

Pattern and Distribution of Fatal Injuries in Wildlife Vehicle Collisions (2010-2019)

Andreia Garcês^{1*}, Justina Prada^{2,3}, Ana Silva², Filipe Silva^{2,3}, Celso Santos², Isabel Pires^{2,3}

¹Inno – Laboratory Veterinary, Microbiology Department, Rua Cândido de Sousa 15, 4710-503 Braga, Portugal

²Department of Veterinary Science of the University of Trás-os-Montes and Alto Douro Quinta dos Prados, 5001-801, Vila Real,

³CECAV - University of Trás-os-Montes and Alto Douro, Quinta dos Prados, 5001-80, Vila Real, Portugal.

*andreiagarcês@inno.pt

ABSTRACT

One of the major causes of death of wild animals worldwide is vehicle collisions. In Portugal the number of animals killed by vehicle collisions has grown in the past years. In order to characterize pathologic lesions sustained by wild animals injured in wildlife vehicle collisions, the necropsy reports of 170 animals (mammals and birds) performed between January 2010 and June 2019 were evaluated. The majority of the animals were wild mammals (73.5%), being Red Fox (*Vulpes vulpes*) (26.5%) the most common species. There were more collisions adult animals (88.2%) than young animals. There was no difference between sexes. The most commonly affected body area was abdomen and pelvis injuries in mammals (80.8%) and coelomic cavity in birds (80.0%). Musculoskeletal lesions were the most commonly identified on both Classes. The evaluation of post-mortem lesions in animals who suffered from traffic collisions may provide important information to enhance knowledge in this area of study. Integration of this data with animal behaviour may, in the future, allow the establishment of measures to prevent wild animal's traffic collisions.

Keywords

collision; Portugal; roadkill; road ecology, vehicles; wildlife; trauma

Introduction

The increase in human population and urban areas that has taken place in recent decades has had an impact at various levels, for instance economic, politic, geographic and ecologic. The last one can be compromised, either directly, by increased pollution, habitat destruction or climate change, or indirectly through genetic and population losses resulting from the construction of roads and other transport routes (Jackson & Fahrig, 2011).

The emergence and globalization of highways invariably implies the invasion of wild habitats, forcing wild animals to relocate or adapt their behaviors to the new conditions. This often leads to road traffic accidents (Knutson, 1987). Death due to the vehicle collisions is one of the major cause of death in wild animals as described in several works around the world (Edwards et al., 2000; Molina-López et al., 2011; Rodríguez et al., 2010; Taylor et al., 2002; Visintin et al., 2017).

Roads are one of the main anthropogenic factors responsible by habitat fragmentation and destruction (Fahrig, 2003; Hostetler et al., 2009; Lindenmayer et al., 2000; Riley et al., 2003). They can lead to alteration of social behaviors, reduce reproductive rates and/or genetic diversity, increase mortality and in later stages lead to extinction of population (Fahrig, 2003; Hostetler et al., 2009; Lindenmayer et al., 2000; Riley et al., 2003). Other negative indirect effects of traffic are noise and artificial light, which are majors contributors for the reduced density of birds near roads (Silva et al., 2012).

The impact of vehicles and roads on wildlife has been a matter of concern for several governmental and environmental entities, which has already led to measures such as the construction of animal passages or the maintenance of non-specific passages, such as water or agricultural channels, allowing a safe passage for these animals (Marcantonio et al., 2013). Several organizations have been monitoring the impact of roads on wildlife, such as Canada (Bishop & Brogan, 2013), Australia (Brendan et al., 2010) United States (Editorial et al., 2011; Loss et al., 2014), Brazil (Ascensão & Mira, 2006; Bager & Da Rosa, 2011; Corrêa et al., 2017), Poland (Elzanowski & Ciesio, 2009; Morelli et al., 2014) or Spain (Canal et al., 2018; Hernandez, 1988; Sillero, 2008).

In Portugal, according to data provided by Infrastructures of Portugal (IP), the number of wildlife vehicle collisions on Portuguese roads increased gradually between 2011 and 2015 (Garcia, 2012, 2013, 2014, 2015, 2016). The increase in the number of road casualties coincides with the increase in the general awareness of this phenomenon. This led to a rise in the reporting of such incidents. As a result in 2015 an IP partnership with the LIFE LINES (project of the University of Évora), in a daily and methodical performed a search for struck by vehicles cases in diverse municipalities (Universidade de Évora, 2018a).

An analysis of the data provided by the LIFE LINES project reveals that amphibians and wild birds represent an important slice of the identified species of animals involved in vehicle collision incidents in Portugal (Universidade de Évora, 2018b). The Order Anura (Classe Ammphibia) has the highest number of occurrences, followed by the Order Passerines (Class Aves). The high number of amphibian deaths reported may raise questions about the future of amphibian populations, which have been declining in recent years, with many species being classified with conservation status of vulnerable or critically in danger (Hels & Buchwald, 2001)

The purpose of this work was to identify and systematize the injuries resulting from vehicle collisions involving wild animals in the North region of Portugal.

Methods

The necropsy files of the Laboratory of Histology and Pathological Anatomy of the University of Trás-os-Montes and Alto Douro (Vila Real, Portugal) between January 2010 and July 2019 were consulted between January 2010 and July 2019. Only animals with both a history and macroscopic lesions compatible with vehicle collision were included. All the necropsies were performed by veterinary pathologists (I.P. and J.P.). The cadavers of the animals were delivered by the road concessionaires or the police authorities (when human or material damage are involved)

From all cases, data were collected regarding the date of necropsy, species, Order, Class, sex and age. Animal were categorized as juvenile and adult based on their external characteristics. (Bencatel et al., 2017; Cabral et al., 2005; Matias et al., 2007; Meirinho et al., 2014). The necropsy of each animals was performed according to standard techniques described for each of the classes, following the standards of security and hygiene (Garcês & Pires, 2017).

The systematization of the observed lesions was performed by anatomical region and type of lesion. Lesions were evaluated considering all their features in order to separate that antemortem or postmortem injuries. Only injuries that were associated with evident ante-mortem trauma were considered in this study. Thus, categorization was made into four anatomical regions: head and neck, chest, abdomen and pelvis, and limbs with the exception of birds, were three anatomical regions were considered: head and neck, celomic cavity and

limbs. Injuries were further classified according to the type of lesion as musculoskeletal, organ, or cranioencephalic. Musculoskeletal injuries encompassed all alterations with fracture or dislocation of long bones, ribs, pelvis, and spine. Organs lesions included all those involving only internal organs (Ressel et al., 2016; Rogers & Stern, 2018). The presence of blood in body cavities was distinguished between thoracic (hemothorax), abdominal (hemoperitoneum) or coelomic cavity in birds. Cranioencephalic injuries included head injuries with hemorrhages and skull fractures (Akinrinmade, 2002; Kolata, 1980).

All data were entered into a database using Excel and the statistical analysis was performed using IBM SPSS Statistics (version 24, advanced Models TM 21.0, SPSS Inc. 233 South Wacker Drive, 11th Floor Chicago, IL 60606-6412). In order to study the differences between the observed and expected frequencies of categories of a field, one-sample nonparametric tests were used (Binomial test or Chi-Square test, depending of the number of categories of the categorical field).

Results

Characterization of the sample

Of the 170 animals included in the study, 45 belonged to the class Aves (26.5%) and 125 to the Mammalia class (73.5%). Table 1 describes the species examined in this study.

Our study included 81 females and 89 males, with the majority being adult animals (n=150). The differences observed in animal sex was not significant (p=0.706), but for animal age, they were statistically relevant (p<0,001).

Table 1: Species, sex and age range of 170 animals included in this study.

Class	Specie	N	Sex		Age	
			Male	Female	Adult	Juvenile
Aves	Northern Goshawk (<i>Accipiter gentilis</i>)	1	0	1	1	0
	Short-Eared Owl (<i>Asio flammeus</i>)	1	0	1	1	0
	Little Owl (<i>Athene noctua</i>)	5	2	3	4	1
	Eurasian Eagle-owl (<i>Bubo bubo</i>)	1	1	0	1	0
	Common Buzzard (<i>Buteo buteo</i>)	9	6	3	8	1
	Western Marsh Harrier (<i>Circus aeruginosus</i>)	1	0	1	1	0
	Great Spotted Woodpecker (<i>Dendrocopos major</i>)	1	0	1	1	0
	Common Kestrel (<i>Falco tinnunculus</i>)	1	0	1	1	0
	Common Snipe (<i>Gallinago gallinago</i>)	1	0	1	1	0
	Eurasian Jay (<i>Garrulus glandarius</i>)	2	2	0	2	0
	European Honey Buzzard (<i>Pernis apivorus</i>)	1	1	0	1	0

	Tawny Owl (<i>Strix aluco</i>)	11	5	6	11	0
	Common Blackbird (<i>Turdus merula</i>)	5	3	2	4	1
	Common Barn Owl (<i>Tyto alba</i>)	4	2	2	4	0
	European Nightjar (<i>Caprimulgus europaeus</i>)	1	1	0	1	0
	TOTAL	45	23	22	42	3
Mammalia	European Roe Deer (<i>Capreolus capreolus</i>)	16	7	9	11	5
	European Hedgehog (<i>Erinaceus europaeus</i>)	2	0	2	2	0
	Common Genet (<i>Genetta genetta</i>)	14	12	2	11	2
	Egyptian Mongoose (<i>Herpestes ichneumon</i>)	1	1	0	1	0
	Eurasian Otter (<i>Lutra lutra</i>)	6	3	3	5	1
	Beech Marten (<i>Martes foina</i>)	7	6	1	7	0
	European Pine Marten (<i>Martes martes</i>)	8	4	4	8	0
	European Badger (<i>Meles meles</i>)	10	5	5	9	1
	European Polecat (<i>Mustela putorius</i>)	1	1	0	1	0
	Eurasian Red Squirrel (<i>Sciurus vulgaris</i>)	4	2	2	4	0
	Wild Boar (<i>Sus scrofa</i>)	11	5	6	11	0
	Red Fox (<i>Vulpes Vulpes</i>)	45	20	25	37	8
	TOTAL	125	66	59	108	17

In the ten years included in this study (between January 2010 and July 2019), the highest number of cases was in 2016 (n=40, 23.6%), as shown in Figure 1A. Regarding the months of study, most fatalities submitted were in March (n = 41, 24.1%), followed by October, according to the data presented in Figure 1B.

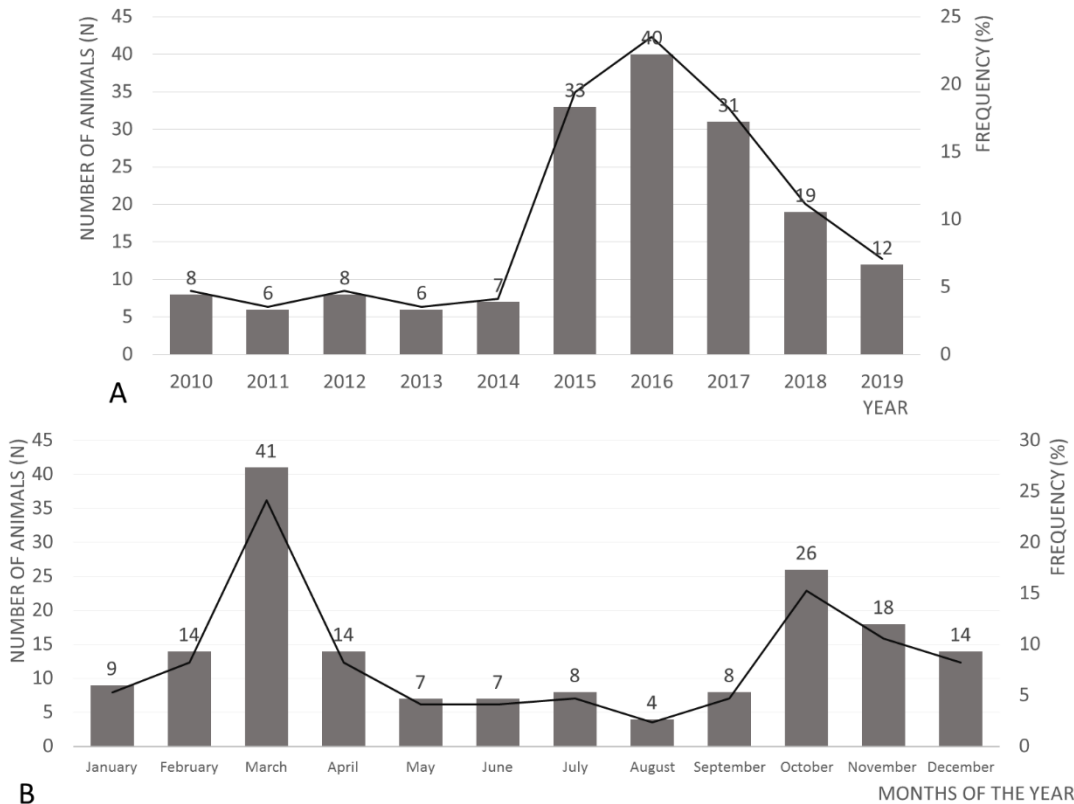


Figure 1- Distribution of vehicle collision cases by year of study (A) and months of the year (B) (n; %).

Distribution of lesions by anatomical region

The lesions in body cavities (thorax, abdomen and pelvis or coelomic cavity) are significantly higher than the lesions in the other anatomic regions considered ($p < 0,001$). Of all animals, 85 had thorax or 101 abdomen and pelvic injuries (mammals) and 36 had coelomic cavity injuries (birds), 70 had head and neck injuries (41.2%) and 52 had limb injuries (30.6%). Figure 2 records the prevalence of lesions in these regions on the necropsied animals.

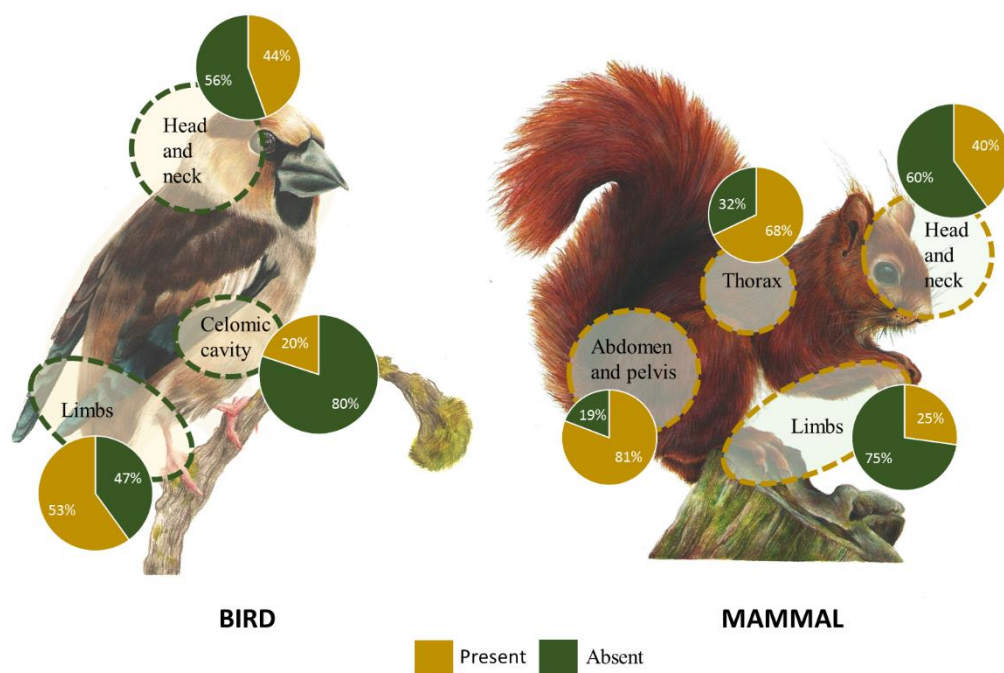


Figure 2- Distribution of lesions by anatomical region in mammals and wild birds included in this study (illustrations ©Andreia Garcês).

As seen in Table 2, most animals (n=126, 74.1%) had lesions in more than one anatomical area, while 44 animals (25.9%) had only one area affected (p<0,001).

Table 2: Number of anatomical areas affected in necropsied animals (n%).

	Number of anatomical areas affected	
	One	Multiple
<i>Class Aves</i>	20 (44.4)	25 (55.6)
<i>Class Mammalia</i>	24 (19.2)	103 (80.8)
<i>Total</i>	44 (25.9)	126 (74.1)

Lesions description

Most animals had musculoskeletal lesions (n=120, 70.6%), followed by organ lesions (n=112, 65.9%) and cranioencephalic lesions (n=59, 34.1%), (p<0.001). Table 3 describes the type of lesions by Class Aves and Mammalia.

Table 3: Distribution of lesions by type (musculoskeletal, organ and cranioencephalic) (n%).

	Type of lesion					
	Musculoskeletal		Organ		Cranioencephalic	
	Present	Absent	Present	Absent	Present	Absent
<i>Class Aves</i>	32 (71.1)	13(28.9)	17(37.8)	28(62.2)	17 (37.8)	28 (62.2)
<i>Class Mammalia</i>	88 (70.4)	37(29.6)	95(76.0)	30(24.0)	42(33.6)	83(66.4)

Total	120(70.6)	50(29.4)	112(65.9)	58(45.2)	59(34.1)	111(65.3)
-------	-----------	----------	-----------	----------	----------	-----------

Skeletal injuries included muscular lesions and bones fractures, that on the majority of the cases was associated to muscle hemorrhages. Of the 117 animals with bone fractures, 59 had one single bone affected while 58 presented multiple fractured bones. Fractures occurred on the jaw (n=15), vertebrae with fracture of spine with complete spinal cord lesion (n=30), pelvic bone (n=19), fore limb (n=22), hind limb (n=23), ribs (n=53), keel (n=4) and multiple fracture (n=6). Eighty-seven animals presented with cutaneous lesions associated with the fracture point (e.g., lacerations, avulsions), 25 in the eyes (e.g. eye prolapse, enucleation, ulceration, hemorrhage) and 15 in the ears (e.g. hemorrhage).

Cranioencephalic lesions included cranial skull fractures and intracranial hemorrhage. Twenty-six animals had skull fractures and 29 presented intracranial hemorrhage without bone fracture.

Internal organs affected by trauma were liver (n=54), lungs (n=29), spleen (n=28), kidney (n=10) and heart (n=4), in both wild birds and mammals, with rupture, concussion, contusion and hemorrhage. The presence of blood in body cavities, as a result of organ damage was also recorded.

Class Mammalia

Regarding mammals, 59 females and 66 males (p=0.592) were included, with 108 adults and 17 juveniles (p<0.001).

Considering the distribution of lesions by anatomical region, lesions in thorax and abdominal cavity were significantly higher (p<0.001); 85 of the necropsied mammals had thorax injuries (68%), 101 abdomen and pelvis injuries (80.8%), 50 had head and neck injuries (40%), and 31 had their limbs affected (24.8%).

One hundred and one animals had lesions in multiple body regions (80.8%) and 24 animals had only one region affected (19.2%), (p<0.001).

Regarding lesion type, the organs lesions were more frequent ((p<0.001) being observed in liver (n=43), lungs (n=28), spleen (n=28), kidney (n=4) and heart (n=4). Considering musculoskeletal lesions, 85 animals showed bone fractures, being 46 in one single bone and 39 in multiple bones. Three animals present muscular lesions without bone fracture associated. Bone fractures occurred in the ribs (n=45), vertebrae (n=21) pelvis (n=16), limbs (14 in the fore limbs and 16 in the hind limbs), and jaw (n=15). Cutaneous lesions were observed in 63 animals, ocular lesions in 19 animals and ear lesions in 11 mammals. Cranioencephalic lesions were the less frequent (n=42) and included cranial skull fractures and intracranial hemorrhage. Twenty animals had skull fractures and 22 presented intracranial hemorrhage without bone fracture.

Concerning cavities lesions, 62 animals had hemothorax and 73 had hemoperitoneum. Lesions in mammals also included pneumothorax (n=3), diaphragmatic hernia (n=8) and inguinal hernia (n=2). Organ evisceration was observed in 8 animals, with blunt exposition of abdominal organs. Table 4 summarizes the lesions presented in mammals.

Table 4: Lesion presented in the cavities of wild mammals.

<i>Species</i>	Hemothorax	Hemoperitoneum	Pneumothorax	Diaphragmatic hernia	Inguinal hernia	Evisceration
European Roe Deer (<i>Capreolus capreolus</i>)	8	4	2	1	0	1
European Hedgehog (<i>Erinaceus europaeus</i>)	1	1	0	0	0	0
Common Genet (<i>Genetta genetta</i>)	9	11	0	0	0	1
Egyptian Mongoose (<i>Herpestes ichneumon</i>)	1	0	0	0	0	0
Eurasian Otter (<i>Lutra lutra</i>)	1	3	0	2	0	0
Beech Marten (<i>Martes foina</i>)	3	3	0	0	0	0
European Pine Marten (<i>Martes martes</i>)	5	5	0	1	0	1
European Badger (<i>Meles meles</i>)	0	1	0	0	0	0
European Polecat (<i>Mustela putorius</i>)	6	6	0	0	0	0
Eurasian Red Squirrel (<i>Sciurus vulgaris</i>)	2	2	0	0	0	0
Wild Boar (<i>Sus scrofa</i>)	7	9	0	1	0	1
Red Fox (<i>Vulpes</i>)	18	28	1	3	2	2

Vulpes)

Class Aves

The necropsied birds were similarly distributed between the two sexes (n=22 female and 23 male) (p=1), most frequently adults (n=42; p<0.001).

Regarding the distribution of lesions by anatomical region, there is a significant difference (p=0,044), being the coelomic cavity the most affected region (n=36; 80.0%). Twenty animals presented head and neck injuries (44.4%), and 21 injuries in the limbs (46.7%).

Multiple injuries on different anatomical regions were significantly higher (p<0.001). Twenty-five birds had multiple body regions affected, corresponding to 55.6% of the 45 autopsied birds, and 20 had involvement of only one body region (44.4%).

Regarding lesion type, the musculoskeletal lesions were more frequent (p=0.033). Musculoskeletal injuries included bones fractures on 32 animals (13 in one single bone and 19 in multiple bones). Fractures occurred on vertebrae with fracture of spine with complete spinal cord lesion (n=9), synsacrum (n=3), fore limb (n=8), hind limb (n=7), ribs (n=8), keel (n=4), coracoid bone (n=2). Twenty-four animals presented with cutaneous lesions, 6 and 4 in the ears. Internal organs affected were kidney (n=6), liver (n=10) and lungs (n =1). Cranioencephalic lesions (n=17) included cranial skull fractures and intracranial hemorrhage (n=6) and intracranial hemorrhage without bone fracture (n=11).

Concerning cavities lesions, 28 birds had hemorrhage in coelomic cavity and two animals presented organs evisceration.

Discussion

Animal deaths as a result of vehicle collisions is increasing, probably due to the impact caused by the construction of new roads in wild habitats and the intensification of urban areas. However, the existing data may be underestimated because, depending on the size of the animal, the collision may not cause damage to the vehicle, or mortal casualties or animals may die elsewhere (Childs & Ross, 1986; Kinne, 2016; Rochlitz, 2004).

Studies on the fatal injuries associated with wildlife vehicle collisions are also scarce in Portugal, with some studies performed in amphibian on limited geographic areas (Carvalho & Mira, 2011; Matos et al., 2012; Silva et al., 2012). Additionally, animals killed by vehicle collisions are often not necropsied, due to the evidence of the cause of death, lack of partnerships between authorities and specialized laboratories and the advanced state of decay in which the carcasses of these animals are often found.

In this study, the number of wild mammals necropsied was higher than wild birds. However, the results of this study may not reflect the true relative incidence of mammal versus avian deaths. It is important to note that the low number of birds examined can be due to the fact that due to the small size of birds they may be overlooked as roadkill and bird lesions can be so severe that it is not possible to perform the necropsy. Another factor could be the fact that insurance companies or highway concessionaires' resort to the postmortem examination of animals involved in accidents when material or human damage occurs. Birds generally do not cause damage to vehicles or passengers.

For mammals, the species most identified in this study were Red Fox (*Vulpes Vulpes*) (n=45), Roe Deer (*Capreolus capreolus*) (n=16) and Common Genet (*Genetta genetta*) (n=14). In the group of birds, the most common species were Common Barn Owl (*Strix aluco*) (n=11) and Common Buzzard (*Buteo buteo*) (n=9). These results are similar to the species most often identified in other European countries in similar studies (Balčiauskas, 2009; Hell et al., 2005;

Morelle et al., 2013), but not with the data provided by the LIFE LINES project, although these species are present on their lists (Universidade de Évora, 2018b). The different results obtained could be due to the variation of the common and representative species in different geographical areas. Indeed, Vila Real is located in north Portugal, characterized by a mountainous terrain and colder climate while LIFE LINES project was performed in Évora that is located in the South of Portugal, characterized by a plain terrain and warm climate. (Bencatel et al., 2017; Cabral et al., 2005; Matias et al., 2007)

Regarding sex, our study showed that there is a higher percentage of male victims. However, in our sample, the differences between animal sex are not statically significant. We believe this is due to the fact that males will be more at risk especially in mating season (Bruinderink & Hazebroek, 2003; Rodríguez-Morales et al., 2013; Thomas et al., 2014; Zuberogoitia et al., 2014), since they have vast hunting grounds or undefined territory. Contrarily, in several studies there was a significant different between female and males, and young animals are the ones that suffer more traffic accidents, which was not observed in our study (Akinrinmade, 2002; Canal et al., 2018; Colino-Rabanal et al., 2011; Intarapanich et al., 2016; Rochlitz, 2004; Rodríguez-Morales et al., 2013).

The years with the highest number of submissions were 2016 (n=40) and 2015 (n=33). In fact, in those years there was a protocol with a concessionaire company for fast circulation roads in the northern region and animals were brought under that protocol. There are seasonal differences regarding wildlife vehicle collisions, which shows a clear superiority in March (n=41) compared to other months of the year. This can be explained with March being the beginning of spring, which is the mating season for many species.

As example, the most common species described in our study Red Fox, Common Genet, Barn Owl, Common Buzzard their mating season last from February to April in Portugal. In the case of the roe deer the fawns are born in May, and the males are more active in October, which also can explain the second pick in the autumn (Bencatel et al., 2017; Cabral et al., 2005; Loureiro et al., 2012; Mullineaux et al., 2003). During this season the most vulnerable individuals are males and animals undergoing migration (Clarke et al., 1998; Gonzalez-Prieto et al., 1993; Haigh et al., 2014; Kambourova-Ivanova et al., 2012; Philcox et al., 1999; Roedenbeck & Voser, 2008; Slater, 2002). There is also a small peak in October (n=26) can be associated to the migration of some species (particularly birds such as the common swifter) to warmer areas and the beginning of the hibernation season in other species (e.g. hedgehogs), that can be more active during this time of the year (Clarke et al., 1998; Gonzalez-Prieto et al., 1993; Haigh et al., 2014; Kambourova-Ivanova et al., 2012; Philcox et al., 1999; Roedenbeck & Voser, 2008; Slater, 2002).

According to our study, the most affected anatomical regions are body cavities both in mammals (abdomen and pelvis and thorax) and birds (celomic cavity). The numerical superiority of lesions in the thorax and abdomen is largely supported by other studies (Akinrinmade, 2002; Figuera et al., 2008; Klainbart et al., 2018; Kolata, 1980), which may be related to the animal's escape reflex when in distress. The animal escape reflex is associated to the tendency of animals to try to escape when they sense the approaching of a moving vehicle (which would then hit them toward their rear) leading to a prevalence of skeletal and muscular injuries in anatomical location of the pelvis and sacrum (Intarapanich et al., 2016).

Regarding the type of injury, this study revealed that most necropsied animals had musculoskeletal lesions (70.6%), followed by organ lesions (65.9%) and cranioencephalic lesions (34.7%). When we analyze these data by animal Class, the organs lesions were more frequent in mammals while, in birds, musculoskeletal lesions are the most frequent. Although

internal organ are more delicate structures than bone tissue constituents, is important consider that bones in birds are hollow and therefore more fragile to trauma (Brooks, 2018).

Regarding the presence or absence of blood in body cavities, it has been found in several studies that hemoperitoneum is more common in wildlife vehicle collisions situations than hemothorax. The ribs are highly resilient and therefore difficult to break, particularly in young animals (Intarapanich et al., 2016). Thus, the organs of the abdominal cavity are more susceptible, as confirmed by the large number of cases with rupture of the liver and spleen, the main causes of hemoperitoneum. Organs such the liver cannot dissipate the energy it absorbs making it more likely to rupture. Also, could be due to the animal escape reflex mentioned before (Intarapanich et al. 2016).

The main causes of hemoperitoneum were rupture of the spleen and liver, and in hemothorax were laceration of the lung and heart and rupture of the great vessels. In some cases, the presence of blood in the body cavities could not be directly associated to organs or vessel rupture or, perhaps due to unidentifiable vessel rupture or due to postmortem autolysis of the carcass.

Necropsy of wildlife vehicle collisions, although not generally critical for the diagnosis of the cause of death, may provide important data to increase knowledge in this area of pathology. This data may prove important in the evaluation and systematization of fatal injuries caused by collision with a motor vehicle. However, the real number of necropsied animals from traffic accidents could be underestimated because the authors only added to this study the animals in which there as complete history associated to vehicle collision and information regarding to the lesions observed. The “flatten” animals or in an advanced state of putrefaction were eliminated without necropsy. Further studies are needed including where is included information regarding the geographic location in which the animals were found, months of the year, species, type of fractures, complementary exams such as radiography as well as other data. In the future is necessary to improve the gathering of information regarding the animal or carcasses when they were found, associated with a more practical and simpler digital registry of the data as a LIMS system. The inclusion of animals arising from collision involving road vehicle transporting wildlife is also of interest in future studies.

Conclusion

The evaluation of vehicle collision animals and their injuries may contribute to the understanding of the associated causes and to the proposal of measures to reduce not only the number of animals involved but also the number of human injuries that occur with collision of vehicles and animals.

This study provides important information regarding which animal species are the most affected in North of Portugal by vehicle collision. The data available in this area is still very scarce. Although, most of the species recorded as casualties in this study are considered common within the adjacent natural areas it is important determining the impacts of road and railways, since this type of structure is expanding rapidly across the world and how they are affecting the wild fauna.

It seems impossible to avoid all collisions but is possible to mitigate them. Studies of this kind can be used to identify measures which may reduce the number of accidents: 1) advise greater caution of drivers when in forested areas during mating season and migration; 2) clearing the vegetation adjacent to roads to allow the driver to see the animal before it enters the roadway; 3) construction of animal passages or the maintenance of non-specific passages for wildlife; 4) reducing vehicle speed in high-risk areas allowing the driver more time to

react after spotting the animal; 5) noise barriers and low-noise road surfaces to reduce traffic sound that can act as a physical barrier.

Also, the authors hope these first results will be useful in convincing stakeholders (police, local authorities, insurance companies, forest administration, naturalists, drivers) to collaborate so that better data collection could be achieved in the future.

Acknowledgement

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

References

- [1] Akinrinmade, J. F. (2002). Evaluation of Pattern of Pet Animal Trauma at the Veterinary Teaching Hospital, Ibadan, Nigeria. *Nigerian Veterinary Journal*, 35(3), 1007–1014.
- [2] Ascensão, F., & Mira, A. (2006). *Impactes das vias rodoviárias na fauna silvestre*.
- [3] Bager, A., & Da Rosa, C. A. (2011). Influence of sampling effort on the estimated richness of road-killed vertebrate wildlife. *Environmental Management*, 47(5), 851–858. <https://doi.org/10.1007/s00267-011-9656-x>
- [4] Balčiauskas, L. (2009). Distribution of species-specific wildlife-vehicle accidents on lithuanian roads, 2002-2007. *Estonian Journal of Ecology*, 58(3), 157–168. <https://doi.org/10.3176/eco.2009.3.01>
- [5] Bencatel, J., Álvares, F., Moura, A. E., & Barbosa, A. M. (Eds.). (2017). *Atlas de Mamíferos de Portugal* (1st ed.). Universidade de Évora.
- [6] Bishop, C. A., & Brogan, J. M. (2013). Estimates of Avian Mortality Attributed to Vehicle Collisions in Canada Estimation de la mortalité aviaire attribuable aux collisions automobiles au Canada. *Avian Conservation Ecology*, 8(2).
- [7] Brendan, A., Taylor, D., Ross, A., & Goldingay, L. (2010). Roads and wildlife : impacts , mitigation and implications for wildlife management in Australia. *Wildlife Research*, 37, 320–331.
- [8] Brooks, J. W. (2018). *Veterinary Forensic Pathology* (J. W. Brooks (Ed.); Vol. 2). Springer International Publishing. <https://doi.org/10.1007/978-3-319-67172-7>
- [9] Bruinderink, G. W. T. A. G., & Hazebroek, E. (2003). Ungulate Traffic Collisions in Europe. *Conservation Biology*, 10(4), 1059–1067. <https://doi.org/10.1046/j.1523-1739.1996.10041059.x>
- [10] Cabral, M. J., J. Almeida, P. R., Almeida, T., Delliger, N., Ferrand de Almeida, M.E. Oliveira, J. M., Palmeirim, A. I., Queirós, L., Rogado, M., & Santos-Reis. (2005). *Livro Vermelho dos Vertebrados de Portugal* (M. S.-R. Cabral, M.J. ; J. Almeida, P.R. Almeida, T. Delliger, N. Ferrand de Almeida, M.E. Oliveira, J.M. Palmeirim, A.I. Queirós, L. Rogado (Ed.)). Instituto da Conservação da Natureza. <https://doi.org/http://hdl.handle.net/10174/6006>
- [11] Canal, D., Camacho, C., Martin, B., de Lucas, M., & Ferrer, M. (2018). Magnitude, composition and spatiotemporal patterns of vertebrate roadkill at regional scales: a study in southern Spain. *Animal Biodiversity and Conservation*, 41(2), 281–300.
- [12] Carvalho, F., & Mira, A. (2011). Comparing annual vertebrate road kills over two time periods, 9 years apart: A case study in Mediterranean farmland. *European Journal of Wildlife Research*, 57(1), 157–174. <https://doi.org/10.1007/s10344-010-0410-0>
- [13] Childs, J. E., & Ross, L. (1986). Urban cats: characteristics and estimation of mortality due to motor vehicles. *American Journal of Veterinary Research*, 47(7), 1643–1648.
- [14] Clarke, G. P., White, P. C. L., & Harris, S. (1998). Effects of roads on badger *Meles meles* populations in south-west England. *Biological Conservation*, 86(2), 117–124. [https://doi.org/10.1016/S0006-3207\(98\)00018-4](https://doi.org/10.1016/S0006-3207(98)00018-4)
- [15] Colino-Rabanal, V. J., Lizana, M., & Peris, S. J. (2011). Factors influencing wolf *Canis lupus* roadkills in Northwest Spain. *European Journal of Wildlife Research*, 57(3), 399–409. <https://doi.org/10.1007/s10344-010-0446-1>

- [16] Corrêa, L. L. C., Silva, D. E., Oliveira, S. V. de, Finger, J. V. G., Santos, C. R. dos, & Petry, M. V. (2017). Vertebrate road kill survey on a highway in southern Brazil. *Acta Scientiarum. Biological Sciences*, 39(2), 219. <https://doi.org/10.4025/actascibiolsci.v39i2.33788>
- [17] Editorial, G., Ree, R. Van Der, Jaeger, J. A. G., Grift, E. A. Van Der, & Clevenger, A. P. (2011). Effects of Roads and Traffic on Wildlife Populations and Landscape Function : Road Ecology is Moving toward Larger Scales. *Ecology and Society*, 16(1).
- [18] Edwards, P. J., Fletcher, M. R., & Berny, P. (2000). Review of the factors affecting the decline of the European brown hare , *Lepus europaeus* (Pallas , 1778) and the use of wildlife incident data to evaluate the significance of paraquat. *Agricultures, Ecosystems and Environment*, 79, 95–103.
- [19] Elzanowski, A., & Ciesio, J. (2009). Amphibian road mortality in Europe : a meta-analysis with new data from Poland. *European Journal of Wildlife Research*, 55, 33–43. <https://doi.org/10.1007/s10344-008-0211-x>
- [20] Fahrig, L. (2003). Effects of habitat fragmentation on biodiversity. *Annu. Rev. Ecol. Evol. Syst.*, 34, 487–515. <https://doi.org/10.1146/annurev.ecolsys.34.011802.132419>
- [21] Figuera, R. A., Silva, M. C. da, Souza, T. M. de, Brum, J. S., Kommers, G. D., Graça, D. L., Irigoyen, L. F., & Barros, C. S. L. de. (2008). Aspectos patológicos de 155 casos fatais de cães atropelados por veículos automotivos. *Ciência Rural*, 38(5), 1375–1380.
- [22] Garcês, A., & Pires, I. (2017). *Manual de Técnicas de Necropsia em Animais Selvagens* (A Garcês & I. Pires (Eds.); 1st ed.). artelogy.
- [23] Garcia, G. (2012). *Monitorização da Mortalidade de Fauna nas Estradas da EP: Relatório Síntese, 2011*.
- [24] Garcia, G. (2013). *Monitorização da Mortalidade de Fauna nas Estradas da EP: Relatório Síntese, 2012*.
- [25] Garcia, G. (2014). *Monitorização da Mortalidade de Fauna nas Estradas da EP: Relatório Síntese, 2013*.
- [26] Garcia, G. (2015). *Monitorização da Mortalidade de Fauna nas Estradas da EP: Relatório Síntese, 2014*.
- [27] Garcia, G. (2016). *Monitorização da Mortalidade de Fauna nas Estradas da IP: Relatório Síntese, 2015*.
- [28] Gonzalez-Prieto, S., Villarino, A., & Freán, M. (1993). Mortalidade de vertebrados por atropello en una carretera nacional del NO de Espana. *Ecologia*, 7, 375–389.
- [29] Haigh, A., O’Riordan, R. M., & Butler, F. (2014). Hedgehog *Erinaceus europaeus* mortality on Irish roads . *Wildlife Biology*, 20(3), 155–160. <https://doi.org/10.2981/wlb.12126>
- [30] Hell, P., Plavý, R., Slamečka, J., & Gašparík, J. (2005). Losses of mammals (Mammalia) and birds (Aves) on roads in the Slovak part of the Danube Basin. *European Journal of Wildlife Research*, 51(1), 35–40. <https://doi.org/10.1007/s10344-004-0068-6>
- [31] Hels, T., & Buchwald, E. (2001). The effect of road kills on amphibian populations. *Biological Conservation*, 99(3), 331–340. [https://doi.org/10.1016/S0006-3207\(00\)00215-9](https://doi.org/10.1016/S0006-3207(00)00215-9)
- [32] Hernandez, M. (1988). Road mortality of the little owl (*Athene noctua*) in Spain. *Journal of Raptor Research*, 22(3), 81–84. <http://elibrary.unm.edu/sora/jrr/v022n03/p00081-p00084.pdf>
- [33] Hostetler, J. A., Walter McCown, J., Garrison, E. P., Neils, A. M., Barrett, M. A., Sunquist, M. E., Simek, S. L., & OIL, M. (2009). Demographic consequences of anthropogenic influences: Florida black bears in north-central Florida. *Biological Conservation*, 142(11), 2456–2463. <https://doi.org/10.1016/j.biocon.2009.05.029>
- [34] Intarapanich, N. P., McCobb, E. C., Reisman, R. W., Rozanski, E. A., & Intarapanich, P. P. (2016). Characterization and Comparison of Injuries Caused by Accidental and Non-accidental Blunt Force Trauma in Dogs and Cats. *Journal of Forensic Sciences*, 61(4), 993–999. <https://doi.org/10.1111/1556-4029.13074>
- [35] Jackson, N. D., & Fahrig, L. (2011). Relative effects of road mortality and decreased connectivity on population genetic diversity. *Biological Conservation*, 144(12), 3143–3148. <https://doi.org/10.1016/j.biocon.2011.09.010>
- [36] Kambourova-Ivanova, N., Koshev, Y., Popgeorgiev, G., Ragyov, D., Pavlova, M., Mollov, I.,

- & Nedialkov, N. (2012). Effect of traffic on mortality of amphibians, reptiles, birds and mammals on two types of roads between Pazardzhik and Plovdiv region (Bulgaria) - Preliminary results. *Acta Zoologica Bulgarica*, 64(1), 57–67.
- [37] Kinne, J. (2016). Postmortem Examination. In J. Samour (Ed.), *Avian Medicine* (3rd ed., pp. 567–578). Elsevier. <https://doi.org/10.1016/B978-0-7506-3598-1.50020-5>
- [38] Klainbart, S., Bibring, U., Strich, D., Chai, O., Bdolah-Abram, T., Aroch, I., & Kelmer, E. (2018). Retrospective evaluation of 140 dogs involved in road traffic accidents. In *Veterinary Record* (Vol. 182, Issue 7, p. 196). <https://doi.org/10.1136/vr.104293>
- [39] Knutson, R. M. (1987). *Flattened Fauna: A Field Guide to Common Animals of Roads, Streets and Highways* (10th ed.). Ten Speed Press.
- [40] Kolata, R. J. (1980). Trauma in dogs and cats: an overview. *The Veterinary Clinics of North America. Small Animal Practice*, 10(3), 515–522. [https://doi.org/10.1016/S0195-5616\(80\)50051-3](https://doi.org/10.1016/S0195-5616(80)50051-3)
- [41] Lindenmayer, D. B., Mccarthy, M. A., Parris, K. M., & Pope, M. L. (2000). Habitat fragmentation , landscape context , and mammalian assemblages in southeastern australia. *Journal of Mammalogy*, 81(3), 787–797.
- [42] Loss, S. R., Conservation, S., Park, N. Z., & Box, P. O. (2014). Estimation of Bird-Vehicle Collision Mortality on U . S . Roads. *The Journal of Wildlife Management*, 78(5), 763–771. <https://doi.org/10.1002/jwmg.721>
- [43] Loureiro, F., Pedrodo, N., Santos, M. J., & Rosalino, L. M. (2012). *Um olhar sobre os Carnívoros Portugueses*. Carnivora.
- [44] Marcantonio, M., Rocchini, D., Geri, F., & Bacaro, G. (2013). Biodiversity , roads , & landscape fragmentation : Two Mediterranean cases. *Applied Geography*, 42, 63–72. <https://doi.org/10.1016/j.apgeog.2013.05.001>
- [45] Matias, R., Catry, P., Costa, H., Elias, G., Jara, J., Moore, C. C., & Tomé, R. (2007). Lista sistemática das aves de Portugal Continental. *Anuário Ornitológico*, 5, 74–132.
- [46] Matos, C., Sillero, N., & Argana, E. (2012). Spatial analysis of Amphibian road mortality levels in northern Portugal country roads. *Amphibia Reptilia*, 33(3–4), 469–483. <https://doi.org/10.1163/15685381-00002850>
- [47] Meirinho, A., Barros, N., Oliveira, N., Catry, P., Lecoq, M., Paiva, V., Geraldés, P., Granadeiro, J. P., Ramírez, I., & Andrade, J. (2014). *Atlas das Aves Marinhas de Portugal*.
- [48] Molina-López, R. A., Casal, J., & Darwich, L. (2011). Causes of morbidity in wild raptor populations admitted at a wildlife rehabilitation centre in Spain from 1995-2007: A long term retrospective study. *PLoS ONE*, 6(9). <https://doi.org/10.1371/journal.pone.0024603>
- [49] Morelle, K., Lehaire, F., & Lejeune, P. (2013). Spatio-temporal patterns of wildlife-vehicle collisions in a region with a high-density road network. *Nature Conservation*, 73, 53–73. <https://doi.org/10.3897/natureconservation.5.4634>
- [50] Morelli, F., Beim, M., Jerzak, L., Jones, D., & Tryjanowski, P. (2014). Can roads, railways and related structures have positive effects on birds? - A review. *Transportation Research Part D: Transport and Environment*, 30, 21–31. <https://doi.org/10.1016/j.trd.2014.05.006>
- [51] Mullineaux, E., Best, D., & Cooper, J. E. (2003). *BSAVA manual of wildlife casualties*. British Small Animal Veterinary Association.
- [52] Philcox, C. K., Grogan, A. L., & Macdonald, D. W. (1999). Patterns of otter *Lutra lutra* road mortality in Britain. *Journal of Applied Ecology*, 36, 748–762.
- [53] Ressel, L., Hetzel, U., & Ricci, E. (2016). Blunt Force Trauma in Veterinary Forensic Pathology. *Veterinary Pathology*, 53(5), 941–996.
- [54] Riley, S. P. D., Sauvajot, R. M., Fuller, T. K., York, E. C., Kamradt, D. A., Bromley, C., Wayne, R. K., Monica, S., National, M., Area, R., Hillcrest, W., & Oaks, T. (2003). Effects of Urbanization and Habitat Fragmentation on Bobcats and Coyotes in Southern California. *Conservation Biology*, 17(2), 566–576.
- [55] Rochlitz, I. (2004). Clinical study of cats injured and killed in road traffic accidents in Cambridgeshire. *Journal of Small Animal Practice*, 45(8), 390–394. <https://doi.org/10.1111/j.1748-5827.2004.tb00253.x>

- [56] Rodríguez-Morales, B., Díaz-Varela, E. R., & Marey-Pérez, M. F. (2013). Spatiotemporal analysis of vehicle collisions involving wild boar and roe deer in NW Spain. *Accident Analysis and Prevention*, 60, 121–133. <https://doi.org/10.1016/j.aap.2013.07.032>
- [57] Rodríguez, B., Rodríguez, A., Siverio, F., & Siverio, M. (2010). Causes of Raptor Admissions to a Wildlife Rehabilitation Center in Tenerife (Canary Islands). *Journal of Raptor Research*, 44(1), 30–39. <https://doi.org/10.3356/JRR-09-40.1>
- [58] Roedenbeck, I. A., & Voser, P. (2008). Effects of roads on spatial distribution, abundance and mortality of brown hare (*Lepus europaeus*) in Switzerland. *European Journal of Wildlife Research*, 54(3), 425–437. <https://doi.org/10.1007/s10344-007-0166-3>
- [59] Rogers, E., & Stern, A. W. (2018). *Veterinary Forensics Investigation, Evidence Collection, and Expert Testimony* (1st editio). CRC Press Taylor& Francis Group.
- [60] Sillero, N. (2008). Amphibian mortality levels on Spanish country roads: descriptive and spatial analysis. *Amphibia Reptilia*, 29, 337–347.
- [61] Silva, C. C., Lourenço, R., Godinho, S., Gomes, E., Sabino-Marques, H., Medinas, D., Neves, V., Silva, C., Rabaça, J. E., & Mira, A. (2012). Major Roads Have a Negative Impact on the Tawny Owl *Strix aluco* and the Little Owl *Athene noctua* Populations. *Acta Ornithologica*, 47(1), 47–54. <https://doi.org/10.3161/000164512x653917>
- [62] Slater, F. M. (2002). An assessment of wildlife road casualties - The potential discrepancy between numbers counted and numbers killed. *Web Ecology*, 3, 33–42. <https://doi.org/10.5194/we-3-33-2002>
- [63] Taylor, S. K., Buergelt, C. D., Roelke-parker, M. E., Homer, B. L., & Rotstein, D. S. (2002). Causes of mortality of free-ranging florida panthers. *Journal of Wildlife Disease*, 38(1), 107–114.
- [64] Thomas, R. L., Baker, P. J., & Fellowes, M. D. E. (2014). Ranging characteristics of the domestic cat (*Felis catus*) in an urban environment. *Urban Ecosystems*, 17(4), 911–921. <https://doi.org/10.1007/s11252-014-0360-5>
- [65] Universidade de Évora. (2018a). *LIFE LINES*. Universidade de Évora.
- [66] Universidade de Évora. (2018b). *LIFE LINES National Roadkill Database - Public data - Lizmap*. Universidade de Évora.
- [67] Visintin, C., Ree, R. Van Der, & Mccarthy, M. A. (2017). Consistent patterns of vehicle collision risk for six mammal species. *Journal of Environmental Management*, 201, 397–406. <https://doi.org/10.1016/j.jenvman.2017.05.071>
- [68] Zuberogoitia, I., Del Real, J., Torres, J. J., Rodríguez, L., Alonso, M., & Zabala, J. (2014). Ungulate vehicle collisions in a peri-urban environment: Consequences of transportation infrastructures planned assuming the absence of ungulates. *PLoS ONE*, 9(9). <https://doi.org/10.1371/journal.pone.0107713>