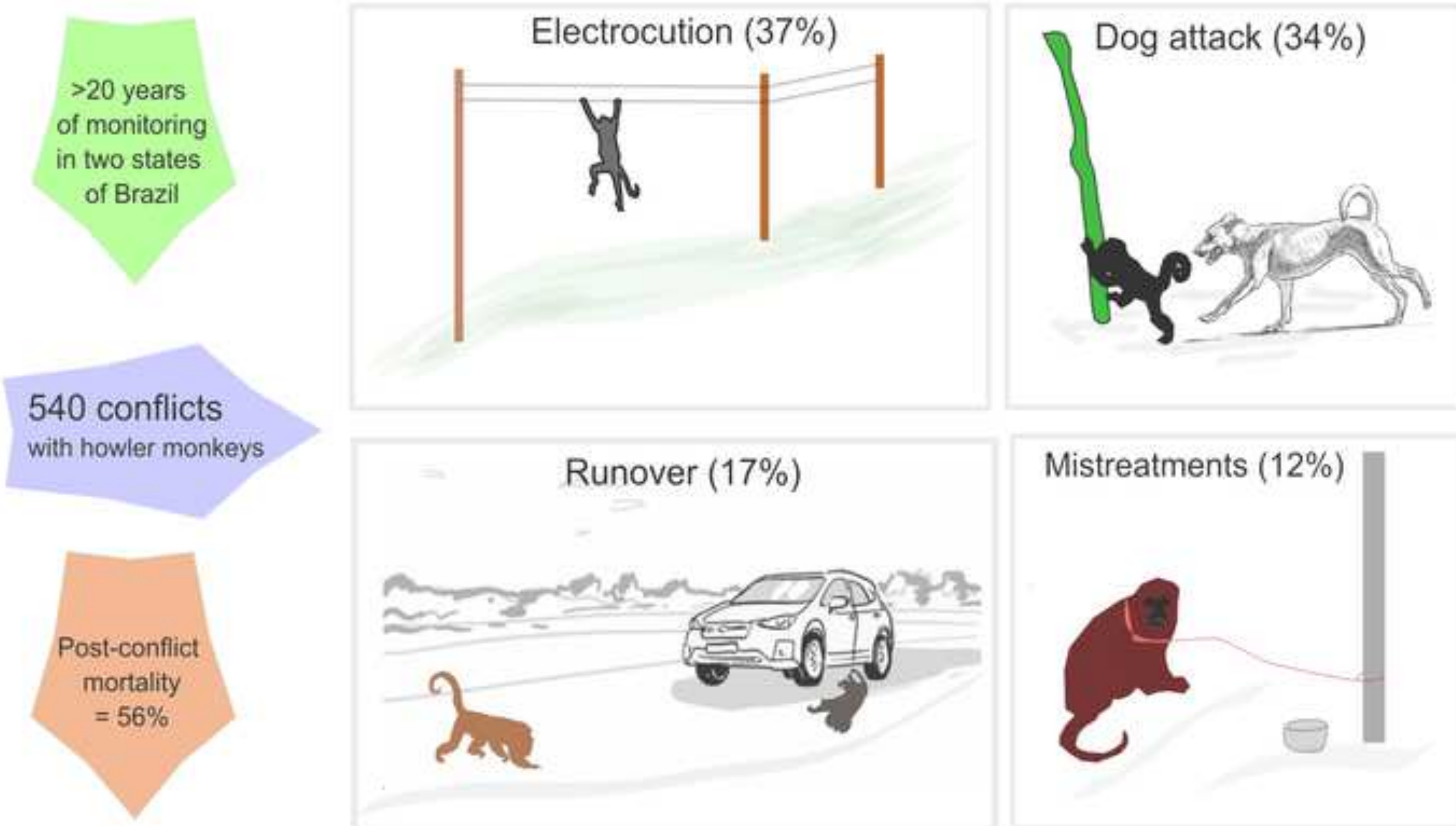
Graphical Abstract



HIGHLIGHTS

- We investigated the threats faced by brown howler monkeys in peri-urban regions of southern Brazil.
- After 20+ years of monitoring, we compiled 540 conflicts between monkeys and humans in the two study regions.
- The main conflicts were electrocution (37%), dog attack (34%), run over (17%), and mistreatment (12%).
- Lethal injuries ranged from 5% to 69% depending on conflict type.
- State, conflict type, and age-sex class explained 83% of the variation in conflict-related mortality.

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ABSTRACT

Urbanization and deforestation impose severe challenges to wildlife, particularly for forest-living vertebrates. Understanding how the peri-urban matrix impacts their survival is critical for designing strategies to promote their conservation. We investigated the threats faced by brown howler monkeys (*Alouatta guariba clamitans*) in peri-urban regions of Rio Grande do Sul (RS) and Santa Catarina (SC) states, southern Brazil, by compiling conflicts reported over more than two decades. We assessed the major conflicts, their distribution among age-sex classes, and the predictors of conflict-related mortality using GLMMs. After 20+ years of monitoring, we compiled 540 conflicts (RS=248 and SC=292). Electrocution in power lines was the most frequent cause of death or injury (37%), followed by dog attack (34%), run over (17%), and human mistreatment (12%). The occurrence of lethal injuries ranged from 5% to 69% depending on the type of conflict and state. The overall post-conflict mortality was 56%. Adult males and females were the major victims in both states. The minimal adequate GLMM explained 83% of the variation in conflict-related mortality. State, conflict type, and age-sex class were the main predictors of mortality. Overall, mortality was lower in SC and after human mistreatment, and higher among adult females than in the other classes. We found that the survival of brown howlers in the forest-urban interface is constrained by both the urban infrastructure and the growing interactions with humans and domestic and stray dogs. We propose the placement of aerial bridges, road signs and speed bumps in areas of frequent animal crossing, the sterilization of stray dogs, and the sensitization of local inhabitants on the importance of respecting and protecting wildlife to reduce their conflicts with humans and domestic animals in the forest-urban interface.

Keywords:
Alouatta guariba clamitans; Atlantic Forest; electrocution; dog attack; run over; human-wildlife conflict; environmental disturbance

1. Introduction

The accelerated destruction of natural habitats by human activities, particularly the expansion of farming, cattle ranching and urbanization (United Nations, 2015; Piano et al., 2020), has resulted in large-scale biodiversity loss (Ceballos et al., 2015; Estrada et al., 2017; Piano et al., 2020). Urban expansion in regions characterized by fragmented landscapes is particularly critical because it imposes additional pressures on threatened species that increase the risk of local extirpation (United Nations, 2015; Salomão et al, 2019; Piano et al., 2020), particularly when the adaptations of the remaining wildlife to the urban landscape increase their encounters with humans (Schell et al., 2020). Therefore, identifying the main threats faced by wildlife in peri-urban landscapes (i.e. mixed landscapes of rural and urban elements that experience intense human pressure: Douglas, 2006) is the first step to developing appropriate conservation strategies aimed at preventing or mitigating their impacts on wild populations.

Urbanization-related processes cause negative impacts on animals worldwide (e.g. butterflies and dung beetles: Salomão et al., 2019; Piano et al., 2020; reptiles: Gonçalves et al., 2018; birds: Bernardino et al., 2018; primates and other mammals: Bueno et al., 2015; Katsis et al., 2018; Al-Razi et al., 2019; Jones-Román et al., 2021; wild terrestrial vertebrates in general: Villatoro et al., 2019; Rodríguez et al., 2020; Teixeira et al., 2020). Roads, power lines, houses/buildings, and areas inhabited by domestic and stray dogs increase the risk of death of dispersing individuals, thereby compromising gene flow between isolated populations immersed in impermeable or semi-permeable urban matrices (Sol et al., 2013; Bernardino et al., 2018; Schell et al., 2020).

Arboreal tropical primates are among the most vulnerable vertebrates to urbanization because of their high dependence on emergent trees (Peres, 1994; Arroyo-Rodríguez and Dias, 2009; Rovero et al., 2015) and because the human encroachment into their habitats is growing (Estrada et al., 2017). While the impact of forest fragmentation, selective logging and hunting on primate behavior and demography has received significant attention (e.g. *Procolobus rufomitratus* and *Colobus guereza*: Gillespie and Chapman, 2008; *Alouatta* spp.: Arroyo-Rodríguez and Dias, 2009; *Ateles geoffroyi*: Chaves et al., 2011; see also Marsh, 2003; Marsh and Chapman, 2013), the impact of urbanization on primate survival has been often neglected (but see Gordo et al., 2013; Bicca-Marques, 2017; Katsis et al., 2018; Cunneyworth and Slade, 2021).

Primates inhabiting small habitat patches (i.e. <10 ha; *sensu* Marsh et al., 2003), which are often immersed in peri-urban landscapes, tend to face higher levels of food scarcity, physiological stress and spatial isolation among other adverse consequences of living in these environments (Fahrig, 2003; Arroyo-Rodríguez and Dias, 2009, Bicca-Marques et al., 2020). Species that cope with these peri-urban stressors can exploit food patches containing native and cultivated plants and human-provisioned or wasted foods in the matrix (e.g. *Alouatta guariba clamitans*: Chaves and Bicca-Marques, 2017; Corrêa et al., 2018; Back and Bicca-Marques, 2019; *Cebus imitator*: Mckinney, 2011; *Saguinus bicolor*: Gordo et al., 2013). However, peri-urban primates are also exposed to the aforementioned intense vehicle traffic in roads and highways, powerline networks, dog attacks and human mistreatment while navigating between food patches (e.g. Lokschin et al., 2007; Buss, 2012; Gordo et al., 2013; Bicca-Marques, 2017; Bicca-Marques et al., 2020; Cunneyworth and Slade., 2021; Jones-Román et al., 2021).

This scenario illustrates the encroached Atlantic Forest landscapes (Ribeiro et al., 2009), where 19 of the 27 nonhuman primates are endemic (Culot et al., 2019), three are Near Threatened, four are Vulnerable, seven are Endangered and five are Critically Endangered (IUCN, 2021). Although the Atlantic Forest is the most developed and populated Brazilian biome (Mittermeier et al., 2004), the impact of urbanization on the conservation status of its threatened primate fauna is poorly known.

The brown howler monkey (*Alouatta guariba clamitans*) is a Vulnerable (Buss et al., 2019) endemic Atlantic Forest primate found in isolated forest patches immersed in peri-urban and rural landscapes of south and southeastern Brazil. The taxon's ecology and behavior are well-known, particularly in south Brazil (Martins, 2006; Buss, 2012; Chaves and Bicca-Marques, 2013, 2017; Chaves et al., 2018; Corrêa et al., 2018; Back and Bicca-Marques, 2019). However, the lack of long-term data on the influence of peri-urban threats on its populations compromises our assessments of their conservation importance.

In this study we compiled almost three decades of data on conflicts involving free-ranging brown howlers in peri-urban and rural landscapes in the two southernmost Brazilian states (Rio Grande do Sul and Santa Catarina, hereafter RS and SC, respectively). Specifically, we assessed (i) the types of conflict and their relative frequency, (ii) the level of physical harm caused by each conflict, (iii) the proportion of howlers that recovered from distinct external injuries and the proportion of those that were released back into their habitats, (iv) the relationship between age-sex class and the frequency of each type of conflict, (v) the role played by season and day of the week on the frequency of conflicts, and (vi) the potential predictors of conflict-related outcomes (i.e. if animals survived or died because of the conflict). Based in the aforementioned, we hypothesize that brown howlers are imperiled in peri-urban areas in both states because the presence of hostile urban elements such as power lines, roads, and domestic dogs. In light of our findings, we propose management strategies to prevent and reduce the occurrence of conflicts and fatalities involving howler monkeys and other arboreal mammals in peri-urban landscapes.

2. Materials and methods

2.1. Study species

Brown howler monkeys, alike their congenerics, are known for their high resilience to habitat disturbance, which has been associated with their highly flexible folivorous-frugivorous diet, including the exploitation of cultivated foods (Dias and Rangel-Negrín, 2015; Chaves and Bicca-Marques, 2016, 2017), and their home ranges often <15 ha. Brown howler populations in peri-urban areas in southern Brazil are commonly confined to small (<10 ha) private forest fragments (Printes et al., 2010; Chaves and Bicca-Marques, 2013; Corrêa et al., 2018). These discrete subpopulations may interact in patchy metapopulations and persist longer than those more isolated in the landscape. Therefore, they may play an important role for the conservation of this threatened species that is also highly susceptible to outbreaks of yellow fever (Almeida et al., 2012; Bicca-Marques et al., 2017; Buss et al., 2019).

2.2. Study area and forest remnants

In RS, we conducted this study in a ca. 200-km^2 region in the municipalities of Viamão and Porto Alegre, particularly in the peri-urban and rural areas of Viamão and the district of Lami (Fig. 1, Table 1). We focused >90% of our sampling effort in an area of 110 km^2 (Fig. 1). In SC, we monitored a ca. 800-km^2 peri-urban region in the municipalities of Blumenau, Indaial, Pomerode and Jaraguá do Sul (Fig. 1, Table 1). Additionally, we occasionally monitored other districts of Porto Alegre, RS, and municipalities along the coastal region of SC when local inhabitants reported conflicts with howler monkeys (Fig. 1, Table S1).

Human populations grew as little as 5% in Viamão to as much as 21% in Indaial from 2000 to 2010, reaching densities ranging from 160 people/km^2 in Viamão to 596 people/km^2 in Blumenau (Table 1). Most people (≥86%) live in peri-urban areas in the study regions. The number of houses vary from about 4,000 in Lami to 97,000 in Blumenau (IBGE, 2020; Table 1). Most of the study areas are surrounded by <1-ha to 100-ha Atlantic Forest fragments and scattered vegetation corridors in different successional stages. Subtropical semideciduous forests dominate the vegetation in both study regions.

Regardless of fragment size and level of official environmental protection, brown howlers that move between habitat patches in these peri-urban landscapes face risks imposed by roads, power lines, and human settlements (Table 1). These structural elements together with pastures and cultivated lands reduce matrix permeability, compromising inter-path connectivity. Despite these threats for moving and dispersing individuals and the human pressures on the plant community structure of habitat patches (e.g. selective logging, residential development), brown howler populations have persisted.

2.3. Data collection

We recorded the conflicts between 1995 and 2021 in RS and between 1991 and 2020 in SC (Fig. S1). We used four sources of information: (i) our own field observations, and reports from local (ii) inhabitants, (iii) environmental authorities (i.e. municipal and state environmental secretariats, Polícia Militar Ambiental/SC), and (iv) wildlife rehabilitation centers and veterinary hospitals and clinics (Porto Alegre and Viamão, RS). We visited the location of ~70% of reported conflicts to record the following information: geographic coordinates using a Garmin GPS, type of conflict (electrocution or sub-lethal injuries in power lines, EL; run over by any kind of motor-vehicle, RO; domestic dog attack, DA; and human mistreatment (including illegal captivity and physical mistreatments, MT; Fig. 2), injury level (mild-medium, severe, or lethal) based on the external injuries (Fig. S2), and, whenever possible, the fate of the injured individual. Type MT involved firearm shooting, stoning, and illegal captivity. The last is associated with chaining, inadequate feeding, precarious sanitary conditions, and lack of veterinarian care.

The mild-medium injury level included minor scratches that did not require prompt veterinarian care (e.g. slight skin-burns and teeth loss) and injuries that required surgery (e.g. bleeding, multiple dog bites, bone fractures, amputation of fingers, limbs or tail; Fig. S2). Severe injuries included multiple wounds that could lead to death without urgent veterinarian intervention (Fig. S2). These injuries often impeded the release of the individual back into the wild upon its recovery. Finally, lethal injuries often caused the howler's death up to 5 h after the conflict.

Whenever possible, we frozen the carcasses in the collection of biological material of CEPESBI in SC, and in the Laboratório de Primatologia or the Museu de Ciências e Tecnologia/PUCRS, or the Museu de Ciências Naturais (SEMA/RS) in RS. A subsample of carcasses from RS was necropsied in a study on the taxon's helminth parasite fauna (Jesus et al., 2021). Injured howlers were rescued by local authorities, researchers, or volunteers, whom, then, sent them to veterinarian hospitals/clinics or authorized wildlife rehabilitation centers. The full conflict dataset is available in Chaves et al. (2021).

2.4. Database limitations

Although we recorded conflicts during almost three decades in each study region, we are conservative in extrapolating and interpreting our findings because of the following limitations inherent of this kind of long-term study. First, we certainly missed conflicts (Fig. 1) that were not reported by local people or not forwarded to us by local authorities. This situation is more likely when the injuries were mild-medium and when the monkey returned to its group soon after the conflict (OMC, personal observation). Second, our sampling effort varied over time (Fig. S1) given temporal changes in the number of researchers, volunteers, and local informants. In this respect, the 1990s were poorly sampled because of a lack of volunteers or institutional groups to rescue the monkeys. Finally, local environmental authorities were more active collaborators in SC than in RS. This difference may explain the greater number of records of human mistreatment in SC. Despite these limitations, our database represents a useful description of the main threats faced by brown howlers living in the forest-urban interface for promoting their conservation via the design of appropriate management strategies.

2.5. Characterization of the study peri-urban matrices

We estimated 10 physical traits of the peri-urban matrices for those conflicts for which we have precise geographic coordinates, date of occurrence, type of conflict, and injury level (*n* = 335, 212 in SC and 123 in RS, see Table 2) to assess their relationship to conflict lethality: (1) matrix element where the conflict occurred, (2) conflict type, (3) number of houses within a 500-m radius from the location of the conflict, (4) total number of elements in the peri-urban matrix (e.g. roads, houses, buildings, airports, power lines, gardens, orchards, pastures, and others) within a 500-m radius from the location of the conflict, (5) type of road (primary or secondary), (6) road material (paved or unpaved), (7) distance to the nearest road, (8) distance to the nearest small forest fragment <10 ha, (9) distance to the nearest ≥70 ha-forest fragment, and (10) distance to the nearest house. We estimated these traits by exporting the GPS locations of the conflicts from the software Map Source 6.16.3 (Garmin®) to Google Earth Pro (Google®). For each GPS position we chose a high-resolution satellite image (with a low percentage of clouds and shadows) of the year of the conflict using the option 'historic images,' which includes images from 2002 to 2019. We analyzed Landsat 5 images in the software QGIS 3.6 to estimate the traits for those conflicts that occurred between 1991 and 2001 (*n*=46).

2.6. Statistical analyses

We performed Chi-square tests for proportions using the R function ‘prop.test’ to compare the proportions of occurrence between types of conflict, age-sex classes, seasons, and days of the week. When we found significant differences, we compared the proportion of records in each hour interval via post-hoc proportion contrasts using the R function ‘pairwise.prop.test’ with a Bonferroni correction. We performed generalized linear mixed models (GLMM; Zuur et al., 2009) using the function ‘lmer’ of the R package lme4 to assess the influence of the 16 predictor variables listed in

Table 2 on conflict-related mortality. We set the binomial family error for the response variable (i.e. if animals died or survived after the conflict) and a log link for running the models. We specified the 16 variables as fixed factors and the sampled year-ID as random factor to account for repeated-measures during the same years. We considered only two second-order interactions that are ecologically relevant, namely conflict type*matrix element and conflict type*age-sex class, to minimize overparameterization and problems of convergence of the global model (the model containing all fixed and random factors) due to the inclusion of a large number of variables and their interactions (Grueber et al., 2011). Before running this analysis we tested the variables for multicollinearity problems using the 'vifstep' function of R package dplyr. We included all variables in the global model because their Variance Inflation Factors (VIFs) were <3.

Then, we used the model simplification procedure to determine the minimal adequate (most 'parsimonious') model. In this method, the maximal model is simplified over a stepwise procedure until a model that produces the least unexplained variation or the lowest Akaike's Information Criterion (AIC) is found (Crawley, 2012). We used the AICc to select the 'best' model as recommended when sample size/number of predictor variables <40 (Burnham and Anderson, 2003). We used a likelihood ratio test over the R function 'anova' to test the significance of the 'best model' in comparison with the null model (the model including only the random factor). Finally, we used the 'r.squaredGLMM' function of the R package MuMIn (Barton, 2016) to estimate an equivalent of the coefficient of determination or pseudo-R^2 for the 'best' GLMM. The datasets used to perform these analyses are available in Chaves et al. (2021). We run all statistical analyses in R v.3.6.3 (R CoreTeam, 2020).

3. Results

3.1. Major conflicts involving brown howlers in peri-urban matrices

We recorded 540 conflicts involving brown howlers in the peri-urban matrices of RS (n = 248) and SC (n =292), from which we discarded 56 from further analysis because of incomplete information on the date, conflict type and/or injury level. Then, we collected complete information for 484 conflicts. In addition to our main study regions, our records included conflicts in other 11 municipalities in RS and 24 in SC (6% and 22% of state conflicts, respectively, Table S1). The major conflicts were electrocution (37% of 488 conflicts with complete information), followed by dog attack (34%), run over (17%), and human mistreatment (12%, Figs. 2 and 3 A-C). The vast majority of conflicts occurred at daytime when howler monkeys walked on power lines (Fig. 2 A-D), tried to cross paved or unpaved roads (Fig. 2 E-H), or descended to the ground (Fig. 2 I-J) to cross canopy gaps or to move between forest patches. A high percentage of the dog attacks (66% in RS and 49% in SC; Fig. 3 A, C) were lethal. In none occasion the killing dog(s) ate the monkey's flesh. Dog attacks involved stray and domestic dogs, and in all cases, they abandoned the carcass *in situ* upon the monkey's death. Furthermore, howlers kept illegally in captivity, commonly infants and juveniles, represented most records of human mistreatment (87% of 60 conflicts). The remaining cases were howlers shot with ball-bearing guns by local inhabitants.

The representation of each type of conflict varied between RS and SC. The number of records also differed among conflict types in RS (EL=43% of 222 conflicts, DA=31%, RO=18%, and MT=8%; χ^2=81, d.f.=3, P<0.0001; contrasts, P<0.05 in all significant comparisons; Fig. 3 A) and SC (DA=37% of 262 conflicts, EL=31%, RO=17%, and MT=15%; $\chi^2=45$, d.f.=3, $P<0.0001$, contrasts, $P<0.05$ in all significant comparisons, Fig. 3 B).

3.2. Injury level suffered by the howlers after a conflict

Most RS howlers involved in EL (54%), DA (66%) and RO (69%) suffered lethal injuries (contrasts, $P<0.05$ in all significant comparisons), and the remaining individuals survived with mild-medium (38%, 26%, and 21%, respectively) or severe injuries (8%, 7%, and 10%, respectively; Fig. 3 A). A higher proportion of the animals involved in EL suffered lethal or mild-medium injuries than severe injuries, while a higher proportion of those involved in DA and RO suffered lethal than mild-medium or severe injuries (Fig. 3 A, contrasts, $P<0.05$ in all significant comparisons).

Lethal injuries were less frequent in SC howlers after EL (48%), DA (49%) and RO (36%; Fig. 3 B). The other howlers involved in these conflicts survived with mild-medium (42%, 39%, and 57%, respectively) or severe injuries (10%, 13%, and 7%, respectively; Fig. 3 B). The proportion of victims of these conflicts with lethal or mild-medium injuries was higher than the proportion with severe injuries (Fig. 3 B, contrasts, $P<0.05$ in all significant comparisons). A higher proportion of howlers involved in MT suffered mild-medium (RS=11%. SC=10%) than severe or lethal injuries (RS=5%, SC=15%; Fig. 3 A, B, contrasts, $P<0.05$ in all significant comparisons).

Finally, 56% (269 out of 484 conflicts, Table S2) of the howlers with lethal injuries or with mild-medium or severe injuries that were alive immediately after the conflict died after <1 to 8 h during the transport to the veterinarian clinic or during the emergency veterinarian care. The health problems associated with their deaths included cardiorespiratory problems, lung perforations, internal hemorrhages, myases, and mutilations. This mortality represented 61% and 51% of the total number of conflicts with complete data reported for RS and SC, respectively (Table S2). Injured and/or mutilated survivors that were kept for life in public or private wildlife rescue centers represented 25% (RS) and 15% (SC), whereas individuals released back to their habitats summed only 7% (RS) and 2% (SC). The fate of the remaining survivors is unknown.

3.3. Conflict distribution among age-sex classes

Conflicts involved all age-sex classes in both states with a bias towards adult males and adult females (RS: χ^2=115, d.f.=7, P<0.0001; SC: χ^2=158, d.f.=7, P<0.0001; contrasts, P<0.05 in all significant comparisons; Fig. 3 C, D). The proportion of records per conflict type was often similar within each age-sex class (Fig. 3 C, D). The exceptions were higher EL proportions of records than MT for adults of both sexes in RS and for adult females in SC. In SC, juvenile males were more impacted by DA than by RO, and juvenile females were more impacted by DA than by EL and MT (contrasts, P<0.05 in all significant comparisons, Fig. 3 D).

3.4. Temporal patterns in the number of conflicts

The average number of conflicts per year (mean ± SD) was similar between RS and SC (13 ± 8 *vs* 12 ± 9 conflicts, respectively, Fig. S1). There was a higher frequency of conflicts in the summer and fall than in the winter in RS (χ^2=30, d.f.=3, P<0.0001; contrasts, P<0.05 in all significant comparisons, Fig. 4 A). The number of conflicts also differed among months in each season (χ^2 ranged from 16 to 57, d.f. ranged from 3 to 4 in all cases, P<0.001 in all cases; Fig. 4 A). The month(s) with the highest number of conflicts in summer, fall, winter, and spring were, respectively, January and March, April and May, August and September, and October (contrasts, P<0.05 in all significant comparisons, Fig. 4 A). In contrast, the frequency of conflicts in SC was similar in all seasons (χ^2=5, d.f.=3, P=0.2, Fig. 4 B). However, the number of conflicts also differed among months in each season (χ^2 ranged from 15 to 23, d.f. ranged from 3 to 4, P<0.005 in all cases; Fig. 4 B), and the month(s) with the highest number of conflicts in summer, fall, winter, and spring were, respectively, January and February, April, July-September, and October and November (contrasts, P<0.05 in all significant comparisons, Fig. 4 B).

The frequency of conflicts also differed between the days of the week in RS (χ^2=18, d.f.=6, P=0.005), because of a greater number of conflicts on Fridays than on Tuesdays (proportion contrast, P<0.05; Fig. 4 C). There were also differences in the frequency of conflicts in SC (χ^2=19, d.f.=6, P=0.005, Fig. 4 D). with a greater number of conflicts occurring on Mondays, Tuesdays, Wednesdays and Fridays than on Sundays (contrasts, P<0.05 all significant comparisons; Fig. 4 D).

3.5. Predictors of conflict lethality

The minimal adequate GLMM explained 83% of the variation in conflict lethality and included the predictors 'state,' 'type of conflict,' 'age-sex class,' 'day of the week,' and 'distance to the nearest large forest fragment' (R^2_c=0.83; Table 3). Conflict-related mortality was lower in SC than in RS (β=-1.2, z-value=-3, P<0.01) and for MT than for the other conflicts (β=-1.8, z-value=-3, P<0.01). Lethality was higher for adult females than for victims belonging to other age-sex classes (β=1.9, z-value=2, P<0.05) and for

Tuesday conflicts than for those occurring in the other days (β=-1.2, z-value=2, $P<0.05$; Table 3).

4. Discussion

We found that electrocution is the most frequent conflict affecting the physical integrity of brown howlers in the study peri-urban landscapes, agreeing with findings for wildlife worldwide that powerlines and the associated infrastructure represent major threats. Electrocution kills hundreds of primates (Lokschin et al., 2007; Katsis et al., 2018; Dittus, 2020; Jones-Román et al., 2021) and hundreds of thousands to millions of birds and other vertebrates each year (Bernardino et al., 2018; Biasotto and Kindel, 2018). Therefore, the implementation of management strategies such as an appropriate trimming of tree branches, insulation of powerlines, installation of wildlife crossings (canopy-to-canopy aerial bridges), creation of biological corridors, and an efficient protection of riparian edges are urgent not only to prevent the electrocution of arboreal wildlife, but to increase habitat connectivity and gene flow between animal populations (Table 4).

Attacks by stray or domestic dogs were the second major incident involving brown howlers in both states. This strongly supports the relevance of these animals as 'killers' of primates (e.g. *Alouatta guariba clamitans*: Buss, 2012; Bicca-Marques et al., 2020; *C. nigritus*: Oliveira et al., 2008) and many other terrestrial mammals (e.g. Buttler et al., 2004, Lacerda et al., 2009; Home et al., 2017; Gatti et al., 2018). In the study area, these attacks often occur when brown howlers descend to the ground to access cultivated fruits in subsistence orchards guarded by domestic dogs (Buss 2012; Chaves and Bicca-Marques, 2017; Corrêa et al., 2018) or, when they cross roads, gardens or pastures to access another Atlantic Forest remnant (Óscar M. Chaves and João Claudio Godoy, personal observation). In most cases, death or severe injuries (e.g. organ perforations, mutilations, multiple bites, and skin cuts: see database in Chaves et al., 2021) are the outcome of dog attacks. Critically injured individuals cannot be returned to their habitats. For instance, most (ca. 70%) brown howlers surviving dog attacks in RS and sent to recovery to the Rincão do Araticum Wildlife Rescue Center have never returned to their habitats because of infections in the injured organs (mainly lungs) or amputations in tail, feet, and hands (Silvia B. Ribeiro, personal communication). Therefore, population control of stray dogs is a necessary management strategy to reduce dog-wildlife conflicts in the study peri-urban matrices (Table 4).

Although less frequent, run overs and human mistreatments also deteriorate the health and compromise the survival of brown howlers in the study regions. These conflicts were expected given (i) the high fragmentation and urban encroachment of the Atlantic Forest remnants that howlers inhabit (Ribeiro et al., 2009), (ii) the frequent dispersal of individuals from their natal groups (Strier et al., 2001), (iii) the howlers' diet supplementation with wild and cultivated foods found in scattered food patches separated by roads and other potentially lethal landscape elements (Buss, 2012; Chaves and Bicca-Marques, 2017; Corrêa et al., 2018), (iv) the howlers' incapacity of high-speed travel on the ground, and (v) the inefficient Brazilian public policies to prevent/mitigate road kills (Gonçalves et al., 2018). Run over is a major cause of wildlife mortality in southern Brazil (Teixeira et al., 2020). It is estimated that ca. 1.3 million vertebrates (10% of which are large/medium birds, reptiles, primates, and terrestrial mammals) are killed every day along the extensive Brazilian networks of streets and roads (CBEE, 2019). Finally, the reported percentage of mistreatments is probably underestimated. This conflict is rarely denounced by local inhabitants probably because they are either afraid of retaliations or because they are poorly informed on how to fill out a complaint.

Whereas the larger body of adults compared with that of immature individuals increases their risk of touching cables with opposite charges simultaneously (Printes, 1999), thereby potentially explaining the highest frequency of electrocuted adult howlers, the greatest number of dog attacks and run overs on adults is compatible with their leading role in group travel both on the ground (Bicca-Marques and Calegaro-Marques, 1997) and on the canopy (Fernandéz et al., 2013), as reported for black-and-gold howler monkeys (*Alouatta caraya*).

Irrespective of conflict type, most howlers suffered lethal or mild-medium injuries (e.g. lung perforations, severe skin-burns, and body mutilations; see Fig. S2) and died soon after the incident or a few hours later. The overall fate of howlers involved in conflicts is even worse if we take into account that 93 to 98% of those individuals that survived after receiving veterinarian care were condemned to live in captivity. These findings highlight that howler monkeys are in great danger in urban and peri-urban areas of southern Brazil as have been suggested for the study areas (e.g. Printes, 1999; Buss, 2012; Corrêa et al., 2018; Bicca-Marques et al., 2020). Victims of mistreatments were the exception. Very few died or suffered severe injuries probably owing to their use as pets and the resulting provisioning of food and some veterinarian care by the “owners,” as observed in the study regions (Gerson Buss and Júlio César Souza Jr., personal communication).

Individual removal from wild populations via death or life in captivity compromise the long-term conservation of brown howlers in the Atlantic Forest with cascading consequences at the community level because of their important role as seed dispersers (Martins, 2006; Chaves et al., 2018). A brown howler can disperse ca. 52,000 seeds >2 mm per year (Chaves et al., 2018). Taking brown howler density data in RS into account (see Table S3), we estimate that the injured individuals that we have reported (*n*=248) represent ca. 10% of the total brown howler' population in this study region. Given the limitations of our data collection mentioned in the Methods and the fact that most conflicts that we reported involved adult females which could be pregnant, the estimate above is quite likely an underestimate. Another implication that needs to be considered is the economic cost associated with the rescue, veterinarian care, and maintenance of handicapped individuals in RS and SC and the maximum lifespan reported for captive howlers (i.e. 20 years). These costs can reach US$45,000/animal during a 15-year period (Table S4). Considering only the basic costs of maintenance in captivity, rescue centers spend considerable monthly amounts (e.g. US$ 177/month/animal in SC, Júlio César Souza Jr., personal communication) that are rarely reimbursed by local or state governments.

The lack of a clear pattern in the seasonal distribution of conflicts between the two states may have resulted from regional differences in the use of the landscapes. Although incidents occurred throughout the year in RS, alike that reported by Buss (2012), there were higher frequencies of records in the summer and fall compared with the winter. In SC, however, the distribution of conflicts was similar between seasons. Whether this difference simply reflects the temporal characteristics of the sampling efforts in RS and SC (see Methods) or legitimate differences resulting from higher numbers of people living or visiting the RS study region or driving through it during their summer vacation remains to be investigated. In SC, the sampling conducted by Projeto Bugio-FURB (https://www.furb.br/web/5579/ projeto-bugio/apresentacao) was more uniformly distributed throughout the year. The leading role of Projeto Bugio, a research institute with a consolidated history of rescuing, caring, and rehabilitating brown howlers in SC, may also explain the marked influence of state on conflict-related mortality (Table 4).

5. Conclusions

To the best of our knowledge, this is the first study collating long-term data on the main threats faced by wild Neotropical primates living in peri-urban landscapes. We confirmed the aforementioned negative impact of urbanization on wildlife health and survival described in short-term studies of primates and other vertebrates. We found that the destruction of the Atlantic Forest and the encroachment of its remnants by urban centers represent serious (and often ignored) conservation challenges for the long-term survival of arboreal primates (and probably many other vertebrates). The severity of this scenario is further highlighted by the fact that despite flexibly adjusting their behavior to diverse anthropogenic landscapes (including peri-urban regions), alike other vertebrates (Sol et al., 2010; Schell et al., 2020), the long-term persistence of howler monkey (*Alouatta* spp.) in fragmented peri-urban landscapes is at high risk (Bicca-Marques et al., 2020). As we have shown conservatively, hostile elements of the urban matrix, such as power lines, roads, domestic dogs, and wildlife traffickers, impose a much higher death rate to peri-urban populations than that seen in habitats more isolated from people. Therefore, designing and implementing appropriate strategies to prevent or mitigate human-wildlife conflicts are crucial to save urban- and peri-urban-tolerant species from extirpation.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/

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Figure legends

Fig. 1. Location of conflicts involving brown howler monkeys in Santa Catarina and Rio Grande do Sul states, southern Brazil. Color markers represent the type of incident: white (run over), yellow (unknown), rose (electrocution), cyan (dog-attack), blue (natural causes), concentric circle (mistreatment). The black star in the marker indicates that the monkey suffered mild or severe injuries, while its absence indicates that it died soon after the incident or during the transport to the veterinary hospital. Observe the difference in scale between the two state regions.

Fig. 2. Main threats faced by brown howlers (*Alouatta guariba clamitans*) in urban and peri-urban areas in Rio Grande do Sul state, southern Brazil. Adult female using a power line to cross a road (A), individuals electrocuted and/or mutilated on power lines (B-D), monkeys crossing roads to access food patches (E,F), adult and sub-adult males run over (G, H), adult male on cultivated tree in a subsistence orchard guarded by dogs (I), juvenile individual walking on the ground near a domestic dog (J), victim of mistreatment in a peri-urban area of RS (K). Photos by Ó. M. Chaves (A-C, H), J. C. Godoy (D-F), G. Buss (G, K) and J. P. Back (I-J).

Fig. 3. Comparison of the proportion of conflicts involving brown howlers according to the type of incident (a, b) and the age-sex class (c, d) in the States of Rio Grande do Sul (top panels) and Santa Catarina (bottom panels). Different Lucida handwriting capital letters on the bars indicate differences among incident types or age-sex classes, and lowercase letters inside the bars indicate differences among injury levels or incident types (proportion contrasts, $P < 0.05$). When no significant differences were detected within each incident type

or age-sex class (proportion contrasts, $P > 0.05$), no lowercase is show. Type of incident: EL=electrocution, DA= dog attack, RO= run over, and MT= human mistreatment (further details in Methods). Numbers in parentheses at the bottom of bars represent the number of conflicts per type of conflict or age-sex class. Age categories: A= adult, S= subadult, J= juvenile, and I=infant. Total number of conflicts considered in each graph: 222 (a), 151 (b), 262 (c), and 225 (d).

Fig. 4. Temporal patterns in the proportion of conflicts according to season (a, b) and day of the week (c, d) in Rio Grande do Sul (top panels) and Santa Catarina (bottom panels) states. Different Lucida handwriting capital letters on the bars indicate differences among seasons or day of the week (proportion contrasts, $P<0.05$). When the proportion of conflicts was similar (proportion contrasts, $P>0.05$) among seasons or days, no capital letter is shown. Lowercase letters on the bars in (a) and (b) indicate differences among months within each season (contrasts, $P<0.05$). When the proportion of conflicts was similar between months (contrasts, $P>0.05$), no lowercase is shown. Days: Monday (Mon), Tuesday (Tue), Wednesday (Wed), Thursday (Thu), Friday (Fri), Saturday (Sat), and Sunday (Sun). Numbers in parentheses at the bottom of bars represent the number of conflicts per season or day recorded until April 2021. Total number of conflicts considered in each graph: 214 (a), 199 (b), 261 (c), and 260 (d).

Table 1. Demographic characteristics of the main municipalities/cities monitoring in Rio Grande do Sul and Santa Catarina, Brazil

Variable[a]	Rio Grande do Sul			Santa Catarina[b]				
	Viamão	Lami	RS	BL	IN	PO	JS	SC
Area (km^2)	1,496	28.2	281,707	518.6	430.8	214.3	530.1	95,731
Population size in 2019	252,872	4,642	11,377,239	357,199	69,425	33,447	177,697	7,164,788
Urban population	224,943	—	9,100,291	294,773	52,927	23,823	132,800	5,247,913
Rural population	14,441	—	1,593.64	14,238	1,927	3,936	10,323	1,000,523
% population in urban areas	94.0	—	99.8	95.4	96.5	85.8	92.8	84.0
Population density (ind./km^2)	160	165	39.8	596.1	127.3	129.3	270.3	65.3
Population growth (%)	5.3	—	20.0	13.5	21.0	17.0	19.5	12.8
#vehicles in 2018	95,734	—	5,365,382	197,586	33,894	18,100	84,776	3,672,593
#Urban residences	70,514	4,030	3,084,215	96,866	16,753	7,423	42,070	—
#Rural residences	4,883	—	515,589	4,196	614	1,130	3,036	—

[a]Information sources were: Instituto Brasileiro de Geografía e Estatística (population census 2010; IBGE, 2020) and Departamento Nacional de Trânsito (number of vehicles 2018; Denatran, 2020). Population growth was based on the last 10 years (i.e. the differences between the population estimate of 2019 and the population census of 2010. Vehicles included: cars, pickups, trucks, buses, and microbuses.

[b]Municipality abbreviations: BL=Blumenau, IN= Indaial, PO=Pomerode, and JS=Jaraguá do Sul.

— Information not available.

Table 2. Potential predictors of post-incident lethality in brown howlers in the peri-urban matrices of Rio Grande do Sul and Santa Catarina states, southern Brazil

Predictor[a]	Description	Effect[b]
	Physical traits of urban matrix	
1) Matrix element	Element of the urban matrix where the incident occurred, including roads, gardens, cities, fragment edges, forest remnants, etc.	(+)
2) Conflict type	Main incident involving howlers: electrocution (EL), dog-attack (DA), runover (RO), and mistreatment (MT).	N.A.
3) # houses	Number of houses or clearly identifiable roofs around a 500-m radius from the incident location.	(+)
4) # elements	Total number of elements constituting the anthropogenic matrix (e.g. roads, airports, power lines, houses, buildings, gardens, and pastures) around a radius of 500 m from the incident location.	(+)
5) Type of road	Type of road nearest to the incident location: primary large road (>15 m wide) with high vehicle traffic (P), and secondary small roads (<10 wide) with low vehicle traffic (S).	P>S
6) Road material	If the road was paved or unpaved	paved>unpaved
7) DNR	Distance from the incident location to the nearest primary or secondary road (m).	(-)
8) DNS	Distance from the incident location to the nearest small fragment <10 ha.	(+)
9) DNL	Distance from the incident location to the nearest large fragment > 80 ha.	(+)
10) DNH	Distance from the incident location to the nearest house.	(-)
	Other factors	
11) Age-sex	Age-sex class of the animal, including adults (A), subadults (S), and juveniles (J) of both sexes.	N.A.
12) State	Study state: Santa Catarina (SC) and Rio Grande do Sul (RS)	N.A.
13) Season	Season of the year in which each incident occurred: summer (Su), fall (F), winter (W), and spring (Sp).	>S
14) Day	Day of the week in which each incident occurred: Monday (Mon), Tuesday (Tue), Wednesday (Wed), Thursday (Thu), Friday (Fri), Saturday (Sat), and Sunday (Sun).	>Sat/Sun
15) Incident*element	Interaction between the type of incident and the matrix element	N.A.
16) Incident*age-sex	Interaction between the type of incident and the age-sex category	N.A.

[a]Ten physical traits of the peri-urban matrices estimated in this study enhanced in bold.
[b] Expected effect according to available evidence: positive (+), negative (-), higher (>), and non-assessed (N.A.).

Table 3. Minimal adequate GLMM predicting the lethality of conflicts involving brown howlers in the urban anthropogenic matrix of southern Brazil

Predictors	Parameters[a]				
	β	SE	z-value	AICc	R^2_c
Model: **state+incident+age-sex+day+dlf**			6.2**	320	0.83
State					
Santa Catarina	-1.19	0.36	-3.3**		
Type of incident (incident)					
Mistreatment	-1.76	0.57	-3.1**		
Run over	-0.61	0.44	-1.4		
Electrocution	-0.30	0.39	-0.8		
Age-sex class (age-sex)					
Adult female	1.92	0.93	2.1*		
Subadult male	1.66	1.10	1.6		
Adult male	1.32	0.87	1.5		
Juvenile female	1.44	1.06	1.4		
Day of the week (day)					
Tuesday	1.19	0.59	2.1*		
Thursday	0.72	0.57	1.3		
Distance to the nearest large forest (dlf)	-0.00	0.00	-0.9		

[a]Parameters shown: partial regression coefficients (β_i), standard errors that incorporate model uncertainty (SE), Akaike's Information Criterion for small samples (AICc), and pseudo-R^2 (R^2_c) indicating the percentage of the variance explained by the fixed and random factors in the minimal adequate model. Significance level: * P<0.05, ** P<0.01.

Table 4. Fifteen potential management strategies to minimize the number of conflicts involving brown howler monkeys in the anthropogenic urban matrix of south Brazil

Type of incident	Management strategy	Description	Ref.[b]
EL, DA, RO, MT	1) Environmental education program	Activities related to the sensitization of local inhabitants on the importance of preventing conflicts with wildlife.	1-3
	2) Creation of a specialized network	Interdisciplinary group devoted to monitor, rescue, care, rehabilitate, and release recovered animals back to nature. This group needs to operate 7 days/week.	1-3
EL, DA, RO	3) Establishment of urban protected areas	Protected areas inside or adjacent to urban or peri-urban centers. These areas not only contribute to protect wildlife but also promote human health and well-being.	1,4
	4) Establishment of biological corridors	Biological corridors are crucial to increase the connectivity between habitat patches used by brown howlers and other terrestrial vertebrates.	1, 5
	5) Efficient protection of riparian edges	The density of howler monkeys in urban/disturbed areas often increases along the riparian edges. These forests contain food sources, shared microhabitats, and water for the wildlife, while they increase the connectivity between isolated habitat patches.	1, 4
	6) Installation of wildlife crossings	The construction and installation of aerial rope bridges (elaborated with PVC tubes and rope) in hotspots of conflicts.	1, 5-7

	7) Creation and/or protection of natural canopy bridges	Natural bridges used frequently by primates and that are composed by the canopies of two or more large trees in the opposite sides of a road.	1, 4
	8) Creation of public policies to assist injured animals	Elaboration of municipal laws that enforce energy companies, road departments, and urbanization secretaries to cover the costs related to monkey rescuing, veterinarian assistance, and captive maintenance.	1-3
EL	9) Creation of buffer zones between the forest dossel and power lines	Buffer zones can be established over the trimming of tree branches that allow access to power lines, particularly in medium-voltage power lines.	1, 3, 5, 7
	10) Insulating of power lines	Insulation of low-tension power lines (and high-tension power lines, whenever possible) and installation of physical barriers in the cables to difficult the access of the animal to the cables.	1-3, 5, 7
DA	11) Sterilization of domestic and stray dogs	Sterilization campaigns of domestic and stray dogs inhabiting the urban matrix.	8, 9
	12) Prevention that dogs roam free	Installation of fences, dog-kennels, and other barriers in gardens to avoid that domestic dogs roam free and interact with primates and other wild vertebrates.	8, 9
	13) Relocation and euthanasia of stray dogs	When possible, part of these animals should be captured, sterilized, and relocated to public and private dog-kennels. Lethal control of stray dogs when their population is high and the frequency of dog-wildlife conflicts justify it.	8, 9

RO	14) Installation of speed reducers	Installation of speed bumps and posting of signs to persuade vehicle drivers to reduce the speed near locations with frequent crossing of howler monkeys and other terrestrial vertebrates.	10
MT	15) Efficient surveillance and penalty systems	The surveillance and quick penalty on environmental crimes associated with illegal captivity, traffic, and mistreatment.	3

[a] Electrocution (EL), dog-attack (DA), run over (RO), and mistreatment (MT).

[b]References: 1- Jones-Román et al (2021), 2-Gordo et al. (2013), 3- Buss (2012), 4-Trzyna (2014), 5-Lokschin et al. (2007), 6-Printes (1999), 7-Rodríguez et al. (2020), 8- Home et al. (2017), 9-Villatoro et al. (2019), 10-Texeira et al. (2020)

Figure 1


60°0'0"W
N
BOLIVIA
BRAZIL
PARAGUAY
ARGENTINA
30°0'0"S
30°0'0"S
URUGUAY
Atlantic
Ocean
60°0'0"W
State of Santa Catarina
30 km
State of Rio Grande do Sul
2.5 km

Figure 2

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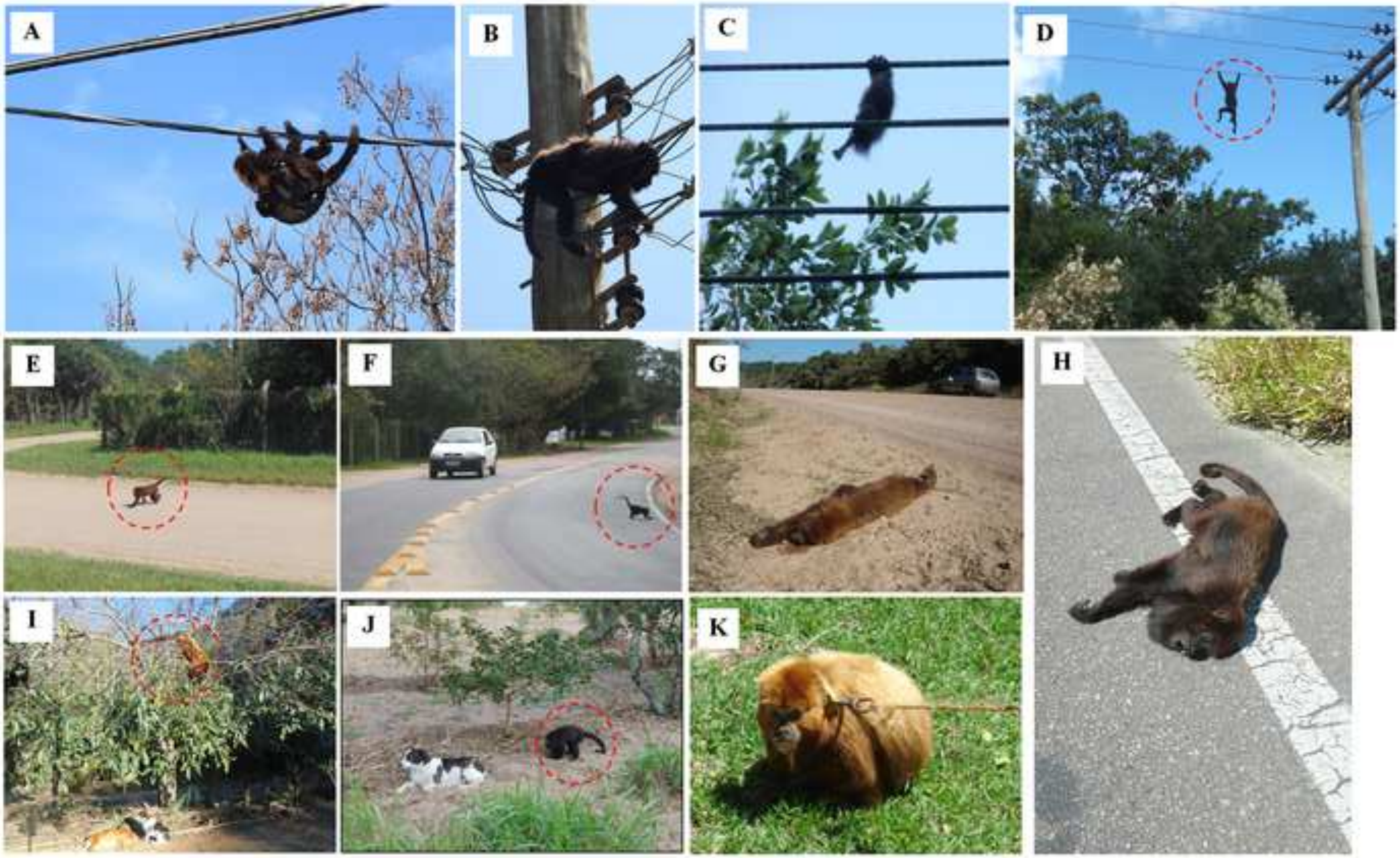

Figure 3

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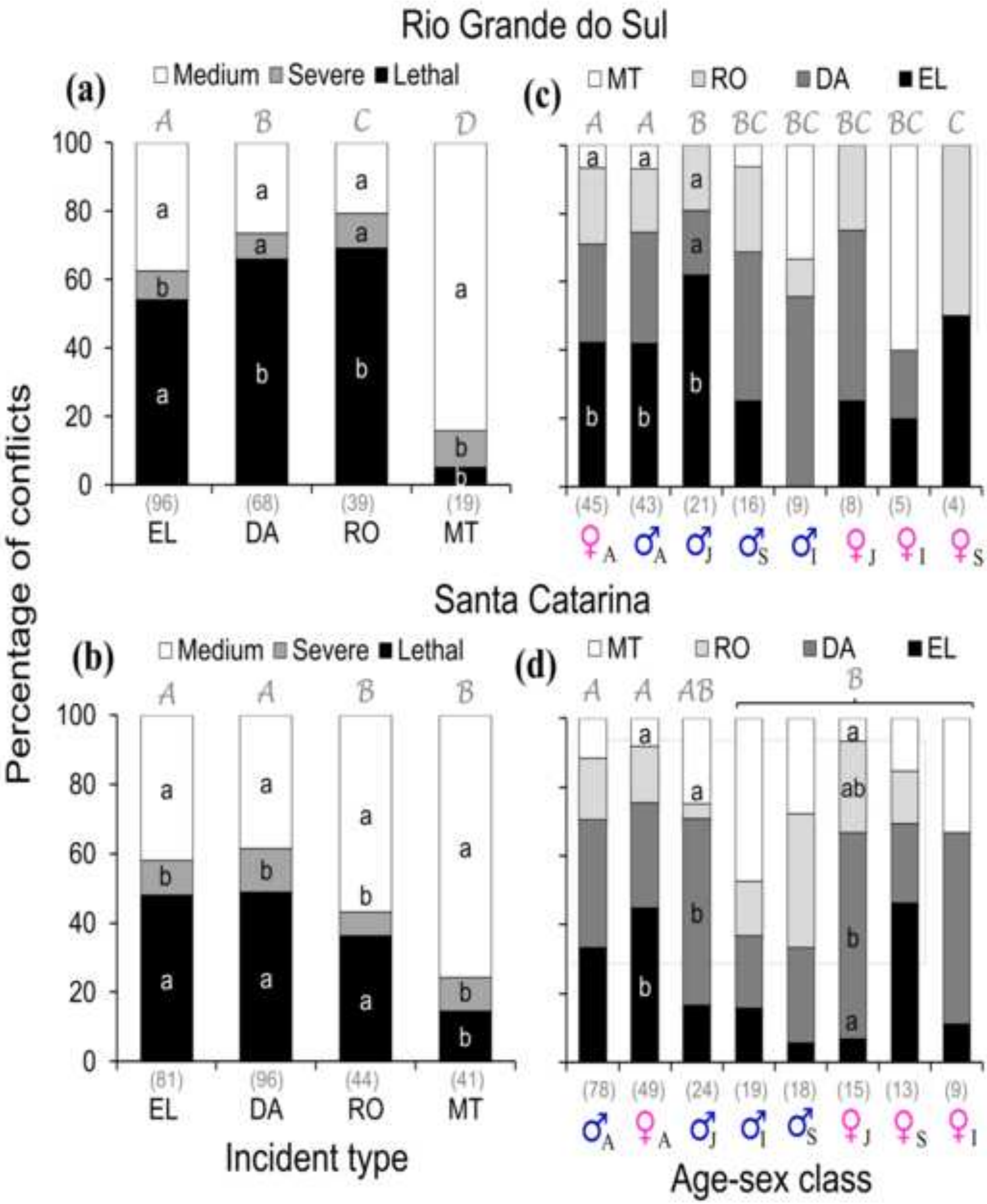
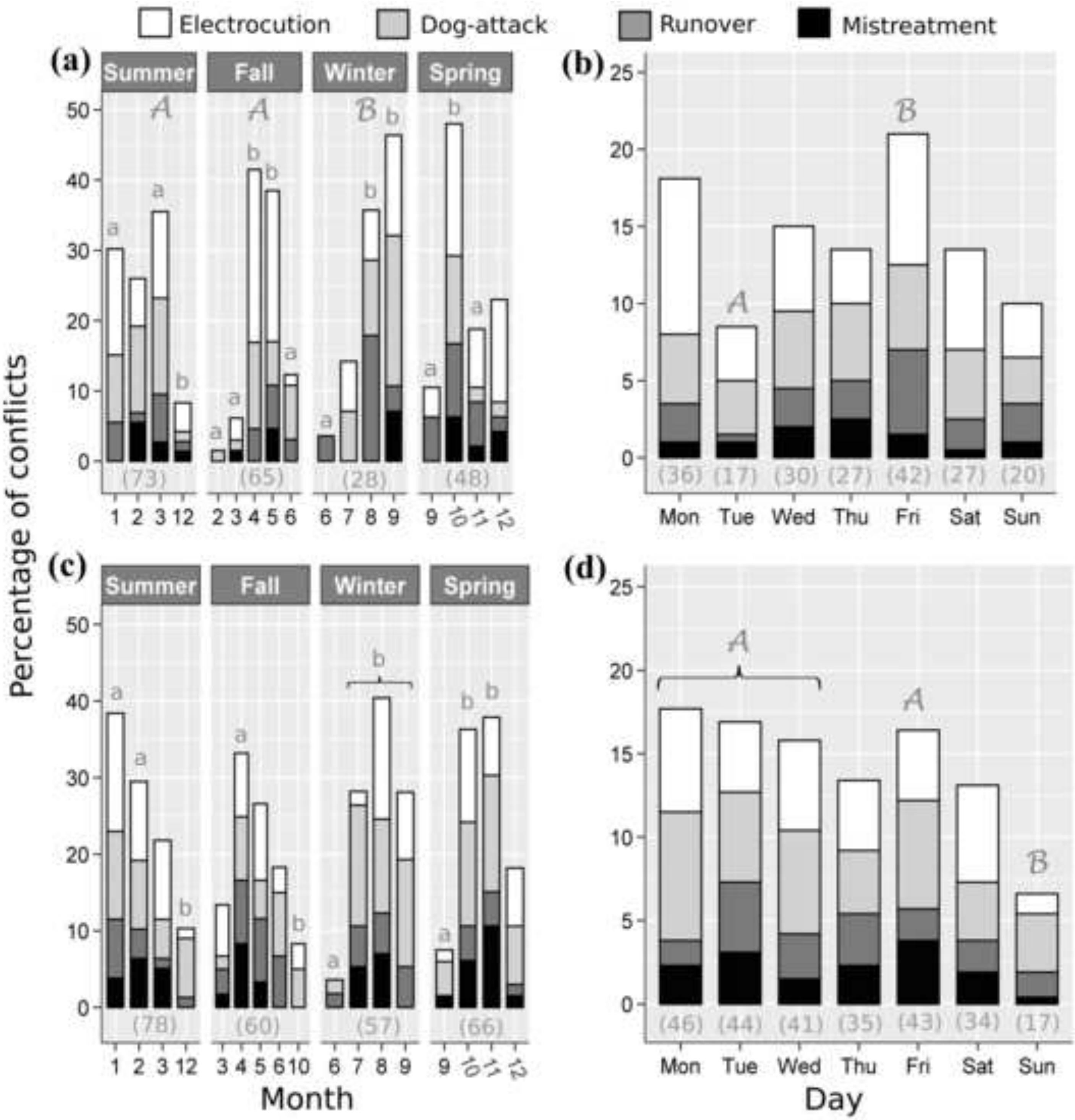


Figure 4


Electrocution
Dog-attack
Runover
Mistreatment
(a)
Summer
Fall
Winter
Spring
(73)
(65)
(28)
(48)
(b)
Mon
Tue
Wed
Thu
Fri
Sat
Sun
(36) (17) (30) (27) (42) (27) (20)
Percentage of conflicts
(c)
(78)
(60)
(57)
(66)
(d)
(46) (44) (41) (35) (43) (34) (17)
Month
Day

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Supplementary Material
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Credit authorship contribution statement

ÓMC: Investigation, Conceptualization, Data collection, Data curation, Formal analysis, Writing original draft, Manuscript revision. **JCSJ:** Data collection, Data curation, Manuscript revision. **GB:** Data collection, Data curation, Manuscript revision, **ZMBH:** Data curation, Funding acquisition, Manuscript revision. **MMAJ:** Data collection, Data curation, Methodology, Manuscript revision. **ELSA:** Data curation, Methodology, Manuscript revision. **JCG:** Data collection, Methodology. **ARP:** Data collection.**TM:** Data collection, Data curation. **JCBM:** Funding acquisition, Supervision, Methodology, Manuscript revision, English revision.

Declaration of conflict of interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.