



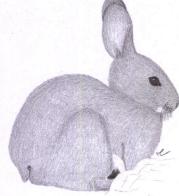
### Mestrado em Biologia da Conservação



# Factores que afectam a mortalidade de predadores por atropelamento: será a disponibilidade de presas importante?



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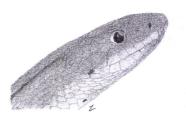




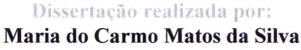
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# Factores que afectam a mortalidade de predadores por atropelamento: será a disponibilidade de presas importante?

#### Resumo

Potencialmente, as bermas de estrada providenciam habitat para micromamíferos e coelhos, incrementando a sua abundância e atraindo predadores para a proximidade das estradas. Este estudo pretendeu determinar a relação entre a abundância de presas, micromamíferos e coelhos, nas bermas e a mortalidade de predadores por atropelamento, tendo em consideração características da estrada e da berma e a paisagem adjacente, a Sul de Portugal. As relações entre a mortalidade de predadores e os diferentes tipos de variáveis exploratórias foram analisados através de GLM (Generalized Linear Models) e RDA (Redundancy Analysis). Os resultados demonstraram que as presas são um factor importante aumentando a ocorrência de mortalidade de predadores, particularmente dos mamíferos carnívoros e rapinas nocturnas. Adicionalmente, este grupo de variáveis, juntamente com a paisagem, explica a maioria da mortalidade de *Elaphe scalaris*, *Tyto alba* e *Herpestes ichneumon*. São analisadas medidas de gestão de bermas e propostas acções de mitigação dos efeitos negativos da estrada.

## Factors influencing predator road-kills: does prey availability matter?

#### **Abstract**

Verges provide potential habitat for small mammals and rabbits, especially if habitat of adjacent areas is not suitable. The increasing use of verges by these preys might attract predators to the road neighbouring. This study aimed to access the effect of prey availability (rabbits and small mammals) on verges and predator road casualties, taking into account road and verge characteristics and the surrounding landscape, in southern Portugal. Relationships among predator mortality and the different kinds of explanatory variables were evaluated with Generalized Linear Models and Redundancy Analysis. Results have shown that prey is an important factor enhancing occurrence of predator mortality especially for carnivore mammals and owls. Moreover, this set of variables, along with landscape, explains most of the fatalities of *Elaphe scalaris*, *Tyto alba* and *Herpestes ichneumon*. Implications for verge management and implementation of mitigation actions are discussed.

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#### Introdução

A ecologia de estradas é a área da ecologia que estuda a interacção entre os organismos e o sistema estrada-ambiente (Forman *et al.*, 2003). As estradas afectam os componentes bióticos e abióticos da paisagem, através da alteração da dinâmica das populações de plantas e animais, da modificação dos fluxos de materiais, introdução de elementos exóticos e alteração da disponibilidade de recursos, como a água, luz e nutrientes (Coffin, *in press*). Visto que, para além disso, a quantidade de estradas e o tráfego continuam, globalmente, a aumentar (Forman, 2000), torna-se relevante analisar os efeitos destas infra-estruturas nos ecossistemas.

O aumento do número de estradas afecta, de diversas formas, os ecossistemas e a conservação de biodiversidade (e.g. Forman and Alexander, 1998). A perda e fragmentação de habitats, o efeito barreira, a perturbação e poluição, o papel ecológico das bermas e a mortalidade por atropelamento são considerados os maiores impactos na biodiversidade (e.g. Forman and Alexander, 1998; Forman et al., 2003; Iuell et al., 2003).

A perda e fragmentação de habitats e o efeito barreira afectam a dinâmica das populações animais, subdividindo populações inicialmente grandes em populações mais pequenas, à medida que surgem áreas de menores dimensões, devido à densidade crescente de estradas. Este facto provoca o aparecimento de populações isoladas, menos estáveis e mais vulneráveis à extinção, que utilizam áreas distantes das estradas (e.g. Forman and Alexander, 1998; Seiler, 2003). O uso de habitats adjacentes à estrada pela fauna também pode ser alterado pelo efeito da perturbação e poluição causadas pelas estradas em si e pelo tráfego a elas inerente (e.g. Iuell *et al.*, 2003). A poluição altera as características químicas e ecológicas dos habitats (e.g. Iuell *et al.*, 2003; Seiler, 2003), enquanto a perturbação sob, por exemplo, a forma de ruído e vibração pode repelir algumas espécies (e.g. Forman and Alexander, 1998; Iuell *et al.*, 2003; Seiler, 2003).

Apesar das bermas da estrada poderem ter um efeito negativo, aumentando a mortalidade da fauna e facilitando o estabelecimento de espécies exóticas nos ecossistemas, também podem funcionar como corredores ecológicos, permitindo o movimento e conectividade entre populações e proporcionando, simultaneamente, habitat para algumas espécies de plantas e animais (e.g. Forman *et al.*, 2003; Iuell *et al.*, 2003). As bermas podem fomentar a manutenção de populações de herbáceas anuais, como gramíneas, (Forman *et al.*, 2003; Santos *et al.*, 2007) e de arbustivas, na maioria

constituídas por espécies resistentes a perturbações antropogénicas, como o corte, regularmente aplicado como forma de controlo da vegetação, e o geralmente elevado nível de nitratos no solo (Spooner et al., 2004; Hovd and Skongen, 2005; Quintana-Ascencio et al., 2007). A variabilidade e riqueza de espécies vegetais variam conforme o habitat adjacente e as perturbações que afectam as bermas, como o período entre os cortes e a concentração de poluentes (Hovd and Skongen, 2005; Truscott et al., 2005). Em regiões de agricultura intensiva ou em zonas urbanas, as espécies autóctones (Forman et al., 2003) e as que incluem plantas mais altas e escapam ao impacto da gestão humana (por exemplo pastoreio; Forman et al., 2003; Santos et al., 2007) são as que mais frequentemente persistem nas bermas. Dadas as características da sua vegetação, as bermas proporcionam habitat potencial para espécies animais, por exemplo de micromamíferos, em regiões onde o habitat adequado é um factor limitante, como observado por diversos autores (Bellamy et al., 2000; Bolger et al., 2001), incluindo em Portugal (Ramalho 2007; Santos et al., 2007). De acordo com Ramalho (2007), em regiões de pastoreio intensivo, a altura e densidade da vegetação são reduzidas, levando a uma maior abundância de micromamíferos nas bermas, relativamente à matriz envolvente.

A mortalidade por atropelamento tem aparentemente um efeito mínimo no tamanho das populações de espécies abundantes, como os coelhos e os pardais (Forman and Alexander, 1998; Seiler, 2003), mas é considerada uma das causas mais importantes de mortalidade para algumas espécies raras (Seiler, 2003), como Lynx pardinus (Ferrera et al., 1992). Dados recolhidos em muitas regiões do mundo revelam que os atropelamentos são responsáveis pela morte de milhões de animais de diversas espécies e por ferimentos num número ainda mais elevado (Forman and Alexander, 1998; Iuell et al., 2003). Nas últimas décadas, o impacto da morte por atropelamento terá, provavelmente, superado o da caça, até então considerada a primeira causa directa de mortalidade de predadores terrestres (Forman and Alexander, 1998). Entre os diferentes factores causadores de mortalidade, destacam-se os meteorológicos, como a temperatura e a precipitação (Clevenger et al., 2003), a abundância, distribuição e comportamento das espécies como, por exemplo, os padrões sazonais relativos à reprodução e distribuição (Clevenger et al., 2003; Seiler, 2003), a densidade e velocidade de tráfego, os padrões espaciais da paisagem (e.g. Forman and Alexander, 1998; Clevenger et al., 2003) e as características da estrada e da berma, nomeadamente a topografia local, a vegetação e a localização e tipo de cercas (Clevenger et al., 2003). No entanto, nenhum estudo considera a influência da abundância de presas como uma possível causa da mortalidade de predadores. Bellamy *et al.* (2000) sugerem que a presença de micromamíferos nas bermas pode influenciar a probabilidade de atropelamento de predadores, uma vez que aumenta o uso das bermas por estas espécies. Com o propósito de elaborar e aplicar medidas efectivas de mitigação para reduzir os atropelamentos, é importante quantificar o efeito destas causas na mortalidade.

Assim, com este estudo pretendeu-se determinar a relação entre a abundância de micromamíferos e coelhos nas bermas e a mortalidade de predadores por atropelamento, tendo em consideração características da estrada e da berma e a paisagem envolvente. Através da definição dos factores mais importantes para a mortalidade por atropelamento, espera-se contribuir para o estabelecimento de linhas orientadoras para a gestão adequada das estradas e bermas, de forma a reduzir o efeito de uma das mais mortíferas armadilhas criadas pelo homem.

#### Área de estudo

O estudo foi realizado ao longo de um troço de cerca de 10 km da Estrada Nacional 4 (EN4) (Figura 1), localizada no Concelho de Montemor-o-Novo, sendo a maior estrada que liga Lisboa a Madrid. Esta estrada tem uma largura de cerca de 12 m e duas faixas de rodagem. O troço seleccionado foi dividido em 40 secções de 250 m, cada uma das quais foi caracterizada a nível das diferentes variáveis estudadas.

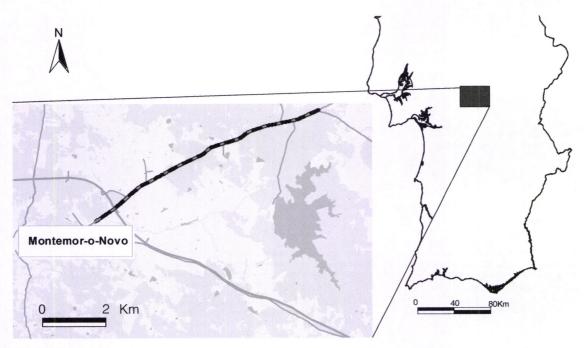


Figura 1 – Localização da estrada amostrada (representada a preto; adaptado de UBC, *in press*). A zona florestal está representada a cinzento-claro e outras estradas e as albufeiras a cinzento-escuro.

Em termos biogeográficos, a área de estudo localiza-se no Reino Holártico, Região Mediterrânica (Costa *et al.*, 1999; Rivas-Martínez and Arregui, 1999), Subsector Araceno-Pacense, Superdistrito Alto Alentejano (Costa *et al.*, 1999).

O concelho de Montemor-o-Novo apresenta um Bioclima Oceânico do Termotipo Mesomediterrânico (Rivas-Martínez and Arregui, 1999), variando a precipitação mensal entre 1,2 mm e 75.3 mm (SNIRH, 2007).

Relativamente aos usos do solo, na área de estudo dominam os montados, com um sub-coberto variável entre matos, culturas anuais e pastoreio. Para além dos montados, as pastagens e as culturas anuais também apresentam uma representatividade significativa (UBC, in press.), patente na Figura 2.

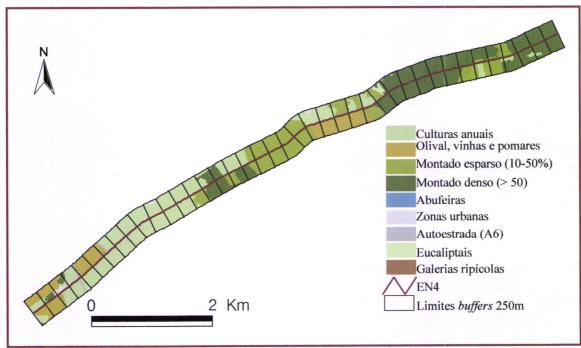


Figura 2 – Mapa de usos do solo do troço estudado na estrada EN4, em *buffers* de 250 m (UBC, *in press*).

Os usos do solo foram considerados em *buffers* de 250 m, de modo a incluir os efeitos causados pela estrada nos habitats adjacentes. Em termos percentuais, cerca de 57% da área são ocupados por montado, 29% por culturas anuais e 12% por vinhas e olivais (UBC, *in press*). Estes usos do solo resultam essencialmente das actividades humanas no concelho, uma vez que as actividades agrícolas, pecuárias e florestais continuam a assumir um peso relevante (Câmara Municipal de Montemor-o-Novo, 2007). Das principais produções destacam-se a cortiça, o azeite, o vinho e o mel (Câmara Municipal de Montemor-o-Novo, 2007).

Este concelho apresenta uma população residente de cerca de 18 578 indivíduos (INE, 2001). No ano 2005, a média de circulação de veículos na estrada EN4 foi de 3688 veículos por dia e 493 por noite, sendo a média em cada período de 24 horas de 4181 (EP, 2005), patente na Tabela 1. Esta estrada tem uma largura média de 12 m e apresenta cercas em ambas as bermas em 65% do seu comprimento.

Tabela 1 – Recenseamento de tráfego da N4, no posto de contagem 670, durante os períodos diurno, nocturno e diário, para o ano de 2005 (EP, 2005).

Designação	Diurno (16h)	Nocturno (8h)	Diário (24h)	
Velocípedes	11	0	11	
Ligeiros	2774	369	3143	
Pesados	903	124	1027	
Total	3688	493	4181	

## Factors influencing predator road-kills: does prey availability matter?

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#### **Abstract**

Road mortality is the most noticeable effect of roads on wildlife. However, verges may provide important refuges for small mammals and rabbits, particularly when roads cross intensive agricultural or grazed areas. In these circumstances, the increasing use of verges by small prey species may attract predators to road surroundings potentiating their road-kills. The aim of this study is to quantify the role of prey availability (rabbits and small mammals) on predator road casualties, taking into account road and surrounding landscape. We analyzed this effect on different kinds of predators including snakes, owls and mammal carnivores. The study took place in a 10 km stretch of a main National Road (EN4) in southern Portugal. Relationships among predator mortality and the different kinds of explanatory variables were evaluated with Generalized Linear Models and multivariate Redundancy Analysis (RDA). Results GLM have shown that prey are an important factor enhancing occurrence of predator mortality especially for carnivore mammals and owls. Moreover RDA reveals that an important part of all predators road-kills is related with prey abundance on verges and that this set of variables, along with landscape, explains most of the fatalities of Elaphe scalaris, Tyto alba and Herpestes ichneumon. Implications for verge management and implementation of mitigation actions are discussed.

**Key words:** Mammal carnivores, owls, snakes, road-kills, prey availability, landscape road verges, variance partitioning.

#### **Introduction**

The road network has increased worldwide in the last two decades (Forman, 2000). Moreover, many roads were rebuilt in order to account for a large raise in the number and speed of circulating cars. The high dominance of these linear infrastructures on modern landscapes has strong impacts on wildlife populations of which, road-killings are the most noticed.

Road-kills apparently have minimal effects on population size for abundant species, like rabbits and sparrows (Forman and Alexander, 1998; Seiler, 2003). They are, however, considered as one of the most important causes of mortality for some rare species (Seiler, 2003), like Lynx pardinus (Ferrera et al., 1992). Data gathered worldwide show that road-kills are responsible for killing millions of animals of different species and injured many more (Forman and Alexander, 1998; Iuell et al., 2003). This impact has probably overtaken hunting during the last decades as the primary direct cause of vertebrate mortality on land (Forman and Alexander, 1998). Several factors influencing road-kills have been widely studied. Factors such as weather, like temperature and precipitation (Clevenger et al., 2003), animal abundance, distribution and seasonal behaviour, such as breeding and dispersion patterns (Clevenger et al., 2003; Seiler, 2003), vehicle traffic levels and speed, landscape patterns (e.g. Forman and Alexander, 1998; Clevenger et al., 2003), road and verges characteristics, like local topography, vegetation and fence location or type (Clevenger et al., 2003) are often referred as influencing wildlife road patterns.

However, despite the negative effect in favouring the dispersion of alien species, road verges may also provide shelter and wildlife corridors for native species allowing smaller species to persist and move in these roadside habitats (e.g. Forman et al., 2003; Iuell et al., 2003). For instance, road verges may have higher densities of annual herbaceous, like grasses and forbs (Forman et al., 2003; Santos et al., 2007) and shrub species than the surrounding areas (Spooner et al., 2004; Hovd and. Skongen, 2005; Quintana-Ascencio et al., 2007). Nevertheless, variability and richness of plant species on verges changes according to the level of agriculture intensification in adjacent patches and the kind of disturbances that affect most verges, like mowing periodicity and pollutants concentrations (Hovd and. Skongen, 2005; Truscott et al., 2005). In intensive farmed lands or urbanized areas, spontaneous species and taller plants that escaped human management impact (e.g. grazing) remain easily on road verges than on

the surrounding landscape (Santos et al., 2007; Forman et al., 2003). These characteristics of verges provide potential habitat for animal species, like small mammals, in regions where suitable habitat is a limiting factor, as observed by several authors (Bellamy et al., 2000; Bolger et al., 2001) including in Portugal (Ramalho (2007; Santos et al., 2007; Marques et al., in prep.). According to Ramalho (2007), the higher density of small mammals on road verges relative to adjacent areas might be related to the reduction of vegetation height on the latter, due to intensive grazing, which would therefore offer poor shelter and food conditions for these species.

Bellamy *et al.* (2000) have already pointed out that the presence of small mammals on road verges could increase the probability of predators being road-killed. However, although several studies have already provided information on causes of road mortality, there are no studies focusing the role of prey availability or attempting to quantify its importance on predator causalities.

This study aimed to quantify the contribution of small mammal and rabbit abundance on road verges for predator road-kills, taking into account two other important factors: i) road and verge characteristics and ii) surrounding matrix characteristics. Moreover, we studied these effects on different types of predators including snakes, owls and mammal carnivores. Therefore, our results might provide information to lay down general guidelines for proper road verge management, in order to reduce the impacts of one of the deadliest human made traps.

#### Methodology

#### Study area

Our study took place in southern Portugal on a 10 km stretch of National Road 4 (EN4), a major road linking Lisbon to Madrid (Figure 1). The road is 12 m wide, has two traffic lanes and partially (1-2 m wide) paved verges. In 2005, the surveyed stretch presented moderate traffic volume, (a mean of 4181 vehicles day<sup>-1</sup>; EP, 2005). Fences were present in both sides of the road on 65% of its entire length.

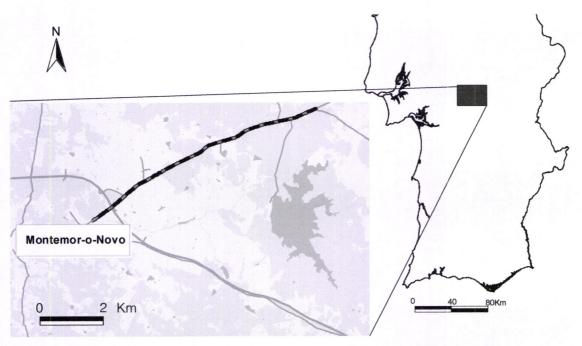


Figure 1 – Map of the surveyed road (in black) and its location on Portuguese territory (adapted from UBC, *in press*). Forested areas are depicted in light grey and other roads and water reservoirs in dark grey.

The studied road crosses typical Mediterranean agro-pastoral woodlands, locally know as *montado* (57%), open areas including cereal crops and fallows (29%) and vines and olive yards (12%). Topography is mainly plain, with altitude ranging from 100 to 200 m (Instituto do Ambiente, 2007).

In terms of bioclimate typology, this region is included in the Mesomediterranean thermotype of the Mediterranean pluviseasonal-oceanic bioclimate (Rivas-Martínez and Arregui, 1999), with dry and hot summers and cold and wet winters. During the study year (2007), monthly average precipitation ranged from 1.2 mm to 75.3 mm (SNIRH, 2007).

#### Sampling design

For analysis purposes, the surveyed 10 km road stretch was divided into forty 250 m length segments. This segment length was considered adequate to both study purposes and implementation of detailed management actions aiming to mitigate predator road-kills. All response and explanatory variables were defined for each of

these segments. The geographical coordinates location of all predator and prey records were obtained through a global positioning system (GPS) unit.

#### Predator road-kills surveys

During the year 2007, road stretch was systematically surveyed for road fatalities of carnivores, owls and snakes. Surveys were performed by car at 40-50 km per hour, in the morning, on a daily basis. Road-killed animals were collected and identified to species level, whenever possible.

#### Prey surveys

#### Aboveground small mammals trapping

Small mammals were captured using medium size Sherman live traps baited with oats and sardine, and using cotton for bedding. Traps were placed alternately along both road verges, at 50 m intervals, in a total of 200 trapping points. Surveys were conducted for four consecutive nights, during the last week of November 2007. Traps were checked everyday at sunrise and captured small mammals were identified to species level, sexed and weighted. Additionally, each captured individual was marked with a fur clip, in order to set up a unique mark identifying verge and day of capture. Afterwards, animals were released at site of capture.

Data was processed by sector as number of individuals of wood mouse (Apodemus sylvaticus), Algerian mouse (Mus spretus) and White-toothed shrew (Crocidura russula) captured and total number of individuals of all species. Species with captures lower than 3 individuals (brown rat, Rattus norvegicus and black rat, Rattus rattus) were not considered individually.

#### Fossorial vole presence signs

The abundance of Mediterranean pine vole (*Microtus duodecimcostatus*) was accessed by presence signs detection, since fossorial mammals (e.g. voles) are highly adapted for subterranean life and therefore are not easily captured by traditional trapping methods (Wilson, *et al.*, 1996). In fact, the underground *M. duodecimcostatus* presence can easily be detected by the typical form and relative position of the aboveground mole-hills (Mira, 1999). Vole survey was carried out through mole-hill

counting on a line transect of 10 km along each verge. Mole-hills distancing less than 3 metres were considered as belonging to the same colony for abundance evaluation proposes. Results are presented as the number of colonies per road sector.

#### Rabbit counting

To estimate the abundance of rabbits (*Oryctolagus cuniculus*) five surveys were conducted on each road direction, in a total of ten surveys, driving at 30-40 km per hour and counting rabbits on a 20 m distance band from the paved boundary. Surveys were carried out twice a week between 6 h and 8 h a.m., during May and June 2007, corresponding to the daily peaks of maximum activity and maximum annual abundances periods (Díez *et al.*, 2005).

The mean number of rabbits observed on each 250 m road sector was used as surrogate of rabbit abundance.

#### Road and landscape variables

Road verges were characterized, from paved edge to fences limit, for their width and shrub species composition and cover at 50 m intervals, coinciding with small mammal trapping points. At each road side, two classes for road verge width (Verg s) were considered: up to 5 m (1) and larger than 5 m (2). Shrubs were identified to species level, whenever possible, and grouped into four dominance classes (Dshrub) regarding physiognomic characteristics of species: shrubs up to 1 m high (Sm); from 1 to 2 m high, spiny (Sp); from 1 to 2 m high, without spines and with pod (P); and from 1 to 2 m high, without spines and without pod (nP). Cover of different shrub classes was estimated on a 10 m radius circle centred on trapping points. Additionally, total length of each elm-leaf blackberry (Rubus ulmifolius) patch was measured on each sector counting both sides of the road. For analysis purposes, fenced stretches included small square mesh (<15 cm) and hexagonal wire-meshand fences and barbed wire fences were included in the classes no fences because they are not an obstacle for any of the studied species. Location and length of verge stretches without fences on each side (1 fence) and on both sides (N fence) of the road were also considered. The presence of culvert (P culv) was also registered for each sector.

Main land uses obtained through interpretation of aerial photos (year 2005) and field surveys, on 250 m buffers centred on the studied road, were: crops and meadows, olive groves, orchards and vine, dense *montado*, and sparse *montado*. *Montado* is an

open savannah like multi-use system dominated by holm and/or cork oak trees (Pinto-Correia, 1993). The number of trees bordering the road (on the closest 35 m) was also considered. As an indicator of landscape heterogeneity on road surroundings, the Patch Structural Shannon's Diversity Index (SDI) was obtained with Patch Analyst 4 for ArcGis 9. (Rempel, 2008). With this tool the edge and mean patch size of forested and open areas were also computed (Rempel, 2008). Distance from the midpoint of each sector to main water courses, reservoirs, urban areas, main national roads and motorways were derived with ArcGIS (ESRI, 1999), from land use map.

On the basis of digital cartography for 2007 (DGRF, 2007), hunting regime classes (Hunt) were established, on the same 250 m buffers used for land uses characterization considering the following groups: associative (A), areas under hunters associations management; municipal hunting (M), areas under municipal and local hunters management; and areas with no hunting regime defined (NM). These classes may be interpreted as corresponding to an increasing gradient of habitat management for prey, often associated with predator control actions.

#### Data analysis

Relationships among predator mortality and the different kinds of explanatory variables were evaluated with Generalized Linear Models (GLM), (binomial with *logit* link for carnivore, owl and snake presence and Poisson with *log* link for *species* richness) and multivariate Redundancy Analysis (RDA).

Prior to analysis, a screening for collinearity of continuous explanatory variables was done. Spearman correlation coefficients were computed for all pairs of variables and pairs showing high collinearity, with a correlation coefficient higher than 0.7 (Tabachnik and Fidell, 2001), were discarded. At the end of this process, twenty two variables were retained for following analysis. The number of trees, edge and patch size, and distances to urban areas, main national roads and motorways were correlated with other variables (e.g. land uses), thus were not analysed. Concerning the prey survey variables, the total number of small mammals was also initially considered but was not analysed, since it was highly correlated with *Mus spretus* abundance.

Table 1 resumes the response variables and the explanatory variables retained in the analyses, their description, units and transformation, when applied.

Table 1 – Preys abundance (N – number of individuals), road verges characteristics and landscape variables, their acronyms, description and other important features.

Variables set	Acronyms	Description	Type (Unit)	Transf.
	Richness	Predator road-kills species richness	Continuous (-)	-
	Carnivores	Presence of carnivores road-kills	Binary (1/0)	-
	Owls	Presence of owls road-kills	Binary (1/0)	-
	Snakes	Presence of snakes road-kills	Binary (1/0)	•
Response M_foina		Number of Martes foina road-kills	N	-
variables	H_ich	Number of Herpestes ichneumon road-kills	N	-
	Salu	Number of Strix aluco road-kills	N	-
	T_alb	Number of Tyto alba road-kills	N	-
	M_mon	Number of Malpolon monspessulanus road-kills	N	-
	E_sca	Number of Elaphe scalaris road-kills	. <b>N</b>	<b>.</b>
	Ap_syl	Number of Apodemus sylvaticus	N	-
Preys	M_spret	Number of Mus spretus	N	-
abundance	C_russ	Number of Crocidura russula	N	-
(Pr)	M_duo	Number of colonies of Microtus duodecimcostatus	N	-
	O_cun	Mean number of Oryctolagus cuniculus detected	N	<b>.</b>
	Dshrub_Sm	Dominance of shrubs up to 1m high - Cistus salviifolium, Cistus crispus	Binary (1/0)	-
Dshrub_Sh Dshrub_Sp	Dominance of shrubs 1 to 2m high, spiny - Calicotome villosa, Ulex spp., Asparagus spp.	Binary (1/0)	-	
	Dshrub P	Dominance of shrubs 1 to 2m high, no spines, with pod - Cytisus spp., Cytisus bacticus, Spartium junceum,	Binary (1/0)	-
Road and verge	-	Retama sphaerocarpa		
characteristics	Dshrub nP	Dominance of shrubs 1 to 2m high, no spines, without pod – Daphne gnidium, Cistus ladanifer	Binary (1/0)	-
(Rvc)	L_Rub	Length of elm-leaf (Rubus ulmifolius) shrubs	Continuous (Meters)	-
	P_culv	Presence of culverts	Binary (1/0)	•
	Verg_s	Road verge size	Ordinal (1/2)	-
	N_fence	Proportion of road without fences on both verges	Continuous (Proportion)	-
	1_fence	Proportion of road with fences only on one side	Continuous (Proportion)	
	Crop_m	Proportion of crops and meadows	Continuous (Proportion)	Arcsen√x
	Oliv_vin	Proportion of olive groves, orchards and vine	Continuous (Proportion)	Arcsen√x
Mon	Mont_S	Proportion of sparse montado (tree canopy cover: 10-50%)	Continuous (Proportion)	Arcsen√x
Landscape Mont D		Proportion of dense <i>montado</i> (tree canopy cover: 50-100%)	Continuous (Proportion)	Arcsen√x
(La)	Hunt _	Hunting estate – hunting regime (M – municipal; A – associative; NM – no hunting management defined)	Nominal (M/A/NM)	-
, -	D_wcou	Shortest distance to water courses	Continuous (Meters)	Log(x+1)
	D_res	Shortest distance to reservoirs	Continuous (Meters)	Log(x+1)
	SDI	Landscape Diversity Index	Continuous (-)	Log(x+1)

#### Generalized Linear Models (GLM)

Species richness and presence of total wild carnivores, owls and snakes were the response variables analysed. A Poisson regression with log link was used for species richness and Binomial regression, with logit link, was used for the other response variables.

Combinations of variables were evaluated systematically and the Akaike Information Criterion (AIC) was used to select the best models (Zuur *et al.*, 2007). Selected models were then subjected to univariate variance partitioning method (Reino *et al.*, 2006). Proportion of the explained null deviance was used as a measure of the explained variance (Zuur *et al.*, 2007) for each regression model. All calculations were done in Brodgar 2.5.1 (Highland statistics, 2006) and R 2.6.

#### Redundancy analysis (RDA)

This analysis was done with all the response variables simultaneously, in the group predators (including carnivores, owls and snakes), by redundancy analysis (RDA) (Heikkinen *et al.*, 2004). This kind of analysis was considered appropriated, since the largest value of lengths of the ordination, obtained through a preliminary Detrended Correspondence Analysis (DCA) was 3.779 s.d. (Lepš and Šmilauer, 2003).

Rare species (found in less than 10% of the forty sampling sites) were not considered for analysis in the multivariate test since they might have a confounding effect on final result (Titeux et al., 2004). This influence can be due to rare species fatalities data which can create an increase in the total inertia of the species data set or distortion of the ordination (Titeux et al., 2004).

Prior to analysis, all explanatory variables and covariables were *standardized to* unit variance, to allow comparison between variables with different units of measurement (Jongman  $et\ al.$ , 1995). The forward manual selection of explanatory variables was considered with statistical significance at the 0.05 p level.

The statistical significance of each partial RDA was tested using Monte Carlo permutation tests for the first ordination axis and all canonical axes together (999 permutations under full model).

All the computations were performed with program CANOCO for Windows, version 4.5 (ter Braak and Šmilauer, 2002).

#### Variance partitioning

Variance from prey (Pr), road and verges characteristics (Rvc) and landscape (La) models was partitioned into eight fractions: i) pure prey abundance effects; ii) pure road and verge characteristics effects; iii) pure landscape effects; and combined variation due to the joint effects of iv) prey abundance and road and verges characteristics, v) preys abundance and landscape, vi) road and verges characteristics and landscape, vii) preys abundance, road and verges characteristics and landscape, and viii) unexplained variation (Heikkinen et al., 2004). Variance partitioning was performed following the procedures proposed by (Bocard et al., 1992) adapted to three sets of variables. Computations aiming to find pure and combined proportions of explained variation were done as described in Heikkinen et al. (2004).

#### Results

#### General survey

In 2007, 86 predator road-kills, belonging to 13 wild species, were recorded (Table 2). Most road-killed animals belong to the following species: 6 beech martens (Martes foina); 5 Egyptian mongooses (Herpestes ichneumon); 19 tawny (Strix aluco), 8 barn owls (Tyto alba), 16 Montpellier snakes (Malpolon monspessulanus) and 16 ladder snakes (Elaphe scalaris).

Table 2 – Number (N) of predator road-kills. Species included in multivariate analysis are in bold.

Scientific name	English name	N
Felis silvestris*	Wild cat	1
Herpestes ichneumon	Egyptian mongoose	5
Martes foina	Beech marten	6
Meles meles	European badger	2
Mustela nivalis	Least weasel	2 2 2
Mustela putorius	European polecat	2
Vulpes vulpes		
Athene noctua	Little owl	
Strix aluco	Tawny owl	19
Tyto alba	Barn owl	8
Coluber hippocrepis Horseshoe whip snal		1
Malpolon monspessulanus	Montpellier snake	16
Elaphe scalaris	Ladder snake	

Felis silvestris\* match all the species anatomical characteristics, but was not genetically confirmed.

Regarding prey, small mammals trapped belong to 5 species (208 individuals in total): M. spretus (130 individuals), C. russula (45), A. sylvaticus (31), R. rattus (1) and R. norvegicus (1). M. duodecimcostatus was present in 31 road sectors out of a total of 40 and 109 colonies were identified. O. cuniculus was detected on 21 sectors and the mean number of counted individuals varied from zero to 5.8.

Mean values and standard errors for each explanatory variable are presented in Table 3.

Table 3 – Descriptive statistics of environmental and road variables for each surveyed road

segment (n=40). \* - Variables transformed.

Variables set	Acronyms	Minimum	Maximum	Mean	Standard Deviation
	Ap_syl	0	3	0.78	0.95
Preys	M_spret	0	9	3.25	2.22
abundance	C russ	0	5	1.125	1.32
(Pr)	M_duo	0	21	2.73	3.94
	O cun	0.00	5.80	0.68	1.39
	Dshrub Sm	0	1	-	-
	Dshrub Sp	0	1	-	-
	Dshrub P	0	1	-	-
Road and verge	Dshrub nP	0	1	-	-
characteristics	L Rub	0	293	69	95,15
(Rvc)	P_culv	0	1	-	-
, ,	Verg_s	1	2	-	-
	N fence	0.00	1.00	0.05	0,20
	1 fence	0.00	1.00	0.29	0,40
	Crop m*	0.00	1.57	0.58	0.52
	Oliv vin*	0.00	1.11	0.20	0.34
	Mont S*	0.00	1.57	0.36	0.48
Landscape	Mont D*	0.00	1.57	0.50	0.59
(La)	Hunt —	-	-	-	-
` '	D wcou*	1,32	3.15	2.82	0.31
	D res*	2.36	3.02	2.69	0.16
	SDI*	0.00	0.44	0.25	0.13

#### **GLM** results

The regression models included three variables (Ap\_syl, L\_Rub and D\_wcou) for species richness; five (O\_cun, Ap\_syl, L\_Rub, P\_culv and D\_wcou) for carnivores: four (Ap\_syl, Dshrub\_Sm, Mont\_D and SDI) for owls and one (D\_wcou) for snakes (Table 4). Prey abundance descriptors were significant, particularly in what concerns Ap\_syl, for all responses variables (species richness, carnivores and owls), except for snakes. O\_Cun was almost significant for carnivore mortality.

Only the landscape set present significant relationships with snake road mortality and so, no further variance partitioning analysis was done for this group.

Table 4 – Regression coefficients ( $\beta$ ) and significance levels (P). NS – not significant.

Group	Variable	Acronyms	Partial models		Full model	
	set		β	р	β	р
<u> </u>	Pr	Ap_syl	0.3235	0.00653*	0.1682038	NS
Richness	Rvc	L_Rub	0.001970	0.0934	0.0008139	NS
	La	D_wcou	1.9442	0.00643*	1.441373	<0.1
	Pr	O_cun	0.6266	0.0894	0.741017	<0.1
		Ap_syl	0.6860	0.0629	0.638465	NS
Carnivores	Rvc	L_Rub	0.0073	0.0516	0.008177	NS
		P_culv	-1.299x00	0.00875*	-1.878670	<0.1
	La	D_wcou	5.086	0.0340*	3.179666	NS
Owls	Pr	Ap_syl	1.2454	0.00541*	0.9783	<0.05
	Rvc	Dshrub_Sm	1.9561	0.00773*	1.3725	NS
	La	Mont_D	1.1724	0.0499*	0.8035	NS
		SDI	-8.2111	0.0110*	-5.9513	<0.1
Snakes	La	D_wcou	2.935	0.0913	2.935	<0.1

<sup>\*</sup> P< 0.05, ns=not significant

The decomposition of variation for species richness showed that the largest fractions of variability were accounted for the pure effect of landscape (8.7%), followed by the combined effect of the prey and landscape sets (7.5%). The pure effect of road and verge characteristics was the less important, explaining 0.9% only (Figure 2 - Richness).

The largest fraction of variability for carnivore presence was the pure effect of preys (11.7%) and road and verge characteristics (11.5%). The combined effects of prey and landscape sets (5.2%), landscape and road and verge characteristics (3.1%) and the combination of all variable sets (2.8%), as well as the landscape pure effect (2.4%), also explained a considerable percentage of variation (Figure 2 – Carnivores).

For owls, the largest fractions of variability were accounted by the pure effects of landscape (8.8%) and preys (8.2%). The combined effect of landscape and road and verge characteristics (4.9%) and the pure effect of the road and verge characteristics (4.5%) also present a relevant effect on explaining owls causalities (Figure 2 – Owls).

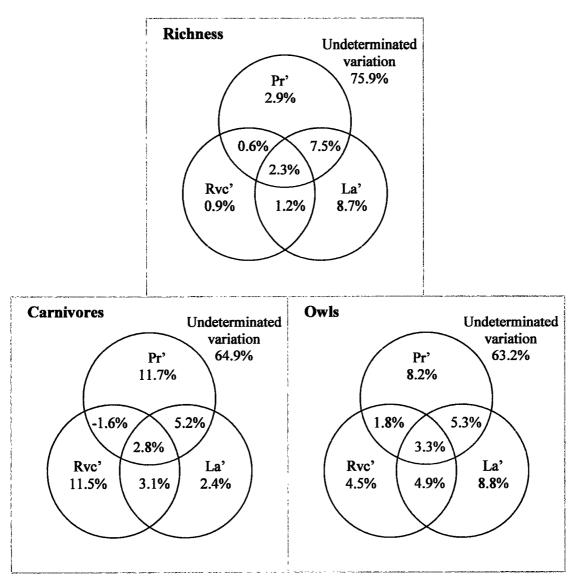


Figure 2 – Proportion of variation explained by each set of variables, distinguishing among pure and combined effects for Species Richness, Carnivores and Owls. Pr' - prey abundance; Rvc –road and verges characteristics; La' –landscape.

#### **RDA results**

Statistically significant variables included in RDA were the number of A. sylvaticus (Ap\_syl, F=3.93, p=0.002) and the mean number of O. cuniculus (O\_cun, F=4.04, p=0.016) from the prey set; small shrub dominance (Dshrub\_Sm, F=2.70, p=0.012) from the road and verge characteristics set; and landscape diversity index (SDI, F=3.39, p=0.006), distance to reservoirs (D\_res, F=3.57, p=0.002) and dense montado proportion (Mont\_D, F=5.95, p=0.001) from the landscape set.

All models performed for each partial RDA were statistically significant (p<0.01), according to Monte Carlo permutations test (Table 5). The amount of explained variance for predator full model was 45.4%.

Table 5 – Explained variance (Exp. var.) and significance of each RDA model, F - F-statistics; p - significance level.

Explanatory variables	Covariates	Values	Total
		Exp. var.	12.0
Pr	Rvc and La	F	3.629
		p	0.001
-		Exp. var.	5.4
Rvc	Pr and La	F	3.280
		p	0.008
		Exp. var.	21.0
La	Pr and Rvc	F	4.221
		p	0.001
	La	Exp. var.	17.2
Pr and Rvc		F	3.466
		p	0.001
-		Exp. var.	38.7
Pr and La	Rvc	F	4.679
		p	0.001
	Pr	Exp. var.	27.1
Rvc and La		F	4.089
		p	0.001
		Exp. var.	45.4
Pr, Rcv and La	-	F	4.567
ŕ		p	0.001

The decomposition of the variation showed that the pure effect of the landscape was a primary responsible for variability (21.0%), followed by the pure effect of preys set (12.0%). The combined effect of landscape and preys sets and the pure effect of the road and verge characteristics had very similar percentages (5.7% and 5.4%, respectively) (Figure 3).

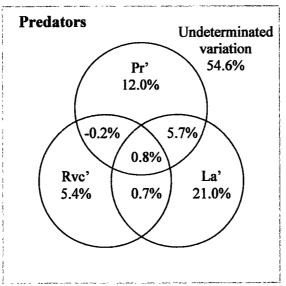


Figure 3 - Proportion of variation explained by each set of variables distinguishing among pure and combined effects. Pr' - prey abundance; Rvc -road and verges characteristics; La' - landscape.

For each species mortality, the proportion of variation explained by each set of variables is shown in Table 6. Overall, landscape set explained most of the road-kills variation (between, 1.11 and 40.81%), and the road and verges set explains the fewest (0.33 to 1.78%). However, prey abundance on verges explained most fatalities of M. monspessulanus (14.00%), Tyto alba (24.60%) and Herpestes ichneumon (20.46%).

Table 6 - Number of individuals (N) and explained variance of partials and total RDA for each predator species analysed.

N Rvc Total Groups **Species** Pr La 14.12 43.60 H. ichneumon 20.46 0.41 5 4.07 0.33 34.65 43.04 M. foina 6 S. aluco 19 2.05 1.29 13.25 38.20 **Predators** 62.06 T. alba 8 24.60 0.93 18.85 1.78 40.81 64.47 M. monspessulanus 16 14.00 1.26 1.11 14.68 E. scalaris 9.45 16

Figure 4 shows the biplots of species and environmental variables for all predators RDA. The first axis accounted for 22.1% of species variance, while the second axis explained 13.4%. The species-environmental correlations values for the first two axes were 0.813 and 0.807, respectively.

The first canonical axis was positively correlated with the variables Ap\_syl, O cun, Dshrub Sm, D res and Mont D, while a negative correlation was found for SDI. All predator species (except *H. ichneumon*) revealed a positive correlation with the first axis (Figure 4).

The second canonical axis was positively correlated with O\_cun, D\_res and SDI and negatively with Ap\_syl, Dshrub\_Sm and D\_res. Only *E. scalaris*, *M. monspessulanus* and *H. ichneumon* presented a positive relation with the second axis.

Considering the main relationships between environmental descriptors and predator mortality, positive correlations where found for Ap\_syl and Dshrub\_Sm with *M. foina* and both owl species (*S. aluco* and *T. alba*); Mont\_D areas with *S. aluco* and *E. scalaris*; and O\_cun with *H. ichneumon* and *M. monspessulanus*. On the other hand, negative correlations occurred for SDI with *M. foina*, *S. aluco* and *T. alba*, and for D\_res with *H. ichneumon* and *M. monspessulanus*.

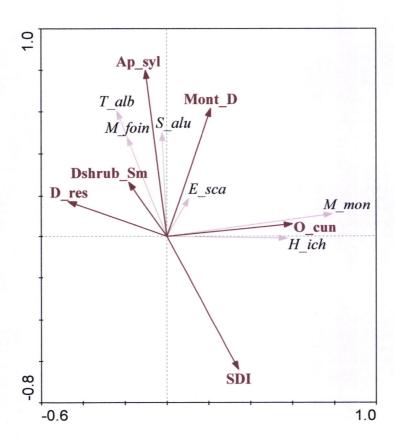


Figure 4 – Ordination biplot for the first two axes of total RDA for the all predators group. Explanatory variables are represented by red arrows and response variables by pink arrows (see Table 1 for variables acronyms).

#### **Discussion**

In the present study, most of the recorded road-killed predators belong to common species widespread in the studied area like red fox, Egyptian mongoose, Montepellier and ladder snakes, and tawny owl. Nevertheless, some threaten, declining or rare species, like the wild cat (identified on the basis of external features and waiting for genetic analysis confirmation), the European polecat and the barn owl were also found.

Considering that the was conducted on a 10 km road stretch and during a period of one year only, results suggest that road causalities may significantly contribute to threaten and decline of wildlife populations and must be seriously considered in issues concerning wildlife conservation.

The dominance of snakes in road-kills (33 casualties belonging to three species) might be related with thermoregulation, since they often use these structures as heating surface mainly in the evening (Vijayakumar et al., 2001; Shine et al., 2004). Moreover, the particular behaviour of some species, as vagility or matting migration, exposes them to an increased risk of road mortality (Jochimsen, 2006).

Concerning owls, the high mortality registered (30 casualties, belonging to three species) could be related to these birds nocturnal behaviour, since they may use roads as hunting grounds and often suffer of temporarily blindness with car lights (Massemin and Zorn, 1998; Gomes et al., 2008).

#### Determinants of predator road mortality

According to a study that took place in southern Portugal, abundance of small mammals and rabbits on verges is two to three times higher than on adjacent habitats (Ramalho, 2007). This pronounced difference suggests that road verge vegetation provides habitat for small mammals and rabbits, as observed in other regions where suitable habitat is a limiting factor for them (Bellamy et al., 2000; Bolger et al., 2001; Santos et al., 2007). Prey abundance on verges seems to attract predators, enhancing thus their mortality in traffic accidents. In fact, the abundance of preys, both small mammals and rabbits, on road verges play a significant role, not only for all predators as a group, but also for the presence of each set, excepting for snakes. Therefore, results

obtained support our initial hypotheses that prey abundance on road verges is of utmost importance in determining predator road-kills, and are in agreement with other studies (Massemin and Zorn, 1998; Bellamy et al., 2000). We must also emphasise that, among abundance of prey species analysed, A. sylvaticus was the most significant in explaining mortality of several predator species. This result is in accordance with the documented importance of this species in the diet of several predators including T. alba (Herrera and Jaksić, 1980; Cramp, 1994; Taylor, 2003) S. aluco (Kirk, 1992; Cramp, 1994), M. foina (Baghli et al., 2002; Posluszny et al., 2007), H. ichneumon (Palomares and Delibes, 1991a; Palomares and Delibes 1991b), E. scalaris (Pleguezuelos, 2006; Pleguezuelos et al., 2007) and M. monspessulanus (Pleguezuelos, 2003). On the other hand, abundance of O. cuniculus, a key prey in Mediterranean ecosystems (Delibes-Mateos et al., 2007), is highly associated with H. ichneumon and M. monspessulanus fatalities, probably reflecting the importance of this species in the diet of both predators (Palomares and Delibes, 1991a; Palomares and Delibes 1991b; Pleguezuelos, 2003; personal observations). Although the attraction of predators for areas with high prey abundance might be mainly related with the privileged food source, we may hypothesised that some species, as snakes, can also be attracted by small mammal burrows which they use as hibernacula (Burger et al., 1988). However, fossorial vole abundance did not show any significant relation with predator mortality, giving little support to this hypothesis.

In general, landscape characteristics explained the highest percentage of variance on predator road-kills, which is in agreement with other studies (Clevenger et al, 2003; Ascensão and Mira, 2006a; Gomes et al., 2008). This must reflect the relation between landscape variables and the abundance and distribution of the predators in the area adjacent to the road (Geneletti, 2003), being road-kills most likely to occur where roads run through or between preferred habitats by the species (Seiler, 2003).

We found higher predator road-kills on dense *montado*, than on sparse *montado*, policultures and open areas, although this relation was more significant for owls. Ascensão and Mira (2006a) also point out high vertebrate road-kills on areas where *montado* is predominant. Considering that *S. aluco* (Cramp, 1994; Massemin and Zorn, 1998) and *M. foina* (Virgós and Garcia, 2002; Ruiz-González *et al.*, 2008) are more common on wooded areas, higher abundance on *montado* and consequently higher fatalities in this habitat should be expected for both species. *E. scalaris* and *M. monspessulanus*, although hunting in open areas such as verges, also use wooded areas for shelter (Pleguezuelos, 2003; Pleguezuelos, 2006). Our results also suggest a

moderate positive influence of dense *montado* cover for *T. alba* road-kills, in contradiction with Gomes *et al* (2008), which report more road-kills of this species in association with open areas than with *montado*. This apparent contradiction may reflect the eclectic behaviour of *T. alba*, concerning habitat use (Cramp, 1994) and road verges are good hunting areas (Massemin and Zorn, 1998), since they usually hunt with low flights on open areas (Cramp, 1994).

Besides the proportion of *montado* areas, landscape heterogeneity (SDI) also affects predator road casualties, being particularly important for owls. In general higher SDI translates into lower predator mortality. In our study site, SDI can be interpreted as a measure of habitat fragmentation due to human intervention, mainly agriculture intensification. Hence, the lower mortality observed may reflect the lower abundance of wild predators on fragmented landscapes reported by Pita *et al.* (in prep.).

The positive relation observed between distance to water reservoirs and *H. ichneumon* and *M. monspessulanus* mortality is in agreement with results of studies on *T. alba* and *S. aluco* (Gomes *et al.*, 2008) and might be due to water availability and to increased food resources in their surroundings. However, as a negative relation was determined for all the other studied species, further radio-tracking on predators and studies on prey availability close to water reservoirs should be made.

The shortest distance to water courses was positively correlated with all the univariate road-kill models, except for owls. Since most close water courses run underneath the road, crossing it through culverts or small bridges, they may be used as corridors by ground dwelling species, like snakes and carnivores, reducing road-kill probabilities. In fact, a significant decrease in carnivore road-kills was observed in road sectors provided with culverts. Moreover, in the study site and its surroundings there is a strong evidence of widespread use of culverts by several carnivore species (Ascensão and Mira, 2006b; Varela, 2007; Grilo *et al.*, 2008).

The road and verges characteristics may attract or repel animals, changing their probability of being road-killed (Forman and Alexander, 1998). However, in contrast with landscape, our study shows that road and verges features usually have a weak overall contribution to explain predator mortality. This must partially reflect the small variability in most of the studied road and verge descriptors, due to the road stretch dimension. Nevertheless, the presence and extend of *R. ulmifolius* along verges significantly increases carnivore fatalities. Also worth to mention that areas dominated by small shrubs are associated with higher owl and *M. foina* road-kills. Concerning

mammals, Clevenger et al., (2001) had already shown that road mortality is enhanced in shrubby areas. The same authors and Seiler (2003) suggested that the presence of shrubs on verges might increase mammal road-kills because they provide refuge, leading thus animals approach to road. Similarly, *S. aluco* road-kills also increased with the presence of shrubs on verges, due to its hunting behaviour between shrub interspaces (Cramp, 1994).

Unexplained variation was significant for all models, achieving a minimum of 54.6% for the predator model. The relatively low percentage of explained variation is not uncommon in ecological studies because species abundance or occurrence data are often noisy (Lepš, and Šmilauer, 2003) and could be explained by several other factors. This variation can be attributed to stochastic space-time fluctuations of the communities and unmeasured environmental variables or spatially-structuring processes that have been missed (Titeux *et al.*, 2004).

#### Mitigation measures

Overall the results show that prey availability on roads is a significant factor affecting predator road-kills, only overtaken by the influence of landscape features. These results have strong implications for the implementation of predator road-kill mitigation measures. Belammy et al (2000) and Ramalho (2007), suggest that verges provide better shelter and food for small mammals and rabbits, increasing their abundance, particularly in regions where roads cross intensive agricultural management. If this is so, soil mowing and vegetation cutting, particularly small shrubs and R. ulmifolius, resources on which some prey species seem to depend on (e.g. A. sylvaticus), should allow for a decrease in predator mortality. Besides, shrub cutting will also prevent the use of roadside as refuge and allows for better awareness of the road presence by ground predators like snakes and carnivores. Another option would be to promote areas of suitable habitat for preys (shrubby and grassy areas) in patches distant to roads, particularly when these cross intensive agricultural or grazed land as is the case of the studied road. As a result, patches with high food resources for prey and predator specie could be maintained. Other important measures would be the construction of culverts that would promote crossing passages to carnivores, diminishing the road impact as a wildlife barrier.

However, preventive measures are always the best management practices. Before building roads, environmental impact assessments should consider not only species present in the area, but also the landscape and its ecological dynamics. Our study have shown, for the first time, that besides landscape features, the dynamics of predator-prey relationships must be taken into account when planning mitigation actions for new or existing roads. In fact, only an assessment of all these features, taken together, will improve our knowledge of the problem and will enhance the efficacy of mitigation measures aiming to reduce predator road-kills.

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#### References

- Ascensão, F. and Mira, A. 2006a. Spatial patterns of road kills: a case study in Southern Portugal. IN: Proceedings of the 2005 International Conference on Ecology and Transportation, Eds. Irwin CL, Garrett P, McDermott KP. Center for Transportation and the Environment, North Carolina State University, Raleigh, NC: pp. 641-646.
- Ascensão, F. and Mira, A. 2006b. Factors affecting culvert use by vertebrates along two stretches of road in southern Portugal. *Ecol Res*, **22(1)**: 57-66.
- Baghli, A., Engel, E., Luxembourg and Verhagen, A. R. 2002. Feeding habits and trophic niche overlap of two sympatric Mustelidae, the polecat *Mustela putorius* and the beech marten *Martes foina*. *Z. Jagdwiss*. **48:** 217-225.
- Bellamy, P. E., Shore, R. F., Ardeshir, D., Treweek, J. R. and Sparks, T. H. 2000. Roads verges as habitats for small mammals in Britain. *Mammal Review.* 30:2, 131-130
- Bocard, D., Legendre, P. and Drapeu, P. 1992. Partialling out the spatial components of ecological variation. *Ecology* 73(3): 1045-1055.

- Bolger, D. T., Scott, T. A. and Rotenberry, J. T. 2001. Use of corridor-like landscape structures by bird and small mammal species. *Conservation Biology* **102**: 213-224.
- Burger, J., Zappalorti, R. T., Gochfeld, M., Boarman, W., Caffrey, M., Doig, V., Garber, S., Mikovsky, M., Safina, C. and Saliva, J. 1988. Hibernacula and summer dens of pine snakes (*Pituophus melanoleucus*) in the New Jersey pine barrens. *J Herpetol* 22:425–433.
- ter Braak, C and Šmilauer, P. 2002. Canoco for Windows version 4.5. Biometrics Plant Research International, Netherlands.
- Clevenger, A. P., Chruszcz, B. and Gunson, K. E. 2001. Drainage culverts as habitat linkages and factors affecting passage by mammals. *Journal of Applied Ecology* **38**: 1340–1349.
- Clevenger, A. P., Chruszcz, B. and Gunson, K. E. 2003. Spatial patterns and factors influencing small vertebrate fauna road-kill aggregations. *Biological Conservation*, 109: 15-26.
- Cramp, S. (Ed.) 1994. Handbook of the Birds of Europe, the Middle East and North Africa Volume IV: Terns to Woodpeckers. Oxford University Press, USA.
- Delibes-Mateos, M., Redpath, S. M., Ângulo, E., Ferreras, P. and Villafuerte, R. 2007. Rabbits as a keystone species in southern Europe. *Biological Conservation* 137: 149-156.
- DGRF 2007. Cartografia digital das áreas de caça. Autoridade Regional de Florestas.
- Díez, C., Pérez, J. A., Prieto, R., Alonso, M. E. and Olmedo, J. A. 2005. Activity patterns of wild rabbit (*Oryctolagus cuniculus* L.1785) under semi-freedom conditions during autumn and winter. *Wildl. Biol. Pract.*, 1 (1): 41-46.
- EP 2005. Recenseamento de Tráfego Évora. Estradas de Portugal, E.P.E.
- ESRI 2004. ArcGIS 9. Environmental Systems Research Institute, Inc., New York.
- Ferreras, P., Aldama, J. J., Beltrdn, J. F. and Delibes, M. 1992. Rates and causes of mortality in a fragmented population of Iberian lynx *Felis pardina* Temminck, 1824. *Biological Conservation* 61: 197-202.
- Forman, R. T. T. 2000. Estimate of the area affected ecologically by the road system in the United Status. *Conservation Biology*, **1(14)**: 31-35.
- Forman, R. T. T. and Alexander, L. E. 1998. Roads and their ecological effects. *Annu. Rev. Ecol. Syst*, 29: 207-231.
- Forman, R. T. T., Sperling, D., Bissonette, J. A., Clevenger, A. P., Cutshall, C. D., Dale, V. H., Fahrig, L., France, R., Goldman, C. R., Heanue, K., Jones, J. A., Swanson, F. J., Turrentine, T. and Winter, T. C. 2003. *Road Ecology: Science and Solutions*. Island Press, USA.
- Geneletti, D. 2003. Biodiversity Impact Assessment of roads: an approach based on ecosystem rarity. *Environmental Impact Assessment Review* 23: 343–365.
- Gomes, L., Grilo, C., Silva, C. and Mira, A. 2008. Identification methods and deterministic factors of owl roadkill hotspot locations in Mediterranean landscapes. *Ecol. Res.*
- Grilo, C., Bissonette, J. A. and Santos-Reis, M. 2008. Response of carnivores to existing highway culverts and underpasses: implications for road planning and mitigation. *Biodivers Conserv* 17:1685–1699.
- Heikkinen, R. K., Luoto, M., Virkkala, R. and Rainio, K. 2004. Effects of habitat cover, landscape structure and spatial variables on the abundance of birds in an agricultural-forest mosaic. *Journal of Applied Ecology* 41: 824-835.
- Herrera, C. M. and Jaksić, F. M. 1980. Feeding ecology of the Barn owl in Central Chile and Southern Spain: a comparative study. *The Auk* 97: 760-767.

- Highland statistics 2006. Brodgar 2.5.1. Highland statistics Ltd, UK.
- Hovd, H. and Skogen, A. 2005. Plant species in arable field margins and road verges in central Norway. *Agriculture Ecosystems and Environment* 110: 257-265.
- Instituto do Ambiente, 2007. Available at: http://www.iambiente.pt.
- Iull, B., Bekker, G. J., Cuperus, R., Dufek, J., Fry, G., Hicks, C., Hlaváč, V., Keller, V., Rosell, C., Sangwine, T., Tørsløv, N., Wandall, B. le Maire (eds.) 2003.
  Wildlife and Traffic: A European Handbook for identifying Conflicts and Designing Solutions. COST.
- Jochimsen, D. M. 2006. factors influencing the road mortality of snakes on the Upper Snake River Plain, Idaho. IN: Irwin CL, Garrett P, McDermott KP. Center for Transportation and the Environment, North Carolina State University, Raleigh, NC: pp. 641-646.
- Jongman R.H.G., ter Braak C.J.F. and van Tongeren O.F.R. 1995. *Data analysis in community and landscape ecology*. Cambridge University Press, Cambridge.
- Kirk, D. A. 1992. Diet changes in breeding Tawny Owls (Strix aluco). J. Raptor Res. 26(4): 239-242.
- Lepš, J. and Šmilauer, P. 2003. *Multivariate Analysis of Ecological Data using CANOCO*. Cambridge University Press, Cambridge.
- Massemin, T. and Zorn, T. 1998. Highway mortality of barn owls in Northeastern France. J. Raptor Res. 32(3): 229-232.
- Mira, A. P. P. 1999. Ecologia do rato-cego-mediterrâneo, Microtus (Terricola) duodecimcostatus (Rodentia, Muridae), em pomares de citrinos Bases a para gestão das populações. Doctoral thesis. Faculdade de Ciências da Universidade de Lisboa. Lisboa.
- Palomares, F. and Delibles, M. 1991a. Alimentacion del meloncillo *Herpestes ichneumon* y de la gineta *Genetta genetta* en la Reserva Biologica de Doñana, S. O. de la Peninsula Iberica. *Acta Vertebrada* 18(1): 5-20.
- Palomares, F. and Delibles, M. 1991b. Dieta del meloncillo *Herpestes ichneumon*, na el Coto del Rey (Norte del Parque Nacional de Doñana, S. O. de España). *Acta Vertebrada* 18(2): 187-194.
- Pinto-Correia, T., 1993. Threatened landscape in Alentejo, Portugal: The Montado and other agro-silvo pastoral systems. *Landscape and Urban Planning* **24**: 43-48.
- Pleguezuelos, J. M. (2003). Culebra bastarda *Malpolon monspessulanus*. En: Enciclopedia Virtual de los Vertebrados Españoles. Carrascal, L. M., Salvador, A. (Eds.). Museo Nacional de Ciencias Naturales, Madrid. Available at: http://www.vertebradosibericos.org/
- Pleguezuelos, J. M. (2006). Culebra de escalera Rhinechis scalaris. *In*: Enciclopedia Virtual de los Vertebrados Españoles. Carrascal, L. M., Salvador, A. (Eds.). Museo Nacional de Ciencias Naturales, Madrid. Available at: http://www.vertebradosibericos.org/
- Pleguezuelos, J. M., Fernández-Cardenete, J. R., Honrubia, S., Feriche, M., Villafranca C. 2007. Correlates between morphology, diet and foraging mode in the Ladder Snake *Rhinechis scalaris* (Schinz, 1822). *Contributions to Zoology*, 76(3): 179-186.
- Posluszny, M., Pilot, M., Goszczyński, J. and Gralak, B. 2007. Diet of sympatric pine marten (*Martes martes*) and stone marten (*Martes foina*) identified by genotyping of DNA from faeces. *Ann. Zool. Fennici* 44: 269-284.
- Quintana-Ascencio, P. F., Weekley, C. W. and Menges, E. S. 2007. Comparative demography of a rare species in Florida scrub and road habitats. *Biological Conservation* 137: 263-270.

- Ramalho, P. A. O. R. 2007. As bermas de estrada como refúgio para os micromamíferos em Monfurado. Degree of licentiate thesis. Universidade de Évora, Évora.
- Reino, L.; P. Beja and A.C. Heitor. 2006. Modelling spatial and environmental effects at the edge of the distribution: the red-backed shrike *Lanius collurio* in Northern Portugal. *Biodiversity and Distributions*, 12: 379-387.
- Rempel, R. 2008. Patch Analyst 4 for ArcGIS. Centre for Northern Forest Ecosystem Research (Ontario Ministry of Natural Resources), Ontario. Available at: http://flash.lakeheadu.ca/~rrempel/patch/index.html.
- Rivas-Martínez, S. and Arregui, J. L. 1999. Bioclimatology of the Iberian Península. *Itinera Geobotanica*, 13: 41-47.
- Ruiz-González, A., Rubines, J., Berdión, O. and Gómez-Moliner, B. J. 2008. A non-invasive genetic method to identify the sympatric mustelids pine marten (*Martes martes*) and stone marten (*Martes foina*): preliminary distribution survey on the northern Iberian Peninsula. Eur J Wildl Res 54:253-261.
- Santos, S. M., Mathias, M. L., Mira, A. and Simões, M. P. 2007 Vegetation structure and composition of road verge and meadow sites colonized by Cabrera vole (*Microtus cabrerae* Thomas). *Pol. J. Ecol.* 55(3): 481–493.
- Seiler, A. 2003 The toll of the automobile: Wildlife and roads in Sweden. Doctoral thesis. Swedish University of Agriculture Sciences. Uppsala.
- Shine, R., Lemaster, M., Wall, M., Langkilde, T and Mason, R. 2004. Why Did the Snake Cross the Road? Effects of Roads on Movement and Location of Mates by Garter Snakes (*Thamnophis sirtalis parietalis*). Ecology and Society 9(1):1-9. Available in: http://www.ecologyandsociety.org/vol9/iss1/art9
- SNIRH, 2007. Disponível em: http://snirh.pt
- Spooner, P. G., Lunt, I. D., Briggs, S. V. and Freudenberger, D. 2004. Effects of soil disturbance from roadworks on roadside shrubs in a fragmented agricultural landscape. *Biological Conservation* 117: 393-406.
- SPSS 2007. SPSS 16.0 for Windows. SPSS Inc., Chicago.
- Tabachnick, B.G. and Fidell, L.S., 2001. *Using Multivariate Statistics*. Allyn and Bacon, Boston.
- Taylor, I. 2003. Barn owls: Predator-prey relationships and conservation. Cambridge University Press. Cambridge.
- Titeux N., Dufrêne M., Jacob J.P., Paquay M. and Defourny P. 2004. Multivariate analysis of a fine-scale breeding bird atlas using a geographical information system and partial canonical correspondence analysis: environmental and spatial effects. *Journal of Biogeography* 31: 1841-1856.
- Truscott, A. M., Palmer, S. C. F., McGowan, G. M., Cape, J. N. and Smart, S. 2005. Vegetation composition of roadsides verges in Scotland: the effects of nitrogen deposition, disturbance and management. *Environmental Pollution* 136: 109-118.
- Varela, N. A. S. 2007. Uso de passagens hidráulicas por carnívoros em estradas do Alentejo. Degree of licentiate thesis. Universidade de Évora, Évora.
- Vijayakumar, S. P., Vasudevan, K. and Ishwar, N. M. 2001. Herpetofaunal mortality on roads in the Anamalai Hills, Southern Western Ghats. *Hamadryad* 26: 265-272.
- Virgós, E. and Garcia, F. J. 2002. Patch occupancy by stone martens *Martes foina* in fragmented landscapes of central Spain: the role of fragment size, isolation and habitat structure. *Acta Oecologica* 23: 231–237.

- Wilson, D. E., Cole, F. R., Nichols, J. D., Rudran, R. and Foster, M. S. (eds.) 1996.

  Measuring and Monitoring Biological Diversity standard methods for mammals. Smithsonian Institution Press, Washington and London.
- Zuur, A. F., Ieno, E. N. and Smith, G. M. 2007. Analysing Ecological Data. Springer. US.

#### Considerações finais

A abundância de micromamíferos e coelhos nas bermas da estrada influenciam a mortalidade de predadores por atropelamento. A abundância de *A. sylvaticus* (excepto no modelo das serpentes) e de *O. cuniculus* (apenas nos modelos de predadores e carnívoros), foram as mais correlacionadas com a mortalidade por atropelamento de predadores e, de facto, estas espécies fazem parte da dieta da maioria das espécies estudadas. Como tal, a sua presença parece aumentar a utilização das bermas por parte do grupo dos predadores e, consequentemente, a sua mortalidade.

A. sylvaticus é considerado uma parte importante da dieta de T. alba (Herrera and Jaksić, 1980; Cramp, 1994; Taylor, 2003), S. aluco (Kirk, 1992; Cramp, 1994), M. foina (Baghli et al., 2002; Posluszny et al., 2007), H. ichneumon (Palomares and Delibes, 1991a; Palomares and Delibes 1991b), E. scalaris (Pleguezuelos, 2006; Pleguezuelos et al., 2007) e M. monspessulanus (Pleguezuelos, 2003). Relativamente à espécie O. cuniculus, esta é uma parte significativa da dieta de H. ichneumon (Palomares and Delibes, 1991a; Palomares and Delibes 1991b) e M. monspessulanus (Pleguezuelos, 2003).

O grupo de variáveis que mais variância na mortalidade por atropelamento de predadores explicou, excepto para os carnívoros, foi a paisagem. Estas variáveis estão relacionadas com a presença e distribuição das espécies na zona adjacente à estrada (Geneletti, 2003). As variáveis distância mais curta a albufeiras (apenas no modelo dos predadores), proporção de áreas densas de montado (entre 70 a 100%; excepto apenas nos modelos dos predadores e rapinas nocturnas), o Índice de Diversidade de paisagem foram as variáveis significativas (apenas nos modelos dos predadores e rapinas nocturnas) e distância mais curta a linhas de água (excepto nos modelos de predadores e rapinas nocturnas).

As características da estrada e da berma podem atrair ou repelir a fauna, alterando a probabilidade de atropelamento das espécies (Forman and Alexander, 1998). Em contraste com a paisagem, este grupo de variáveis foi o que menos explicou a mortalidade por atropelamento, excepto no caso dos carnívoros. As variáveis consideradas mais relevantes foram dominância de arbustos baixos (modelos dos predadores e rapinas nocturnas), o comprimento de silvas (modelo da riqueza especifica e carnívoros) e presença de passagens hidráulicas (modelo dos carnívoros).

As medidas de mitigação para estas espécies de predadores devem passar pela correcta gestão das estradas e bermas. Sugere-se a redução das áreas de vegetação nas bermas, para diminuir a abundância de micromamíferos e coelhos, mantendo zonas com reduzido pastoreio longe da estrada. A construção de passagens hidráulicas adequadas à passagem de carnívoros é uma medida importante para reduzir a mortalidade por atropelamento.

#### Referências bibliográficas

- (Inclui apenas as referências bibliográficas citadas na introdução, área de estudo e conclusões).
- Baghli, A., Engel, E., Luxembourg and Verhagen, A. R. 2002. Feeding habits and trophic niche overlap of two sympatric Mustelidae, the polecat *Mustela putorius* and the beech marten *Martes foina*. *Z. Jagdwiss*. **48:** 217-225.
- Bellamy, P. E., Shore, R. F., Ardeshir, D., Treweek, J. R. and Sparks, T. H. 2000. Roads verges as habitats for small mammals in Britain. *Mammal Review*. **30**:2, 131-139.
- Bolger, D. T., Scott, T. A. and Rotenberry, J. T. 2001. Use of corridor-like landscape structures by bird and small mammal species. *Conservation Biology* **102**: 213-224.
- Câmara Municipal de Montemor-o-Novo, 2007. Disponível em: http://www.cm-montemornovo.pt.
- Clevenger, A. P., Chruszcz, B. and Gunson, K. E. 2003. Spatial patterns and factors influencing small vertebrate fauna road-kill aggregations. *Biological Conservation*, 109: 15-26.
- Coffin, A. W. in press. From road kill to road ecology: a review of the ecological effects of roads. Journal of Transport Geography.
- Costa, J. C., Aguiar, C., Capelo, J. H., Lousã, M. and Neto, C. 1999. Biogeografia de Portugal Continental. *Quercetea*, **0**: 5-56.
- Cramp, S. (ed.) 1994. Handbook of the Birds of Europe, the Middle East and North Africa Volume IV: Terns to Woodpeckers. Oxford University Press, USA.
- EP, 2005. Recenseamento de Tráfego Évora. Estradas de Portugal, E.P.E..
- Ferreras, P., Aldama, J. J., Beltrdn, J. F. and Delibes, M. 1992. Rates and causes of mortality in a fragmented population of Iberian lynx *Felis pardina* Temminck, 1824. *Biological Conservation* 61: 197-202.
- Forman, R. T. T. 2000. Estimate of the area affected ecologically by the road system in the United Status. *Conservation Biology*, **1(14)**: 31-35.
- Forman, R. T. T. and Alexander, L. E. 1998. Roads and their ecological effects. Annu. Rev. Ecol. Syst, 29: 207-231.
- Forman, R. T. T., Sperling, D., Bissonette, J. A., Clevenger, A. P., Cutshall, C. D., Dale, V. H., Fahrig, L., France, R., Goldman, C. R., Heanue, K., Jones, J. A., Swanson, F. J., Turrentine, T. and Winter, T. C. 2003. *Road Ecology: Science and Solutions*. Island Press, USA.
- Geneletti, D. 2003. Biodiversity Impact Assessment of roads: an approach based on ecosystem rarity. *Environmental Impact Assessment Review* 23: 343–365.
- Herrera, C. M. and Jaksić, F. M. 1980. Feeding ecology of the Barn owl in Central Chile and Southern Spain: a comparative study. *The Auk* 97: 760-767.
- Hovd, H. and Skogen, A. 2005. Plant species in arable field margins and road verges in central Norway. *Agriculture Ecosystems and Environment* 110: 257-265.
- INE, 2001. Recenseamento Geral da População e Habitação Resultados Definitivos. Disponível em: www.ine.pt.
- Iull, B., Bekker, G. J., Cuperus, R., Dufek, J., Fry, G., Hicks, C., Hlaváč, V., Keller, V., Rosell, C., Sangwine, T., Tørsløv, N., Wandall, B. le Maire (Eds) 2003.
  Wildlife and Traffic: A European Handbook for identifying Conflicts and Designing Solutions. COST.
- Kirk, D. A. 1992. Diet changes in breeding Tawny Owls (Strix aluco). J. Raptor Res. 26(4): 239-242.

- Palomares, F. and Delibles, M. 1991a. Alimentacion del meloncillo *Herpestes ichneumon* y de la gineta *Genetta genetta* en la Reserva Biologica de Doñana, S. O. de la Peninsula Iberica. *Acta Vertebrada* 18(1): 5-20.
- Palomares, F. and Delibles, M. 1991b. Dieta del meloncillo *Herpestes ichneumon*, na el Coto del Rey (Norte del Parque Nacional de Doñana, S. O. de España). *Acta Vertebrada* 18(2): 187-194.
- Pleguezuelos, J. M. (2003). Culebra bastarda *Malpolon monspessulanus*. En: Enciclopedia Virtual de los Vertebrados Españoles. Carrascal, L. M., Salvador, A. (Eds.). Museo Nacional de Ciencias Naturales, Madrid. Disponível em: http://www.vertebradosibericos.org/
- Pleguezuelos, J. M. (2006). Culebra de escalera *Rhinechis scalaris*. *In*: Enciclopedia Virtual de los Vertebrados Españoles. Carrascal, L. M., Salvador, A. (Eds.). Museo Nacional de Ciencias Naturales, Madrid. Disponível em: http://www.vertebradosibericos.org/
- Pleguezuelos, J. M., Fernández-Cardenete, J. R., Honrubia, S., Feriche, M., Villafranca C. 2007. Correlates between morphology, diet and foraging mode in the Ladder Snake *Rhinechis scalaris* (Schinz, 1822). *Contributions to Zoology*, 76(3): 179-186.
- Posluszny, M., Pilot, M., Goszczyński, J. and Gralak, B. 2007. Diet of sympatric pine marten (*Martes martes*) and stone marten (*Martes foina*) identified by genotyping of DNA from faeces. *Ann. Zool. Fennici* 44: 269-284.
- Quintana-Ascencio, P. F., Weekley, C. W. and Menges, E. S. 2007. Comparative demography of a rare species in Florida scrub and road habitats. *Biological Conservation* 137: 263-270.
- Ramalho, P. A. O. R. 2007. As bermas de estrada como refúgio para os micromamíferos em Monfurado. Degree of licentiate thesis. Universidade de Évora, Évora.
- Rivas-Martínez, S. and Arregui, J. L. 1999. Bioclimatology of the Iberian Península. *Itinera Geobotanica*, 13: 41-47.
- Santos, S. M., Mathias, M. L., Mira, A. and Simões, M. P. 2007 Vegetation structure and composition of road verge and meadow sites colonized by Cabrera vole (*Microtus cabrerae* Thomas). *Pol. J. Ecol.* 55(3): 481–493.
- Seiler, A. 2003 The toll of the automobile: Wildlife and roads in Sweden. Doctoral thesis. Swedish University of Agriculture Sciences. Uppsala.
- SNIRH, 2007. Disponível em: http://snirh.pt.
- Spooner, P. G., Lunt, I. D., Briggs, S. V. and Freudenberger, D. 2004. Effects of soil disturbance from roadworks on roadside shrubs in a fragmented agricultural landscape. *Biological Conservation* 117: 393-406.
- Taylor, I. 2003. Barn owls: Predator-prey relationships and conservation. Cambridge University Press. Cambridge.
- Truscott, A. M., Palmer, S. C. F., McGowan, G. M., Cape, J. N. and Smart, S. 2005. Vegetation composition of roadsides verges in Scotland: the effects of nitrogen deposition, disturbance and management. *Environmental Pollution* 136: 109-118.