

## **ROAD ECOLOGY OF THE NORTHERN DIAMONDBACK TERRAPIN, *MALACLEMYS TERRAPIN TERRAPIN***

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**Abstract:** Diamondback terrapin populations along the East Coast have suffered due to a number of factors since the early 1900's. Overexploitation from commercial harvesting, drowning in fishing gear, and loss of habitat has had a negative impact on the terrapin (Roosenburg 1991). Terrapins in several areas, specifically in New Jersey, are now threatened by an additional source of mortality, road mortality (Wood and Herlands 1997, Hoden and Able 2003), which could cause further declines in the abundance of this species.

Road mortality and ecology of the northern diamondback terrapin, *Malaclemys terrapin terrapin*, in the Jacques Cousteau National Estuarine Research Reserve was examined and compared to traffic patterns during the nesting seasons (May-July) of 2004 and 2005. Traffic-measuring devices were stationed on sections of Great Bay Boulevard (GBB), an access road through salt-marsh habitat to obtain traffic-volume estimates. A total of 1201 terrapins were observed on the road with 104 road mortalities (8.66%). In 2004, a significantly greater proportion of road kills was found in the section of the road with the highest traffic volume.

However, we did not see this same pattern in 2005 as road mortalities across the sections were fairly evenly distributed. There was a positive correlation between road kills and increasing traffic volume throughout the day observed in 2004. Three hundred terrapins were tagged with passive-integrated transponder (PIT) tags over the course of the study. The tagging portion of this study indicated that some females may have been returning more than once in the season to lay multiple clutches along the roadside and demonstrated nest-site philopatry by returning to the area where they were initially tagged.

The information gathered suggests that terrapins are attracted to the roadside as it meets the requirements for a suitable nesting habitat. Future mitigation, such as drift fencing and increased patrolling of the roads, is needed to help reduce road mortalities. Fencing will be proposed to be installed in the areas of greatest road mortality and of greatest nesting activity along Great Bay Boulevard for 2006.

### **Introduction**

Road mortality is becoming a significant problem for the northern diamondback terrapin, *Malaclemys terrapin terrapin*. Since terrapins are a "Species of Special Concern" in New Jersey (N.J. Department of Environmental Protection, Division of Fish and Wildlife, N.J. Endangered and Non-Game Species Program website 2005), understanding the impact of road mortality is critical (Forman et al. 2003). Great Bay Boulevard (GBB), an 8.1-km paved access road running through fairly pristine salt-marsh habitat in the Jacques Cousteau National Estuarine Research Reserve (JCNERR), is the site of many deaths each summer. Since suitable nesting habitat exists along the side of the boulevard, adult female terrapins are at risk from collisions with vehicles. Accordingly, this detailed survey was conducted to evaluate the relationship between traffic, road occurrence, and road mortality along this road for 2004/2005.

The field work was conducted at Rutgers University Marine Field Station, Institute for Marine and Coastal Sciences, Tuckerton, New Jersey.

### **Methodology**

Surveys of Great Bay Boulevard in the JCNERR were conducted during the terrapin-nesting season from May-July of 2004/2005. Transect sections were chosen based on bridges that divide the road and cross over the subtidal marsh creeks (figures 1 and 2). Approximately five-six days per week, eight to 10 surveys were completed each day for a total of 299 samples in 2004 and 272 samples in 2005.

TRAX I Plus Traffic Counters/Classifiers (Jamar Technologies Inc.) were stationed in the middle of the six transect sections of the road to measure traffic patterns.

GPS data points were recorded for all terrapins and plotted on aerial images using ArcView GIS to obtain distances to nearest major and extension creeks and bridges. Comparisons of road mortality between sections and were tested using Pearson Chi-Square Analysis. Spearman's Rank Order Correlation was used to test mortality rate and mean traffic volume during the hours of the day. Three hundred terrapins were tagged with 2 x 12-mm passive-integrated transponder (PIT) tags (Biomark Inc).



Figure 1. Map of the Jacques Cousteau National Estuarine Research Reserve in New Jersey, where Great Bay Boulevard is located.



Figure 2. Aerial image of Great Bay Boulevard with defined transect sections within the Great Bay Wildlife Management Area of the Jacques Cousteau National Estuarine Research Reserve.

## Results

- Numerous terrapins were observed each nesting season (N, 2004 = 601 and N, 2005 = 600) with peaks of nesting along the road around the lunar phases.
- Road mortality in 2004 was found to be significantly greater ( $p < 0.001$ ) in section 6 and exhibited the highest traffic volume. In 2005, no differences were found between sections, except that section 2 had significantly less mortality ( $p < 0.005$ ) (table 1).
- Most road-killed terrapins in section six were killed near creeks and bridges that intersect Great Bay Boulevard (table 2, figure 3).
- Road-mortality rates correlated positively with average traffic volume by hour in 2004 during our survey times of 0900-1600 ( $p < 0.03$ ).
- Sixty five of 300 (21.67%) tagged terrapins were recaptured. One female crossed GBB a minimum of 5 times in the 2004 season.
- Some demonstrated possible multiple clutching and nest-site philopatry within and among years.

Table 1. Summary results of terrapin occurrences, mortalities, and average traffic volume (vehicles/day) by transect section on Great Bay Boulevard during the nesting seasons of 2004 and 2005

Transect Section	Live (2004)	Dead (2004)	Mean Traffic Volume (2004)	Live (2005)	Dead (2005)	Mean Traffic Volume (2005)
1	57	2 (3.39%)	233.70	83	4 (4.60%)	250.38
2	101	9 (8.18%)	262.71	96	<b>1 (1.03%)*</b>	337.71
3	189	13 (6.44%)	363.36 <sup>a</sup>	153	20 (11.56%)	621.39
4	84	8 (8.70%)	363.36 <sup>a</sup>	91	13 (12.5%)	707.22
5	91	10 (9.90%)	464.00	84	7 (7.69%)	749.41
6	25	<b>11 (30.56%)**</b>	936.00	42	6 (12.5%)	905.97
Total	547	53 (8.83%)		549	51 (8.5%)	

\*\* ( $P < 0.001$ ,  $\chi^2 = 22.44$ ,  $df = 1$ )  
 \* ( $P < 0.005$ ,  $\chi^2 = 8.299$ ,  $df = 1$ )  
<sup>a</sup>Estimated traffic volumes.

Table 2. Summary results of terrapin road mortalities in relation to nearest creeks and a bridge along transect section 6 of Great Bay Boulevard

	Mean	Range	N
Straight-line distance (m) to nearest creek (major or extension) from:			
2004 road mortalities <sup>a</sup>	69.87	14.74-334.89	11
2005 road mortalities <sup>b</sup>	93.42	10.57-283.31	6
Straight-line distance (within 150 m) to nearest bridge from:			
2004 road mortalities <sup>a</sup>	130.25	93.36-148.87	6 (54.5%)
2005 road mortalities <sup>b</sup>	141.05	132.97-149.13	2 (33.33%)
<sup>a</sup> Data from GPS telephone pole locations.			
<sup>b</sup> Data from GPS actual locations.			



Figure 3: Aerial image of transect section 6 of Great Bay Boulevard 2004 (circles) and 2005 (plus signs) road mortalities. It appears some terrapins are being killed closer to the bridge and where the extensions of the creeks meet the road.

## Discussion

Our findings are similar to past studies proposing that there are greater road mortalities of herpetofauna where there is greater traffic volume. Snakes have been observed to be more vulnerable to road mortality as traffic peaks during certain time periods (Rosen and Lowe 1994). A correlation has been shown between the number of mortalities and the number of vehicles on a road (Bernardino Jr. and Dalrymple 1992). It has been calculated that, for amphibians, the probability of being killed increases with greater traffic volume (Hels and Buchwald 2001). Fahrig et al. (1995) also suggested that the proportion of dead frogs and toads increased with traffic intensity, while the number of live animals surrounding the roadway decreased.

This study, which has provided us with new information regarding areas of greatest nesting and highest mortality rates, will assist in determining mitigation strategies. A project to install drift fences to reduce terrapin mortality along Great Bay Boulevard has been proposed for the 2006 breeding season.

**Acknowledgments:** We would like to thank Kenneth Able, Michael Kennish, and Roger Hoden of the Rutgers University Marine Field Station facility for technical support and Deborah Lurie of Saint Joseph's University for assistance with data analysis. We would also like to thank the JCNERR volunteers, RUMFS technicians, and John Wnek of MATES for additional support.

Funding for S. Szerlag was provided by NSF GK-12 0139303 (awarded to the SJU Biology Department), Rutgers University Graduate Student Support, Western New York Herpetological Society—Marvin Aures Grant, Sigma Xi Saint Joseph's University Chapter, and Michael Kennish of Rutgers University.

Research was conducted in accordance with the Saint Joseph's University Institute Animal Care and Review Committee (IACUC protocol #: PR-0503) and under the State of New Jersey, Department of Environmental Protection, Division of Fish and Wildlife permits #0423 and # 0521 issued to Rutgers University Marine Field Station and its seasonal employees.

**Biographical Sketch:** Stephanie Szerlag received her B.S. in biology, marine option at Millersville University of Pennsylvania in 2001. She is currently a M.S. in biology graduate student at Saint Joseph's University and plans to defend her thesis in December 2005. This document serves as background material and as partial preparation of her thesis manuscript.

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## **ROAD WATCH IN THE PASS: USING CITIZEN SCIENCE TO IDENTIFY WILDLIFE CROSSING LOCATIONS ALONG HIGHWAY 3 IN THE CROWSNEST PASS OF SOUTHWESTERN ALBERTA**

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### **Abstract**

The municipality of Crowsnest Pass is situated in a rare east-west corridor bisecting the Rocky Mountains in Southwestern Alberta and Southeastern British Columbia. Highway 3, which runs the length of the Pass, is a major transportation route supporting over 13,000 vehicles per day. Wildlife mortality, due to collisions with vehicles, has been identified as a major human-safety and wildlife-conservation issue on this stretch of highway with approximately 109 large mammal deaths per year. Another immediate threat to wildlife populations in the region is the proposed expansion and realignment of Highway 3. The expanded highway footprint and increased traffic will likely affect wildlife use in the area. It is therefore important that decision makers acquire information on where wildlife are most likely to cross the road to ensure effective mitigation measures. Currently, information pertaining to wildlife movement in the Pass is limited.

Road Watch in the Pass is an innovative, community-based research project that engages local citizenry in reporting wildlife observations along Highway 3 through the Crowsnest Pass in southwestern Alberta, Canada. Through the use of a Web-based GIS, interested citizens can participate in data collection that will be instrumental to decision makers in reducing wildlife-vehicle collisions and for developing mitigation measures for highway expansion. Road Watch was designed to test and profile the use of local knowledge and volunteer data collection in the Crowsnest Pass by providing land managers and the community with valuable baseline information related to wildlife highway crossings. The goals of the project are to collect, analyze, and communicate information highlighting crossing locations of wildlife along the highway based on local knowledge and observations, as well as to engage the citizenry of the pass in local issues relating to wildlife movement and safety.

The project was launched in November 2004 after considerable communication with decision makers in the Pass and the hiring of a local project coordinator. There are currently 51 active participants using the website and interactive mapping tool. The 51 participants have recorded over 581 large mammal sightings. These results are provided to the community on a regular basis through the local media, project website, and email messages. Although the project is still new in inception, preliminary results indicate that the community is successfully engaged with an average of five new volunteers joining Road Watch each month. Each volunteer has contributed an average of 12 observations, with 59 percent of the participants submitting observations on more than one occasion. The number of individual observations ranges from one to 167. Participants have recorded the full compliment of large mammals that occur in the pass, including: 243 mule deer (*Odocoileus hemionus*), 106 big horn sheep (*Ovis canadensis*), 66 white-tailed deer (*Odocoileus virginianus*), 64 unidentified deer species (*Odocoileus* spp.), 35 elk (*Cervus elaphus*), 30 moose (*Alces alces*), 11 coyotes (*Canis latrans*), seven black bears (*Ursus americanus*), three wolves (*Canis lupus*), three mountain goats (*Oreamnos americanus*), three grizzly bears (*Ursus arctos horribilis*) and two cougars (*Puma concolor*), with the exception of wolverine (*Gulo gulo*) and lynx (*Lynx canadensis*).

Road Watch observations provide a valuable supplement to mortality data and have the potential to greatly enhance the existing information base. For example, the percentages of species observations from Road Watch correlate to the recorded levels of wildlife mortality, with mule deer as the highest recorded species from both data sources. From preliminary comparisons of these two data sources, we have identified zones with high Road Watch observations corresponding with low mortality records. This may indicate that there are areas where wildlife are successfully crossing, which has important implications for highway mitigation.

Road Watch is an innovative initiative that will generate a unique dataset resulting from a comparative analysis of knowledge sources. Preliminary results demonstrate that this approach increases the knowledge base by providing new emerging knowledge that would not have been explicit from a single source. This initiative also provides the opportunity for the Crowsnest Pass community to actively engage in an important wildlife-conservation issue. This information will be important to citizens in the community and local decisionmakers in relation to human safety and wildlife conservation around Highway 3.

## **THE ROLE OF TRANSPORTATION CORRIDORS IN PLANT MIGRATION IN AND AROUND AN ARID URBAN AREA: PHOENIX, ARIZONA**

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### **Abstract**

While the potential importance of corridors has been acknowledged for both native and non-native species, little is known about how corridors actually function in developed and fragmented landscapes. Transportation corridors, such as roads and freeways, provide fairly consistent habitat conditions traversing nearly all man-made developments, including cropland, suburbs, reserves, and cities, and connect them with undeveloped areas. The combination of the particular conditions along road and freeway verges and the characteristics of the plants that reach these corridors will ultimately determine which species, native or not, will be able to move within cities and developed areas, as well as to and from cities and surrounding undeveloped areas.

This study will advance ecological understanding of the plant species that are able to move through existing corridors in arid and semi-arid urban areas. Urban areas, including freeway corridors, are intensively managed. This study will consider human management and urban development as integral and natural parts of the ecosystem under study. Understanding the similarities and differences in traits that affect movement of plant species along corridors will provide evidence as to whether native and non-native or functional groupings of species actually move differently in corridors. It will contribute to the literature on assessing the potential for particular plant species to invade new areas. Linking local plant processes to the larger landscape scale of movement between cities and undeveloped areas will have important implications for conservation planning in both environments.

Twenty sites were selected along the four major freeways in the cardinal directions around the Phoenix Metropolitan Area. Beginning in March 2004, vegetation surveys have been performed seasonally at each site. In addition, seed-bank samples and bulk-soil samples were collected at each site. The seed-bank samples are germinating in the greenhouse to determine the seed-bank composition; analysis of physical soil characteristics and available and total levels of soil nutrients (nitrogen and phosphorus) is nearly complete.

Initial soil-chemistry results show that levels of plant-extractable nitrate are significantly increased in the surface soil located directly adjacent to the asphalt (ANOVA using log surface soil concentration;  $F = 5.556$ ,  $P = 0.005$ ). There were also significant differences between sites located adjacent to different land uses, with the sites located in the more densely developed city areas having higher nitrate levels than those at the edges of developed areas. The urban residential sites had the highest levels, followed by croplands, then lower density "fringe" development, and desert sites had the lowest levels of extractable nitrate (ANOVA using log surface soil concentration,  $F = 123.67$ ,  $P < 0.001$ ; Fisher's multiple comparison, all combos  $P < 0.001$ ).

The plant community composition and seed-bank composition at these sites will be compared with nutrient levels to determine whether similar patterns emerge. It is likely that in the typically nitrogen-limited Sonoran Desert, the addition of nitrogen as a result of exhaust from combustion engines is significantly impacting which plant species are most likely to grow along the roadsides. This raises the question of whether heavily traveled roadsides in naturally nutrient-limited ecosystems should be considered as potential vegetation reserves, since intense maintenance would likely be needed to maintain a native community. Perhaps these areas are best landscaped with species unlikely to move along the highway corridors, whether native or not.

The results of this research will advance ecological understanding in several ways. I will elucidate the suite of plant traits that allow effective dispersal in fragmented landscapes with well-defined corridors, clarifying whether these corridors favor plants with particular traits, rather than native or non-native species. This study will increase understanding of the connection between urban and extra-urban environments and will have important implications for conservation planning in both types of environments. Finally, this research specifically incorporates humans into ecological theory, including human management and urban development as an integral and natural part of the ecosystem under study.

The project results will also be useful to highway and road managers, particularly in arid areas. The results will illuminate potential management techniques that will enhance or prevent plant migration along transportation corridors, as well as providing information on how management of transportation verges for objectives other than plant dispersal is likely to affect plant community composition.

## **SOFTWARE FOR POCKET PC TO COLLECT ROAD-KILL DATA**

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### **Abstract**

Animal-vehicle collisions are an important issue in North America. Accidents are numerous and result in human injuries and fatalities, property damage, and the death or injury of the animals concerned. Some animal species may be affected at the population level and face increased risk of local or regional extinction due to the high number of road-kills and other negative effects of roads and traffic. Systematically collected road-kill data can help quantify the magnitude of this problem and potential changes in road-kill occurrences and "hot spots" over time. Such data allows for prioritization and focusing of mitigation efforts to avoid or reduce collisions.

However, not all DOT's or DOT districts record animal-vehicle collisions and the DOT's that do record road-kill data often use different methods. A national standard and tool for the recording of animal-vehicle collisions would not only stimulate DOT's and other organizations to collect animal-vehicle collision data, but would also allow for more effective analyses and use of the data.

The Western Transportation Institute at Montana State University (WTI-MSU) has developed software that allows for easy, standardized, and spatially precise collection of animal-vehicle collision data. The software runs on a Pocket PC that is linked to a Global Positioning System (GPS). The software distinguishes between "monitoring" and "incidental observation" modes and tracks the route of the observer. Road-kill data, including species name as well as optional parameters such as the sex of the animal, are stored in a separate file that can be uploaded to a PC and imported into standard spreadsheet or mapping software. Recording road-kill observations with this tool eliminates manual data entry and transcription.

Beyond the basic data-collection software, we anticipate developing data-management and analysis software that will allow for easy merging and analyses of data from numerous sources, including cluster analyses, and linking to other spatial data in a Geographic Information System (GIS). This has the potential to allow for much faster and better feedback to plan and prioritize for mitigation to address human-safety or conservation concerns.

Once mitigation measures have been put in place, the tools and procedures described above allow for proper evaluation of these measures. We expect that the tools and procedures will ultimately result in fewer animal-vehicle collisions, less work for road maintenance crews, and a reduction in the transportation and disposal costs of the carcasses. A CD-ROM that demonstrates the software is available on request. Please contact WTI-MSU if you have further questions or if your organization is interested in helping us with the testing and further development of this tool and procedures.

## SPATIAL PATTERNS OF ROAD KILLS: A CASE STUDY IN SOUTHERN PORTUGAL

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**Abstract:** Roads promote high levels of animal-vehicle collisions and have one of the most visible man-made impacts on wildlife. In Portugal, SW Europe, very few ecological studies have focused on the impacts from roads on vertebrates. Knowledge of the main factors driving the emergence of hotspots of vertebrate mortality is still scarce.

A segment of a main road 26-km long was sampled by car at an average speed of 20 km/h every two weeks for two years (54 surveys) between 1995 and 1997, collecting all road-killed specimens found. We defined road sections with high collision rates, or vertebrate-mortality hotspots (VMH), by detecting clusters of animal collision locations. The analysis was conducted by comparing the spatial pattern of road kills with that expected in a random situation. In such a condition, the likelihood of collisions for each road section would show a Poisson distribution. Differences of explanatory variables between hotspots and low-mortality sections were evaluated with the Mann-Whitney U-test. Also, a direct-gradient analysis (Canonical Correspondence Analysis (CCA)) was executed with the mortality rates of the 24 most-killed species and the explanatory variables considered.

A total of 2421 vertebrate road-killed specimens were collected, which corresponded to nearly 46 specimens per 0.5 km per year. Eighty non-domestic species were recorded. Several sections were defined as VMH, both for all observations and for each vertebrate class. Results suggested that some road sections should receive particular mitigation actions given that mortality hotspots may arise, particularly sections where montado is the dominant habitat and where stream and other water courses run nearby and parallel to the road.

### **Introduction**

One major human agent of habitat fragmentation is the ever-increasing and expanding road network worldwide (Forman et al. 2002) that can be harmful to various faunal groups such as invertebrates (e.g., Haskell 2001), amphibians (e.g., Carr and Fahrig 2001), reptiles (e.g., Gibbs and Shriver 2002), birds (e.g., Kuitunen et al. 1998) or mammals (e.g., Philcox et al. 1999). Roads and traffic can act as barriers which may make animal movements difficult and reduce population connectivity. By diminishing the gene flow and disrupting sink-source population dynamics, roads may increase inbreeding and loss of genetic diversity (Ferrerias 2001). Resultant isolation might lead to higher local population extinction risks due to stochastic effects (van der Zande et al. 1980; Saunders et al. 1991; Fahrig and Merriam 1994; Cooper and Walters 2002).

Roads also promote high levels of animal-vehicle collisions, which is particularly significant for larger species with wider home ranges as carnivores. These collisions are one of the most visible road impacts on wildlife (e.g.: Hodson 1960; Oxley et al. 1974; Fahrig et al. 1995; Philcox et al. 1999; Gibbs and Shriver 2002; Taylor and Goldingay 2004).

In Portugal, SW Europe, very few ecological studies have focused on the impacts of roads on vertebrate populations. Knowledge of the main factors driving to the emergence of hotspots of vertebrate mortality is still scarce. This study refers to a two-year roadkill survey on a main road (IP2) located in southern Portugal (figure 1).

### **Study Area**

The study was conducted in Portalegre District, between the cities of Portalegre and Monforte cities near the Natural Park of S.Mamede (NPSM) (figure 1). This region is in the center of the Iberian Peninsula, generally dominated by smooth areas, except on the natural park where mountain topography reaching 1024 meters a.s.l. The climate is mediterranean, although the NPSM is considered to be an Atlantic Biogeographic island in the middle of a mediterranean region. This biogeographic crossroad enables the coexistence within the same area of several species from both biogeographic regions.

Road vicinity is dominated by characteristic mediterranean agro-forestry areas; cork and holm oak tree stands (*Quercus suber* and *Q. rotundifolia*), hereafter referred as "montado;" open land as pastures, meadows, or extensive agriculture (cereal, fodder); and olive groves (figure 2). This IP2 section has a moderate traffic intensity of about 5000 vehicles day<sup>-1</sup>.

### **Methods**

#### **Sampling**

A segment of the IP2 road (26-km long) was sampled by car at an average speed of 20 km/h every two weeks for two years (54 surveys) between 1995 and 1997. All vertebrates found killed on the pavement were collected and identified to species level *in loco*, whenever possible, or by analysis of skin, scales, feather or hairs, depending on the taxonomic group, in the laboratory.

It should be emphasized that the number of casualties found was most probably biased due to several constraints, namely carrion foraging from other animals, climatic conditions, and physical characteristics of roads, which can mislead correct counting and detection of corpses on roads (see Erritzoe et al. 2003, pers. observ.). Thus, records should be regarded as an underestimation of real carnage occurring on the road. Furthermore, non-daily surveys prevent the detection of all small-bodied animals like amphibians, passerines, or small mammals, since their corpses often remain between one and three days on the traffic lane (António Mira, unpublished data).

### Explanatory variables

For each 0.5-km road section, we created a 500-meter-radius buffer, with its center on the section's middle point. Land cover was assessed for these buffers through orthofoto map analysis, with corrections from field work observations. Five classes of land cover were considered: montado (MNT), open areas (OPEN), olive groves (OLIVE), fruit tree groves and horticultures (FRUIT), and urban areas (URBAN) (figure 2).

On each buffer, we also considered the length of streams present inside each buffer (STREAM\_L) and the distance of the middle point section to nearest stream (STREAM\_D). The number of culverts (CULVERT) and houses (HOUSE) present on each 0.5 km road section were as well considered. All the information was processed with ArcView 3.2 (ESRITM, Redlands, California).

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

We considered that a section was a potential vertebrate-mortality hotspot when its probability summation exceeded the 90 percent threshold, that is  $\sum p(x) > 90\%$ .

### Data analysis

Differences of explanatory variables between hotspots and low-mortality sections were evaluated with the Mann-Whitney U-test (Zar 1999). This analysis was performed for all observations, for anurans and caudata orders (amphibians), and for the vertebrate classes reptiles, birds, and mammals (domestic cat and dog were excluded from analyses).

For multivariate analysis, we used canonical ordination techniques. A direct gradient analysis (Canonical Correspondence Analysis (CCA)) was executed with the mortality rates of the most 24 killed species (species with above 15 casualties, table 1) and the explanatory variables considered, with downweight of rare species and detrending by segments options (Jongman et al. 1995), using CANOCO for Windows version 4.5 (ter Braak and Šmilauer 2002).

We selected the variables MNT, OLIVE, OPEN, FRUIT, CULVERT, STREAM\_L, and STREAM\_D. This option was chosen in order to achieve a compromise between obtaining the maximum percentage of variance explained and the significance of both eigenvalues and correlations of species-explanatory variables with the axis. The significance of species-environment correlation was tested by the Monte Carlo test (499 permutations). Ordination axes were interpreted using the intraset correlations that allow inference on the relative importance of each variable for predicting community composition (ter Braak 1986).

### Results

A total of 2421 vertebrate road-killed specimens were collected, which corresponded to about 46 specimens per 0.5 km per year. At the species level, 2128 individuals were identified. Eighty non-domestic species were recorded (table 1).

Casualties among vertebrate classes were significantly different (chi-test,  $X^2 = 1630$ ,  $df = 3$ ;  $p < 0.001$ ), being higher for amphibians ( $n = 1362$ ), followed by birds ( $n = 681$ ), mammals ( $n = 225$ ), and reptiles ( $n = 153$ ).

Several sections were defined as vertebrate-mortality hotspots (VMH), both for all observations and for each vertebrate class (figure 3). VMH clusters seemed to be mainly aggregated at the first half of the studied road segment.

Regarding amphibians, hotspots of anuran mortality occurred mainly in the proximity of streams ( $U = 203.5$ ,  $n_1 = 35$ ,  $n_2 = 18$ ;  $p < 0.05$ ), and in sections with a lower number of culverts ( $U = 159.0$ ,  $n_1 = 35$ ,  $n_2 = 18$ ;  $p < 0.01$ ). For caudata, a high number of killed specimens were detected in sections with a low number of houses near the road ( $U = 214.0$ ,  $n_1 = 37$ ,  $n_2 = 16$ ;  $p < 0.05$ ).

Concerning reptiles, road sections with high mortality also had a lower number of culverts ( $U = 192.5$ ,  $n_1 = 37$ ,  $n_2 = 16$ ;  $p < 0.05$ ). Stream proximity was also significant, because the hotspots of mortality were closer to stream lines than to other sections ( $U = 203.5$ ,  $n_1 = 37$ ,  $n_2 = 16$ ;  $p = 0.073$ ).

Higher bird mortality occurred in road sections near watercourses ( $U = 145.0$ ,  $n_1 = 39$ ,  $n_2 = 14$ ;  $p < 0.01$ ), with houses in close proximity of the road ( $U = 153.5$ ,  $n_1 = 39$ ,  $n_2 = 14$ ;  $p < 0.01$ ), and with a lower cover of montado ( $U = 153.0$ ,  $n_1 = 39$ ,  $n_2 = 14$ ;  $p < 0.05$ ).

Concerning environmental variables, there were no significant differences between road sections with high and low mortality of mammals.

The direct-gradient analysis (CCA) results are shown in figure 4. The eigenvalues were 0.153 in the first axis and 0.063 in the second. The Monte Carlo test was significant for both the first canonical axis ( $F = 6.099$ ,  $P < 0.001$ ) and all canonical axes ( $F = 1.933$ ,  $P < 0.001$ ). The first two axes explained 74.2% of data variability. First axis reflects mainly the effects of fruit-tree groves and horticulture (FRUIT), which are related to anthropogenic presence, and the montado cover density (MNT). The second axis reflects the proximity and length of watercourses near the road (STREAM\_P and STREAM\_L).

On the CCA plot, we observed that most species are positioned on the left side, suggesting that higher mortality rates occurred in sections dominated by montado. Exceptions to this are the cases of *Passer domesticus* (PD) or *Sylvia atricapilla* (SyA), species that are related to anthropogenic environments and are shown close to the FRUIT variable. Fruit-tree groves and small horticulture are typically located near small urban areas in mediterranean landscapes (as is the case near Portalegre). Anuran mortality seemed to have occurred on sections close to watercourses. Also noteworthy is that the position of several small species (such as the amphibians *Bufo bufo* (BB), and *Bufo calamita* (BC), the reptile *Natrix maura* (NM), and the small mammal *Apodemus sylvaticus* (AS) suggests that higher mortality levels occurred on sections with a lower number of culverts.

## Discussion

Mortality rates on the Portuguese road presented in this study support the ideas that further road expansion should consider impacts on animal populations and that mitigation measures must be taken account on the existing road network. Furthermore, considering that the Iberian Peninsula is included in a global-biodiversity hotspot, namely the Mediterranean Basin (Myers et al. 2000), and that most species are in one way or another threatened by anthropogenic actions such as road expansion (de Vries et al. 2002), high-priority actions should be implemented so that on Iberian roads can provide a more permeable road system to animal movement. This is more relevant for the studied road given its location, which is near the border of an important Portuguese protected area, Serra de S. Mamede Natural Park (figure 1). This area is located in a biogeographic crossroad combining Mediterranean and Atlantic climatic characteristics, which provides multiple habitat patches allowing high species diversity and richness. Probably this is reflected in the highest number of road-killed species and specimens being found on the first kilometers. As suggested by Spector (2002), biogeographic crossroads appear to be areas of high conservation priority and opportunity in both the short and long term and require increased attention in the process of setting conservation priorities.

Results suggest that some road sections should receive particular mitigation actions given that mortality hotspots may arise. This is particularly true in sections where montado is the dominant habitat and where stream and other watercourses run nearby and parallel to the road. Also, the presence of culverts may diminish the collision risk, providing alternative paths for road crossings. This way, as previous authors described (e.g., Yanes et al. 1995; Rodríguez et al. 1996; Cain et al. 2003; Mata et al. 2005), the implementation of several of these or other similar structures, with different sizes and configurations, should be of primary concern.

Presently, an ongoing project using the same methodology is taking place on the same segment of road with the purpose of evaluating and comparing the vertebrate mortality rates and their spatial patterns 10 years after.

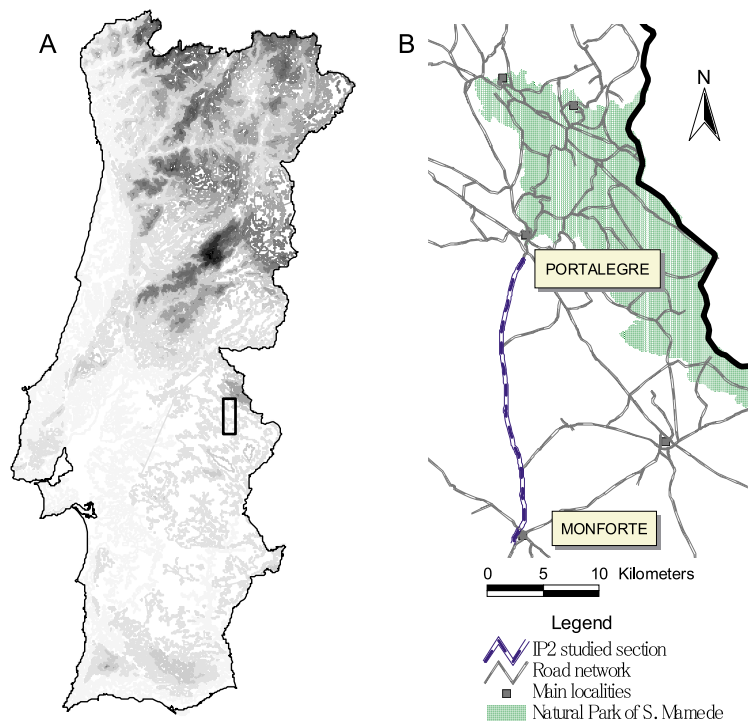


Figure 1. Location of studied IP2 road section (A) and map of study area (B).

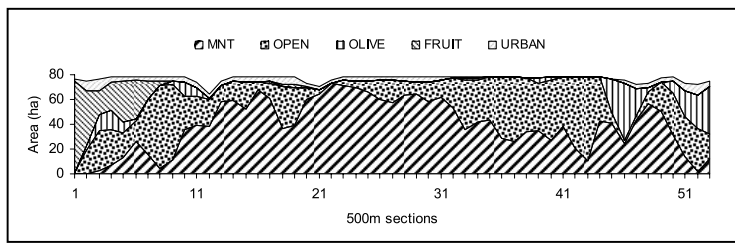


Figure 2. Land cover (main classes) within each 500-meter-radius buffer (near 79 ha) along the studied road.

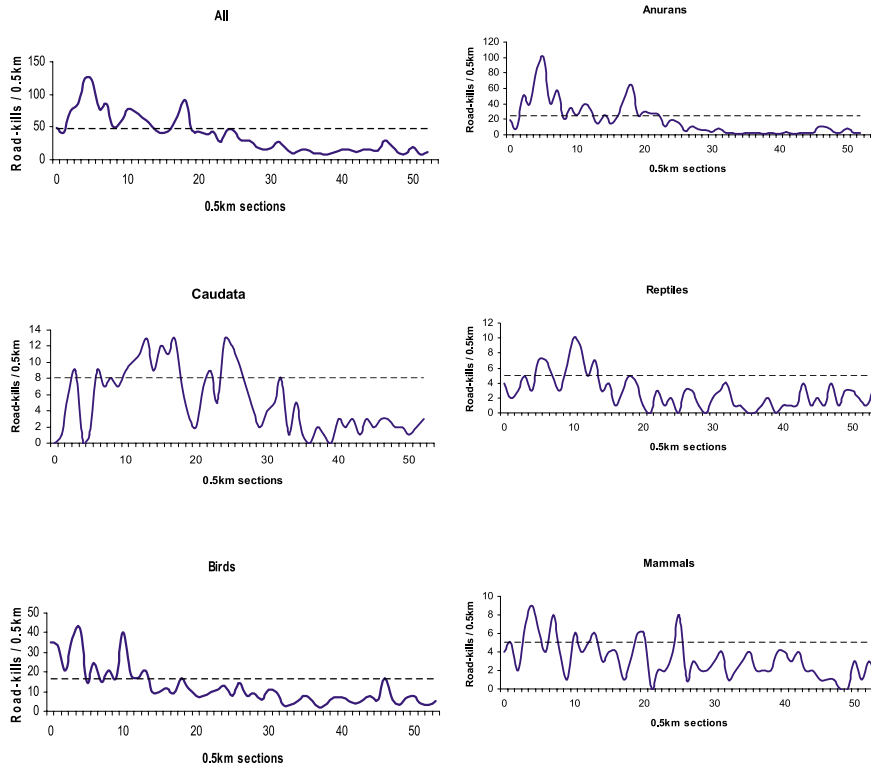


Figure 3. Road mortality along 0.5-km road sections. The dashed line sets the threshold for the definition of vertebrate mortality hotspots (Malo et al. 2004): 46 individuals for all observations, 24 for anurans, eight for caudata, five for reptiles, 17 for birds, and five for mammals.

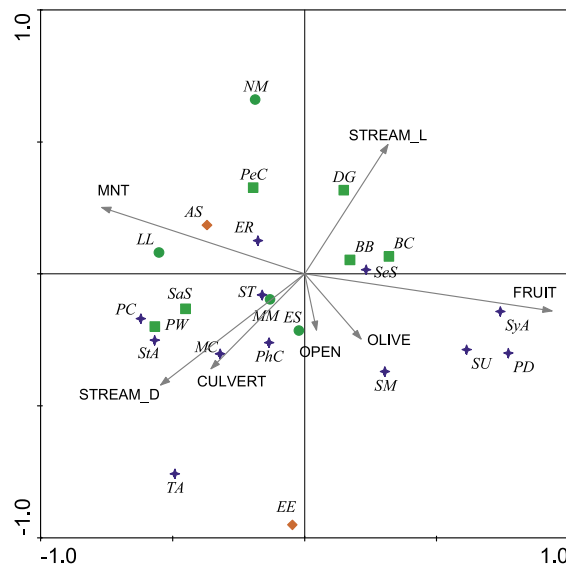


Figure 4. CCA ordination plots of the 24 most killed species (squares are amphibians, circles are reptiles, stars are birds, and diamonds are mammals), with explanatory variables. See text for variables' names. Longer vector lines represent stronger "intrasets correlations" (ter Braak 1986). See text for variables' names and methods.

Species: Amphibians - BB, *Bufo bufo*; BC, *Bufo calamita*; DG, *Discoglossus galganoi*; PeC, *Pelobates cultripes*; PW, *Pleurodeles waltl*; SaS, *Salamandra salamandra*; Reptiles - ES, *Elaphe scalaris*; LL, *Lacerta lepida*; MM, *Malpolon monspessulanus*; NM, *Natrix maura*; Birds - ER, *Erithacus rubecula*; MC, *Miliaria calandra*; PC, *Parus caeruleus*; PD, *Passer domesticus*; PhC, *Phylloscopus collybita*; SeS, *Serinus serinus*; SM, *Sylvia melanocephala*; ST, *Saxicola torquata*, StA, *Strix aluco*; SU, *Sylvia undata*; SyA, *Sylvia atricapilla*; TA, *Tyto alba*; Mammals - AS, *Apodemus sylvaticus*; EE, *Erinaceus europaeus*.

Table 1. Species identified during field work (54 surveys on a 26 kilometer road section); Portuguese red book status (RBS); and number of specimens (N). Species are sorted, within each class, by decreasing number of casualties.

Species	RBS	N
<b>Amphibians</b>		
<i>Bufo calamita</i>	LC	456
<i>Pelobates cultripes</i>	LC	319
<i>Bufo bufo</i>	LC	177
<i>Salamandra salamandra</i>	LC	157
<i>Pleurodeles waltl</i>	LC	106
<i>Discoglossus galganoi</i>	NT	17
<i>Triturus marmoratus</i>	LC	11
<i>Hyla meridionalis</i>	LC	6
<i>Rana perezii</i>	LC	5
<i>Alytes cisternasii</i>	LC	2
<i>Pelodytes sp.</i>	NE	1
<b>Reptiles</b>		
<i>Elaphe scalaris</i>	LC	46
<i>Malpolon monspessulanus</i>	LC	28
<i>Lacerta lepida</i>	LC	21
<i>Natrix maura</i>	LC	16
<i>Mauremys leprosa</i>	LC	12
<i>Psammodromus algericus</i>	NT	9
<i>Coluber hippocrepis</i>	LC	7
<i>Coronella girondica</i>	LC	1
<i>Natrix natrix</i>	LC	1
<b>Birds</b>		
<i>Passer domesticus</i>	LC	80
<i>Miliaria calandra</i>	LC	45
<i>Saxicola torquata</i>	LC	41
<i>Sylvia atricapilla</i>	LC	37
<i>Phylloscopus collybita</i>	LC	33
<i>Sylvia melanocephala</i>	LC	33
<i>Serinus serinus</i>	LC	32
<i>Erithacus rubecula</i>	LC	31
<i>Tyto alba</i>	LC	27
<i>Sylvia undata</i>	LC	26
<i>Parus caeruleus</i>	LC	24
<i>Strix aluco</i>	LC	20
<i>Carduelis carduelis</i>	LC	15
<i>Lanius senator</i>	NT	14
<i>Hirundo daurica</i>	LC	11
<i>Athene noctua</i>	LC	8
<i>Cisticola juncidis</i>	LC	8
<i>Turdus merula</i>	LC	6
<i>Fringilla coelebs</i>	LC	5
<i>Parus major</i>	LC	4
<i>Carduelis chloris</i>	LC	3
<i>Lanius meridionalis</i>	LC	3
<i>Phylloscopus trochilus</i>	LC	3
<i>Alectoris rufa</i>	LC	2
<i>Carduelis cannabina</i>	LC	2
<i>Elanus caeruleus</i>	NT	2
<i>Ficedula hypoleuca</i>	LC	2
<i>Troglodytes troglodytes</i>	LC	2
<i>Turdus philomelos</i>	NT	2
<i>Alcedo atthis</i>	LC	1
<i>Bubulcus ibis</i>	LC	1
<i>Carduelis spinus</i>	LC	1
<i>Ciconia ciconia</i>	LC	1
<i>Falco tinnunculus</i>	LC	1
<i>Galerida cristata</i>	LC	1
<i>Motacilla alba</i>	LC	1
<i>Muscicapa striata</i>	NT	1
<i>Passer hispaniolensis</i>	LC	1
<i>Petronia petronia</i>	LC	1
<i>Phoenicurus ochruros</i>	LC	1
<i>Pica pica</i>	LC	1
<b>Mammals</b>		
<i>Apodemus sylvaticus</i>	LC	37
<i>Erinaceus europaeus</i>	LC	19
<i>Oryctolagus cuniculus</i>	NT	12
<i>Lepus granatensis</i>	LC	11
<i>Vulpes vulpes</i>	LC	10
<i>Mus spretus</i>	LC	9
<i>Pipistrellus kuhlii</i>	LC	7
<i>Crocidura russula</i>	LC	6
<i>Genetta genetta</i>	LC	5
<i>Meles meles</i>	LC	5
<i>Pipistrellus pipistrellus</i>	LC	5
<i>Mustela putorius</i>	DD	4
<i>Microtus cabreræ</i>	VU	3
<i>Microtus duodecimcostatus</i>	LC	3
<i>Herpestes ichneumon</i>	LC	2
<i>Rattus norvegicus</i>	LC	2
<i>Talpa occidentalis</i>	LC	2
<i>Martes foina</i>	LC	1
<i>Rhinolophus ferrumequinum</i>	VU	1

**Acknowledgment:** This work was partially supported by EP-Estradas de Portugal, E.P.E. (Portuguese Roads Institution).

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