Universidade de Aveiro
2020

Mariana Isabel Rossa Mendes

The importance of trophic restoration in the conservation of the Iberian wolf

A importância da restauração trófica na conservação do lobo-ibérico

Universidade de Aveiro
2020

Mariana Isabel Rossa Mendes

The importance of trophic restoration in the conservation of the Iberian wolf

A importância da restauração trófica na conservação do lobo-ibérico

Dissertação apresentada à Universidade de Aveiro para cumprimento dos requisitos necessários à obtenção do grau de Mestre em Ecologia Aplicada, realizada sob a orientação científica do Doutor João Luís Oliveira Carvalho, Investigador Júnior, CESAM & Departamento de Biologia, Universidade de Aveiro e coorientação do Doutor Carlos Manuel Martins Santos Fonseca, Professor Associado Convidado com Agregação do Departamento de Biologia da Universidade de Aveiro.

À minha avó e à minha mãe.

o júri

presidente	Prof.ª Doutora Maria Helena Abreu Silva Professora Auxiliar do Departamento de Biologia da Universidade de Aveiro
arguente	Prof. Doutor José Paulo Cortez Professor Adjunto do Departamento de Ambiente e Recursos Naturais do Instituto Politécnico de Bragança
orientador	Doutor João Luís Oliveira Carvalho Investigador Júnior do CESAM & Departamento de Biologia da Universidade de Aveiro

agradecimentos

Ao meu orientador Doutor João Carvalho por todos os conselhos, orientação e total disponibilidade em ajudar.

Ao meu coorientador Professor Doutor Carlos Fonseca pela oportunidade deste tema de tese e de participar neste projeto.

À Raquel Martins pela companhia nesta jornada e ajuda no trabalho de campo, assim como na recolha dos dados de armadilhagem fotográfica.

A toda a equipa da *Rewilding Portugal* e a todos os parceiros envolvidos no projeto LIFE WolFlux pelo apoio no trabalho de campo e ajuda na recolha de dados.

À Doutora Rita Torres, ao Dário Hipólito e à Associação de Conservação do Habitat do Lobo Ibérico, na pessoa do Doutor Gonçalo Brotas, pela autorização de análise dos excrementos e pela cedência dos dados de armadilhagem fotográfica recolhidos no âmbito do Plano de Monitorização do Lobo-Ibérico – Serra da Arada Freita e Montemuro.

À Daniela Teixeira e ao Professor Doutor Miguel Rosalino pela cedência dos dados de armadilhagem fotográfica, recolhidos na Serra da Malcata no âmbito do projeto POCI-01-0145-FEDER-028204 (WildForests). O projeto WildForests é financiado pelo FEDER, através do COMPETE2020 - Programa Operacional Competitividade e Internacionalização (POCI), e por fundos nacionais (OE), através da FCT/MCTES.

À equipa do Doutor Eduardo Ferreira: Joana Fernandes e Doutora Ana Lino pela realização das análises para confirmação genética dos excrementos de lobo.

À Doutora Ana Valente pela ajuda no tratamento de dados de *Distance Sampling*.

À Paloma pela ajuda na recolha dos dados de armadilhagem fotográfica e no trabalho de campo.

À minha mãe, irmãs e João por toda a paciência, apoio e carinho durante esta jornada.

Esta dissertação foi realizada no âmbito do projeto LIFE WolFlux, financiado pelo Programa LIFE da União Europeia, e do projeto Promover a Renaturalização do Grande Vale do Côa, financiado pelo *Endangered Landscapes Programme* (ELP). O ELP é gerido pela *Cambridge Conservation Initiative* e financiado pela Arcadia, um fundo de caridade de Peter Baldwin e Lisbet Rausing.

palavras-chave

Amostragem de distâncias, corço, dieta, favorabilidade ambiental, gado, índice de abundância relativa, seleção de habitat.

resumo

A população de lobo-ibérico (*Canis lupus signatus*) a sul do rio Douro enfrenta diferentes desafios de conservação causados pelo isolamento genético da população, hibridação, fragmentação do habitat e a possibilidade de conflitos Homem-Lobo desencadeada pela predação de gado doméstico. São vários os motivos que potenciam a predação de gado doméstico por parte do lobo, sendo a ausência ou reduzida abundância de presas silvestres, um dos principais. A restauração da cadeia trófica através de reforços populacionais de corço (*Capreolus capreolus*) surge como uma das possíveis soluções a este problema. O presente trabalho estuda os desafios e oportunidades da restauração desta cadeia trófica na área de distribuição do lobo a sul do rio Douro. O primeiro estudo analisou a dieta do lobo-ibérico na área de estudo. Os resultados demonstraram que o lobo se alimenta principalmente de gado (c. 65% da dieta) e que a presa principal foi a cabra doméstica (*Capra hircus*; ocorrência absoluta, OA: 0.52; volume relativo, VR: 52%). Os animais selvagens que consumiu em maior quantidade foram, por ordem de representatividade, o javali (*Sus scrofa*; OA: 0.22; VR: 16%), aves (OA: 0.15; VR:8%) e a lebre (*Lepus europaeus*; OA: 0.08; VR: 7%). O consumo de corço foi detetado numa amostra recolhida na região oeste onde decorrem anualmente processos de reintrodução desde 2013. Esta presença é indicativa de que os processos de reintrodução foram bem-sucedidos e que o lobo começou a alimentar-se de corço. O segundo estudo teve como objetivo atualizar a distribuição, estimar a densidade e abundância relativa, e estudar a favorabilidade do habitat para o corço na área de estudo com base em técnicas de armadilhagem fotográfica e transectos para deteção de indícios. A abundância relativa estimada demonstrou que a população de corço diminui num gradiente sul-norte e este-oeste, indicando que a população de corço está em expansão. A presença e a abundância relativa de corço são influenciadas pela topografia (*i.e*. sobretudo presente e abundante em vales pouco íngremes) e pela cobertura de solo (*i.e*. sobretudo presente e abundante em florestas nativas com subcoberto composto por herbáceas). A disponibilidade de água e a altitude média apenas tiveram efeito significativo na abundância relativa de corço. A heterogeneidade de habitat e a atividade humana não tiveram efeito significativo na seleção de habitat. Através das características ambientais selecionadas foi desenvolvido um mapa de favorabilidade ambiental. As áreas de grande favorabilidade ambiental e com baixa densidade de corço são potenciais locais para a adoção de medidas que conduzam ao reforço das populações de corço. Adicionalmente, os locais selecionados estão estrategicamente localizados para promover a expansão natural das populações de corço estabelecidas e sobrepõem-se à atual distribuição de lobo-ibérico. Estes resultados constituem um importante passo para a restauração da cadeia trófica do lobo-ibérico e para a redução dos conflitos Homem-Lobo.

keywords

Diet, distance sampling, environmental favourability, habitat selection, livestock, relative abundance index, roe deer.

abstract

The Iberian wolf (*Canis lupus signatus*) population on the south of the Douro river faces different conservation challenges caused by genetic isolation, hybridization with both feral and stray dogs, habitat fragmentation, and the possibility of Human-Wolf conflicts triggered by predation on livestock. The high level of livestock predation reflects, among other things, the absence or reduced abundance of wild preys. The restoration of the trophic chain through the reinforcement of the roe deer (*Capreolus capreolus*) population emerges as a solution to this problem. The present work studies the challenges and opportunities of this restoration of the trophic chain in the current distribution of Iberian wolf south of Douro river. The first study analysed the diet of the Iberian wolf south of the Douro river. The results showed that the Iberian wolf feeds mainly on livestock (c. 65% of the diet) and that the main prey was the domestic goat (*Capra hircus*; absolute occurrence, AO: 0.52; relative volume, RV: 52%). The wild animals most consumed by the Iberian wolf were wild boar (*Sus scrofa*; AO: 0.22; RV: 16%), birds (AO: 0.15; RV:8%) and hare (*Lepus europaeus*; AO: 0.08; RV: 7%). The roe deer consumption was detected in a sample collected in the Western region where reintroduction processes have been taking place annually since 2013. This presence is indicative that the reintroduction processes have been successful and that the Iberian wolf has started to feed on roe deer. The second study aimed to update the distribution, estimate the density and relative abundance and study the habitat favourability of roe deer population in this area using camera trap and transects to detect pellet groups. The relative abundance showed that roe deer population decrease from Southern to Northern and East to Western locations, indicating that the roe deer population is expanding. The roe deer presence and relative abundance are influenced by the topography (*i.e*. mainly present and abundant in valleys with moderate slopes) and by land cover (*i.e*. mainly present and abundant in native forests with understorey composed by herbs). Water availability and mean altitude only had a significant effect on the relative abundance of roe deer. Habitat heterogeneity and anthropogenic presence had no significant effect on habitat selection. Through the selected environmental characteristics, a map of environmental favourability was developed. Locations of great environmental favourability and with low density of roe deer are potential sites for the adoption of measures that lead to a reinforcement of roe deer population in this area. In addition, the selected sites are strategically positioned to promote the natural expansion of established roe deer populations and overlap the current distribution of Iberian wolf. These results are an important step towards the restoration of the trophic chain and to reduce Human-Wolf conflicts.

Table of Contents

List of Tables

List of Figures

General Introduction

Although there are various definitions of trophic cascades, we can generally consider them as direct or indirect interactions between trophic levels, whose disruption produces effects on wild populations and communities (Terborgh *et al*., 2006). The view adopted by Terborgh and colleagues includes both directions of vertical interactions, *i.e.* top-down and bottom-up, which contradicts some other published definitions of trophic cascades (Estes *et al*., 2011; Schmitz *et al*., 2004; Strong and Frank, 2010). The idea of bidirectional trophic cascades conveys a broader and more representative definition of what has been documented in nature (Kagata and Ohgushi, 2006 for review). This definition acknowledges not only the population dynamics derived from a disturbance, but also the dynamics between trophic levels in stationary systems (Ripple *et al*., 2016). Healthy trophic cascades are not static, existing normal fluctuations in the density of predator, herbivore and producer populations. However, these fluctuations can become dramatic after a disturbance in the system caused by the direct or indirect human action (Daskalov *et al*., 2007; Pires *et al*., 2016), or by stochastic events such as fires and disease (Holdo *et al*., 2009; Leahy *et al*., 2015).

The disturbance of trophic cascades may change the dynamics of wild populations (Berger *et al*., 2008; Newsome and Ripple, 2015; Ripple *et al*., 2015), foster the spread of wildlife diseases (Levi *et al*., 2012; Markandya *et al*., 2008) and influence biochemical cycles (Bump *et al*., 2009; Holtgrieve *et al*., 2009; Strickland *et al*., 2013). These disturbances can be aggravated in less diverse ecosystems (Calizza *et al*., 2019). In addition to hindering species conservation and ecosystems functionality, unbalanced trophic cascades and disrupted trophic chains may foster Human-Wildlife conflicts. Human-Wildlife contacts are increasingly common partially due to the expansion of humanized areas into natural environments (Soulsbury and White, 2015). This expansion is often unregulated and quickly turn Human-Wildlife contacts into Human-Wildlife conflicts. For instance, by promoting landscape fragmentation, urban sprawl may have unintended consequences in the surrounding environments leading some species to become “invasive” throughout their original range (Carey *et al*., 2012). There are several other examples of conflicts resulting from Human interferences in trophic cascades. For instance, the absence of predators may results in the burgeoning of wild ungulate populations, which can represent a cause of ecological, health and socioeconomic concern (Linnell *et al*., 2020; Valente *et al*., 2020). On the other hand, the low density of wild preys forces the predator to change its diet and increase livestock consumption causing great economic losses (*e.g.* Athreya *et al*., 2020; Khan *et al*., 2018; Lovari *et al*., 2009). The management of Human-Wildlife conflicts is therefore a contentious issue raising major concerns among the scientific community, wildlife managers and politicians (Lute *et al*., 2020; Redpath *et al*., 2013).

An unbalanced trophic cascade is present in central Portugal due to weakened wild interactions and the creation of new trophic interactions with domestic prey species. The Iberian wolf (*Canis lupus signatus* Cabrera, 1907) population on the south of the Douro river (central Portugal; figure 1) feeds mainly on livestock (Torres *et al*., 2015b; Vos, 2000) at the expense of wild prey species, such as roe deer (*Capreolus capreolus* Linnaeus, 1758) and wild boar (*Sus scrofa* Linnaeus, 1758). This trend is probably due to the low density and diversity of wild prey as well as poor husbandry system (*e.g*. absence of fences or guarding dogs) that do not prevent wolf attacks (Torres *et al*., 2015b). Attacks on livestock trigger misperceptions regarding this carnivore, contributing to Human-Wolf conflicts (Anthony and Tarr, 2019). In this study area, occurred 350 to 400 wolf attacks on livestock in 2002 (Espirito-Santo, 2007). These problems of coexistence, together with the already existing problems of connectivity and instability of packs caused by the habitat degradation and fragmentation (Torres and Fonseca, 2016), make this population an important conservation issue.

The restoration of the trophic chain is envisaged as one of the key solutions to overcome the problems of conservation and mitigate the Human-Wolf conflict in this region. By promoting the expansion of wild prey population, we expect to contribute to a decrease in livestock predation and reduce conflicts with Humans (Imbert *et al*., 2016; Meriggi and Lovari, 1996).

Restoration of trophic chains through species reintroduction or population reinforcements should be studied carefully. Several examples of species introductions have produced complex and unexpected ecosystem changes

(Johnson and Cushman, 2007; Lizarralde *et al*., 2004). In Argentina, the absence of native beavers led the government to introduce an exotic beaver species (*Castor canadensis*). This species quickly grew and became invasive, altering the riparian ecosystem to grassland and wetland, increasing the flooding area (Lizarralde *et al*., 2004). These changes caused the accumulation of organic compounds that alter the composition of water and soil, and the decomposition processes (Lizarralde *et al*., 2004). In this way, the whole process must be thought out in advance, studying the possible effects and not neglecting the social acceptance component. To reduce the unpredictability of reintroduction events, the International Union for Conservation of Nature (IUCN) has developed a set of guidelines that must be respected in all processes of introduction, reintroduction and population reinforcement (IUCN/SSC, 2013). Despite caution, this process can have several positive consequences apart from the restoration of trophic chain, such as seed dispersal (Fernandez *et al*., 2017; Marjakangas *et al*., 2018), mitigation of forest fires (Johnson *et al*., 2018), and mitigation of climate change effects through increased carbon storage (Cromsigt *et al*., 2018). Therefore, wildlife restoration processes can be very useful tools for conservation when carefully planned and evaluated.

In order to proceed with the restoration process of the trophic chain, it is necessary to collect different baseline information. First of all, it will be necessary to understand which species are involved in this trophic chain (Andersen *et al*., 2010). Thus, by studying the diet of the Iberian wolf it will be possible to infer which species of prey are consumed and, particularly, the representativeness of wild prey and livestock on Iberian wolf's diet. Once a thorough analysis of the percentage of livestock consumption is performed, the study of the wild prey communities is necessary. The study of the distribution and abundance of wild ungulates, one of the main wolf prey (Ferretti *et al*., 2019; Figueiredo *et al*., 2020), in the study area will allow the identification of which conservation and management measures are needed and in which areas (Bakker and Svenning, 2018). Of the two species of ungulates present in the study area, the roe deer is the species that best fits into the context of population reintroduction and reinforcements. The roe deer is preyed upon by the wolf in several places in

Europe (Barja, 2009; Imbert *et al*., 2016; Jedrzejewski *et al*., 2002). In the northeast of Portugal, roe deer is the main prey of the Iberian wolf even considering the pervasive presence of red deer (*Cervus elaphus* Linnaeus, 1758) and wild boar in the region (Figueiredo *et al*., 2020). Compared to other ungulates, roe deer is smaller in size, which makes it more vulnerable to wolf attack (Barja, 2009), and is expected to cause less damages in the agriculture and forestry (Reimoser and Putman, 2011). Historically, this species has been successfully reintroduced in several locations south of the Douro river (table 1; Torres *et al*., 2015a). Wild boar, on the other hand, is distributed throughout most of the national territory (Bosch *et al*., 2012) and is a source of conflict with farmers (Valente *et al*., 2020). Red deer was historically reintroduced in Central and Southern Portugal, however, there are no records of significant natural expansion from the *Serra da Malcata* Nature Reserve, located in the far south of the study area (Salazar, 2009). Therefore, the roe deer was considered the ideal ungulate species for the trophic chain restoration process. To proceed to the population reinforcement process should be studied the potential benefits and negative impacts considering the ecological, social and economic aspects (IUCN/SSC, 2013). All of these aspects are important for the success of the restoration process and must be addressed by a multidisciplinary team.

The trophic chain disruption south of the Douro river is related to the instability of wolf packs and the low density of wild prey. It is necessary to study the need and viability of a wild prey population reinforcement for the restoration of the trophic chain and to face the identified Human-Wolf conflicts. Therefore, the aim of this study was to assess the diet of the Iberian wolf and which species of prey are consumed, understanding the dynamics of the trophic chain. Moreover, the roe deer population was assessed in detail to establish the reference situation and identify areas of low prey density. The data collected on roe deer population and the evaluation of environmental favourability will allow to identify areas of conservation interest and possible population reinforcements (Jarvie and Svenning, 2018 for review). The identification of these areas must be followed by discussions among stakeholders such as the local population, hunters and police makers, because not all areas suitable for a species coincide with areas of acceptance and tolerance (Behr *et al*., 2017). In this way, the present work follows the IUCN guidelines to produce the baseline knowledge necessary for the trophic chain restoration. Through the responses to the challenges presented here and the tools provided by this work, a set of conservation processes can be initiated, which include population reinforcements and awareness campaigns towards the involvement of the interested stakeholders.

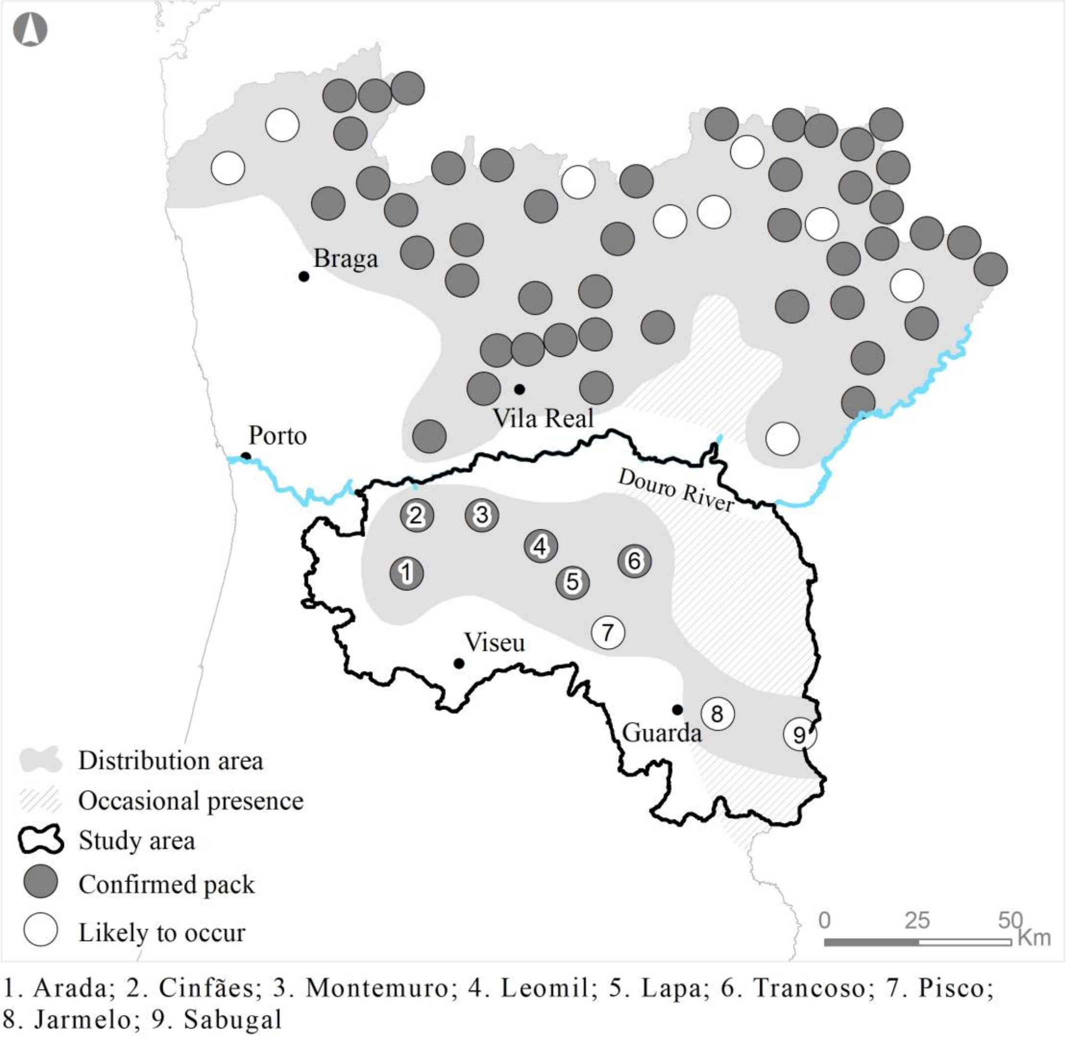


Figure 1 - Distribution of the Iberian wolf in Portugal. Confirmed and probable wolf packs are represented, and the wolf packs studied are depicted and identified. Adapted from Pimenta *et al*. (2005).

Table 1 - Known roe deer reintroductions in the study area (year, sites, number of animals and their origin). Adapted and updated from Torres *et al*. (2015a).

Year	Reintroduction sites	Coordinates (N, W)	Number of animals	Origin
1992	Manteigas	40°22'55.25"; 7°32'15.05"	8	France
1997	São Macário	40°52'50.06"; 8° 3'28.43"	10	Chizé and Trois Fontaines (France)
1999	Manteigas	40°22'55.25"; 7°32'15.05"	2	Chizé (France)
1997	Ribeira de Cadelos	40°33'57.93"; 6°58'13.72"	20	Chizé and Trois Fontaines (France)
1997	Figueira dos Cavaleiros	40°33'5.67";6°55'37.02"	12	Chizé (France)
2000	Aldeia Velha (Sabugal)	40°20'24.47"; 6°53'40.06"	10	Chizé (France)
2000	Ribeira de Cadelos	40°33'57.93"; 6°58'13.72"	10	Chizé (France)
2000	Manteigas	40°22'55.25"; 7°32'15.05"	4	Chizé (France)
2000	Lageosa	40°21'3.99"; 6°50'5.25"	13	Chizé (France)
2000	Lamaçais (Covilhã)	40°18'59.71"; 7°24'13.72"	5	Chizé (France)
2000	Valverdinho (Sabugal)	40°17'56.24"; 7°17'44.52"	14	Chizé (France)
2013	Arada	40°52'0.91"; 8°13'57.85"	12	Valsemana (Spain)
2014	Arada	40°50'46.06"; 8°11'51.33"	24	Valsemana (Spain)
2015	Arada	40°50'26.50"; 8°10'57.45"	22	Valsemana (Spain)
2016	Montemuro	40°56'26.40"; 7°52'30.00"	12	Valsemana (Spain)
2017	Montemuro	40°56'26.40"; 7°52'30.00"	20	Valsemana (Spain)
2018	Montemuro	40°56'26.40"; 7°52'30.00"	12	Valsemana (Spain)

Study area

The study was conducted in Central-North Portugal over an area of 8,941 km^2 (figure 2). The large extension of the study area results in climate differences throughout the territory ranging from temperate climate with strong Oceanic influences (Western region) to Mediterranean regimes (Eastern region; Monteiro-Henriques *et al*., 2016). The topography also has wide variations with ranges from 8 m to 1.382 m. The habitat and land cover vary greatly depending on local conditions and human activities. During the 20th century, socio-economic reasons led the human population to abandon their lands and emigrate to other countries or to the coast of Portugal. This rural exodus allowed the landscape restoration of the region. This mosaic of habitats that characterizes the study area has important consequences for ecosystems, species distributions and community assemblages (Blondel and Aronson, 1999; Peñuelas *et al*., 2017).

I. West part – Freita, Arada and Montemuro mountains

The Westernmost region of study area encompasses two Natura 2000 network sites, the sites of Community Importance *Serras da Freita e Arada*, PTCON0047 and *Serra de Montemuro*, PTCON0025. In this region, natural vegetation passed through several stages of degradation and regeneration. Currently, vegetation is dominated and composed by different scrubland species (*e.g. Cytisus scoparius* (L.) Link, *Cytisus grandiflorus* (Brot.) DC., *Ulex* spp., *Genista triacanthos* Brot., *Erica* spp., and *Pterospartum tridentatum* (L.) Willk.). Some scattered forests can be found (*e.g.* common oak *Quercus robur* L., Pyrenean oak *Quercus pyrenaica* Willd., sweet chestnut *Castanea sativa* Mill., maritime pine *Pinus pinaster* Aiton, in pure or mixed stands with eucalyptus *Eucalyptus globulus* Labill.). This patchwork of habitats is completed with pastures and agricultural fields along this region. The roe deer was recently reintroduced in Freita, Arada and Montemuro mountains (table 1). Apart from roe deer, the wild boar is the only wild ungulate that inhabits the Westernmost region of the study area. Domestic ungulates are represented by goats (*Capra hircus* Linnaeus 1758), sheep (*Ovis aries* Linnaeus 1758), horses (*Equus ferus caballus* Boddaert 1785) and cattle, that generally roamed the mountains in unfenced areas all year around (Torres *et al*., 2015b).

II. Central North part – Aguiar da Beira, Sernancelhe and Trancoso region

The central north region of the study contains the smallest portion of protected reserves in the entire study area, presenting only one Site of Community Importance (*Rio Paiva*, PTCON0059). The climate of this region is characterized by hot summers and cold winters, resulting in large annual thermal amplitudes. It is a region mostly focused on agriculture and husbandry (mainly sheep and goats). The landscape is marked by vineyards and crops of almonds, olive and various cereals. In this way, a mosaic of habitats is formed between permanent crops, scrubland, meadows and some isolated oaks forest (*Quercus pyrenaica* Willd., *Quercus robur* L.) and scattered cork trees. The plantations of pine, eucalyptus and chestnut trees are also predominant in this area. It is an area of confirmed Iberian wolf presence (see Torres and Fonseca, 2016) and is delimited by barriers to the expansion of both wolf and wild prey. These barriers are the Douro river to the north and in the remaining limits by highways (figure 2). In addition to the domestic ungulates, in this central north part of the study area there are also wild boar and roe deer.

III. Northeast part – Côa valley and Douro Internacional region

The Northeast part of study area encompasses one natural park (*Douro Internacional*), two Special Protection Areas (*Douro Internacional e Vale do Águeda*, PTZPE0038; *Vale do Côa*, PTZPE0039), one private protected area (*Faia Brava Reserve*) and one UNESCO World Heritage Site (Côa Archaeological Park). This region experiences a Mediterranean climate of cold winters and hot and dry summers. The Côa river crosses the area from South to North and has shaped the landscape over its course. Mixed patches of cork oak (*Quercus suber* L.), Pyrenean oak and holm oak (*Quercus rotundifolia* Lam.) are interspersed with scrubland composed by *Erica australis* L., *Pterospartum tridentatum* (L.) Willk., *Halimium alyssoides* Lam. and *Cistus ladanifer* L.. Cultivated areas are mainly vines and fields of olives, almonds and cereals. The rupestrian marks identified over the Côa Valley are the evidence that ungulates, including roe deer, red deer and wild boar date back to the Palaeolithic period (Aubry, 2009). Today, however, the only wild ungulates that persist in Côa valley are the roe deer and the wild boar (Fonseca *et al*., 2017). Domestic ungulates are also present.

IV. Central South part – Estrela mountain

This region comprises the largest protected area in Portugal: the *Serra da Estrela Natural Park* with 88,850 ha. It is located in the transition zone of Mediterranean climate with hot and dry summers and the temperate oceanic climate with milder and humid temperatures. The influences of climate and the altitude gradient shape the structure and complexity of the landscapes. Sweet chestnut, pine and eucalyptus plantations and remnant forests of oak (*Quercus rotundifolia* Lam., *Quercus suber* L. and *Quercus pyrenaica* Willd.) can be found in the Natural Park. However, most of the forests have disappeared giving way to degraded vegetation states such as grasslands of *Nardus stricta* L. and scrublands composed by *Arbutus unedo* L., *Frangula alnus* Mill., *Cistus ladanifer* L., *Genista florida* L., *Erica* spp., *Cytisus* spp., *Calluna vulgaris* (L.) Hull and *Juniperus communis* L. subsp. *alpina* (Suter) Čelak (Meireles and Pinto-Gomes, 2012; van der Knaap and van Leeuwen, 1995). At the top of the mountain, vegetation is scarce and the landscape is dominated by rocks. In the region surrounding the Natural Park exists multiple land uses such as woodlands, scrublands, coniferous plantations and agriculture areas (*e.g.* vineyards, olive groves). In the Central South part of the study area, wild ungulates (wild boar and roe deer) and domestic ungulates (mainly sheep) are present.

V. Southeast part – Malcata mountain

The Southeast part of study area comprises one Site of Community Importance (*Serra da Malcata*, PTCON0004). The vegetation is dominated by dense shrubs of *Cytisus* spp., *Halimium* spp., *Cistus* spp., *Erica* spp., *Pterospartum tridentatum* (L.) Willk., and *Arbutus unedo* L.. Scattered woodlands of holm oak and Pyrenean oak trees are also present. Additionally, industrial plantations of *Pinus* spp., eucalyptus, and Douglas firs (*Pseudotsuga menziesii* (Mirb.) Franco) are abundant in this area. Malcata mountain represents the Southern limit of Iberian wolf distribution in Portugal. Roe deer and wild boar have a significative presence in this natural area, whereas red deer population seemed to be limited in number and distribution.

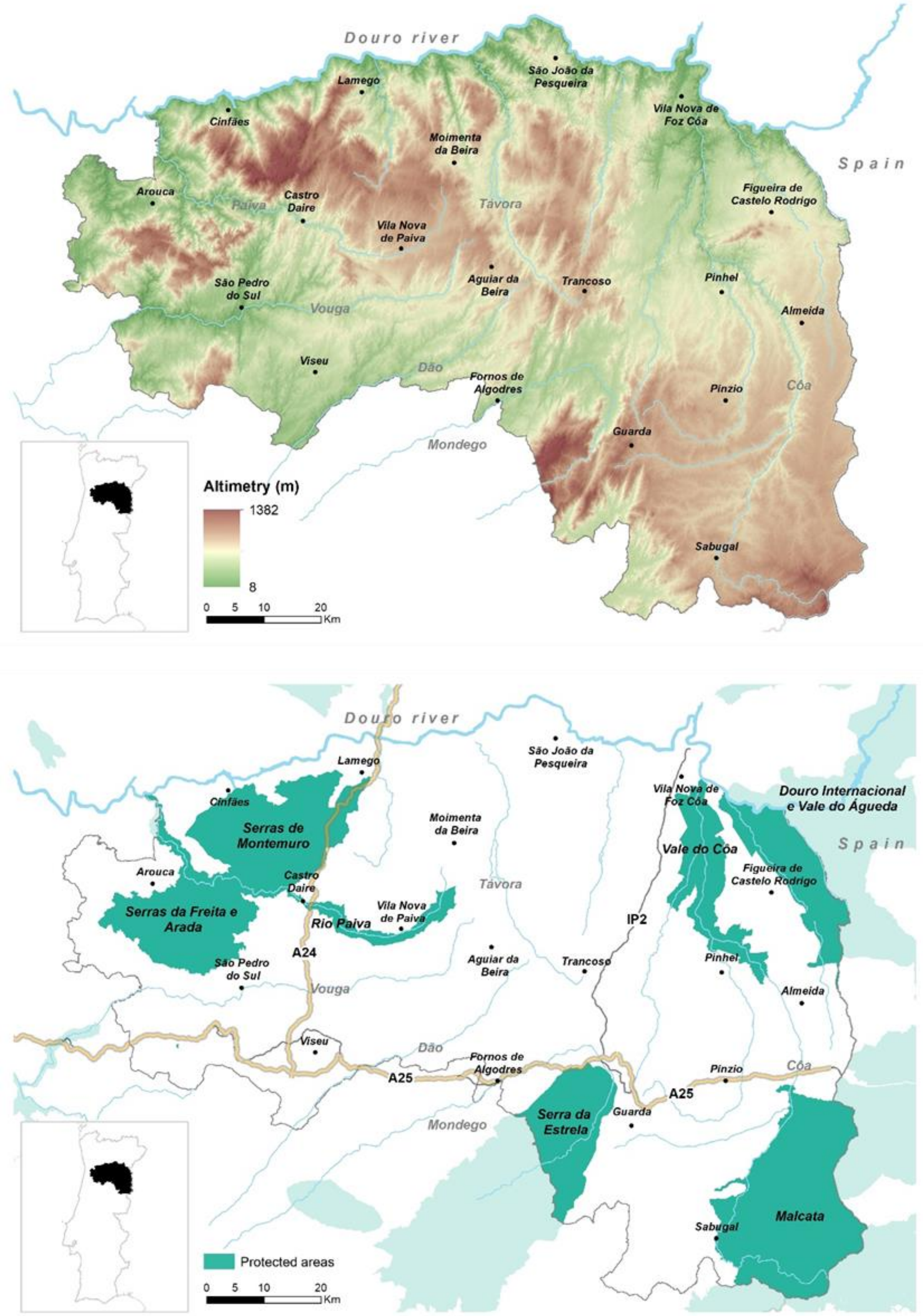
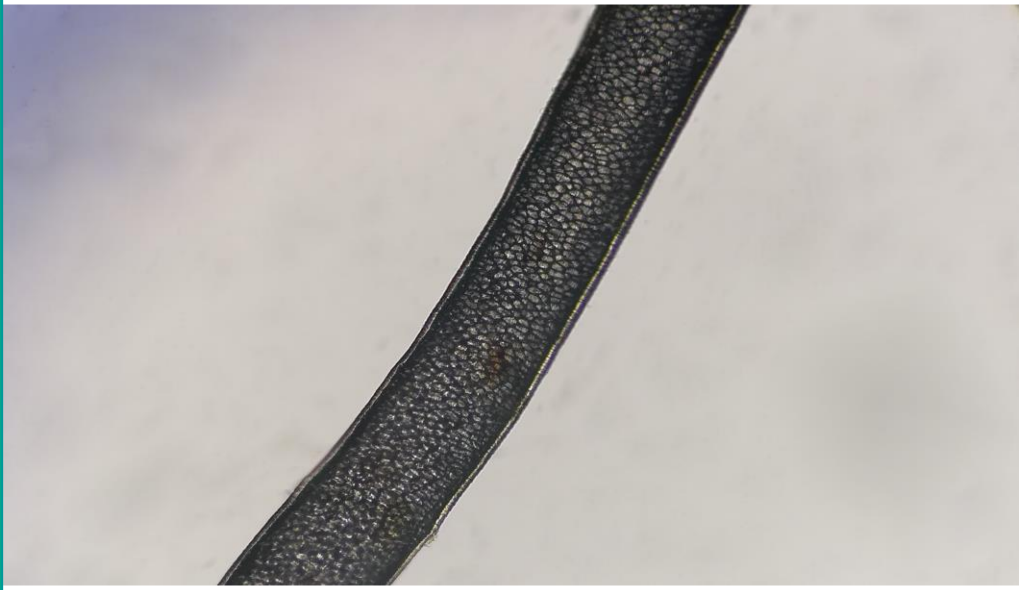


Figure 2 - Location of the study area (8,941 km^2) and geographic variability of elevation and topography. The map below indicates the location of the Natura 2000 network sites: *Serras da Freita e Arada*, PTCON0047; *Rio Paiva*, PTCON0059; *Serra de Montemuro*, PTCON0025; *Malcata*, PTCON0004; *Vale do Côa*, PTZPE0039; *Douro Internacional e Vale do Águeda*, PTZPE0038; *Douro Internacional* and *Serra da Estrela Natural Parks*.

Chapter I

Iberian wolf diet in the southern edge of its distribution



Microscopic observation of the roe deer medulla pattern (400x magnification).

1. Abstract

The conservation of southern Iberian wolf populations is a contentious issue due to the small number and lack of connectivity among packs. The ongoing monitoring of these populations is the most welcome step towards the identification of drivers that limit the expansion and the stability of Iberian wolf population. Knowledge of the feeding habits of large carnivores can be considered an indicator of the carnivore population itself, but also an indicator of wild prey populations. The aim of this study is to analyse the diet of the Iberian wolf south of the Douro River. The samples were collected between 2014 and 2019 in Freita, Arada and Montemuro mountains (Western packs) and between 2019 and 2020 the collection was extended to the entire study area south of the Douro river. All samples were confirmed genetically as being Iberian wolf scats. Through the identification of hairs present in scats it was possible to determine the species of prey. The Iberian wolf in this area consumed more livestock (c. 65% of absolute occurrence, relative occurrence or volume) than wild animals. The goat was the most consumed species representing approximately half of the diet (absolute occurrence, AO: 0.52; relative volume, RV: 52%). This prey was followed by wild boar (AO: 0.22; RV: 16%), birds (AO: 0.15; RV:8%), hare (AO: 0.08; RV: 7%) and sheep (AO: 0.08; RV: 7%). The remaining prey species have little representation in the Iberian wolf's diet (AO: <0.06; RV: ≤4%; individually). The roe deer was found in one sample collected in the Western region, indicating that the roe deer reintroductions conducted since 2013 are slightly increasing the availability of wild prey. The high consumption of livestock can be caused by the low density of wild prey and ineffective livestock protections. Efforts should be made to increase the density of prey (*e.g.* reintroduction or population reinforcements of roe deer) and to apply measures to mitigate wolf attacks (*e.g*. direct surveillance by the shepherd, livestock guarding dogs and fences).

2. Introduction

The knowledge on the feeding habits of large carnivores can be considered a honest indicator about the status of carnivore populations and wild prey communities (Sidorovich *et al*., 2017). Carnivore populations may vary their diet according to social system and/or pack stability (Mills, 1990). For example, lone wolves or dispersers consume preferentially livestock species because are easier to hunt, while stable packs feed preferentially on wild prey (Fritts and Mech, 1981; Imbert *et al*., 2016). The status of wild prey populations is also reflected in the diet of large predators. Three factors related with prey populations rule prey selection by carnivores, i) abundance/accessibility of wild preys, ii) vulnerability, and iii) anti-predatory prey behaviour. The high density of a prey species results in a greater number of encounters with the predator (Hebblewhite and Pletscher, 2002) and, therefore, the likelihood of being preyed and consumed is higher (Gervasi *et al*., 2013; Meriggi *et al*., 1996). However, this pattern is not always linear and may vary for different reasons. The vulnerability of certain individuals due to age class or disease makes them easy to capture, and therefore they are usually selected by predators (Mattioli *et al*., 2011). The defensive behaviour of each prey species is also important (Wirsing *et al*., 2010) as it provides best chances to survive an encounter with the predator (Tambling *et al*., 2015). The diversity of prey present in the ecosystem has also been considered important in the selection of prey (Ferretti *et al*., 2019). When the diversity of wild ungulate species increases, the diet breadth and livestock consumption decreases (Meriggi *et al*., 2015, 1996).

The use of more humanized areas has been a pattern detected in several carnivore species (Athreya *et al*., 2013; Mueller *et al*., 2018). This close contact may become a conservation issue as it usually fosters conflicts between large carnivores and human activities (Anthony and Tarr, 2019). The problems that affect human activities seem to be the most contested, *e.g*. predation on livestock and the reduction of ungulate populations that were once only hunted by man (Andersone and Ozolinš, 2004; Bautista *et al*., 2019). In Europe, approximately 28.5 million euros are spent annually on compensation for damage caused by large carnivores (Bautista *et al*., 2019). These problems cause a negative perception of carnivores by the general public, which leads to conflict and may culminate in retaliatory death (*e.g.* Pohja-Mykrä and Kurki, 2014).

On south of the Douro River, the Iberian wolf population is characterized by isolated packs comprising a small number of individuals, which raises concerns about their conservation (Torres and Fonseca, 2016). Previous studies on the diet of this population showed that the main wolf preys are livestock species (Torres *et al*., 2015b; Vos, 2000). The authors advocated that this result is potentially related to the low density and diversity of wild prey in the area, and poor husbandry practices (Pimenta *et al*., 2017; Torres *et al*., 2015b). Being a region with a lot of livestock breeding and that depends on this activity, the attacks on livestock are a great source of economic loss generating a negative perception of this carnivore by the local communities (Espirito-Santo, 2007). There is a need to apply preventive husbandry methods such as the use of guard dogs and effective fences (Linnell *et al*., 2012). These measures should not be applied in isolation, but in conjunction with measures aimed at increasing the availability of wild preys and actions to raise awareness toward the conservation of the Iberian wolf among the local population.

In the presence of a population of wolves that raises conservation problems, it is important to study and continuously monitoring the feeding habits of this top predator. The knowledge of the Iberian wolf diet will be indicative of the prey community in the study area. The high consumption of livestock could be an indicator of possible conflicts with the local population (Khan *et al*., 2018). The aim of this study is to assess the diet of the Iberian wolf south of the Douro River. The scat collection began in 2014 in Freita, Arada and Montemuro mountains. In 2019, the collection of samples was extended to the entire study area and concluded in January 2020. The present study is not a replication of the work of Torres *et al*. (2015b), but a continuation of the study in a different context of wild prey availability, *i.e*. over a period marked by the reintroduction of roe deer in Freita, Arada and Montemuro mountains (table 1). Thus, the comparison of the wolf's diet before starting the reintroduction process (Torres *et al*., 2015b) and during the process (present study) can be used for evaluating the response of the Iberian wolf to roe deer reintroductions. Reintroduction processes have been implemented in this area on an annual basis, since 2013 (table 1; Torres *et al*., 2015a). Wolf scats collected during this period, were analysed in this chapter and will allow a continuous study of the Iberian wolf diet as the prey population has increased slightly due to reintroductions. The extended collection of samples in the last year make it possible to assess the Iberian wolf's diet throughout the southern edge of the species' distribution.

3. Materials and Methods

3.1. Data collection

Samples were collected between August 2014 and August 2019 in Freita, Arada and Montemuro mountains. In 2019, the collection of scats was expanded to all the study area south of Douro river and the collection period was extended until January 2020 (figure 2). Collections were made along itineraries on foot, randomly distributed in the study area. Scats identification in the field was based on various characteristics such as shape, size and odour. For all samples identified in the field, a portion was collected for genetic identification of the predator following standard procedures (Figueiredo *et al*., 2020). In this way, all the samples used for the determination of the Iberian wolf's diet were confirmed genetically. The samples were stored in plastic bags, identified with the sample code and then kept in a freezer (-5ºC).

3.2. Prey identification

Before the content analysis, each sample remained in the oven for at least 3 hours at 60 ºC. Then, the samples were washed through 1-3mm mesh sieve (Kruuk and Parish, 1981). The hairs and bones in the scat were collected, dried and stored. The identification of prey was performed through the hairs present in the samples and comparison with the reference collection of prey hairs collected in the study area. This comparison was carried out through the analysis of macroscopic and microscopic characteristics. The preparations were made according to Teerink (1991) and applying two techniques: cuticular impression using nail polish to observe the cuticle pattern (figure 3c) and immersion in cedar oil to observe the cortex and the medulla (figure 3a and b). For the microscopic identification of hairs, in addition to the reference collection, illustrated atlas and identification keys for wild (De Marinis and Asprea, 2006a; Teerink, 1991; Valente *et al*., 2015) and domestic ungulates (De Marinis and Asprea, 2006b) were used. Identifications were made using an optical microscope (100-400x magnification). Twenty hairs were collected randomly and identified for each sample for the purpose of quantifying the diet (Lovari *et al*., 2015).

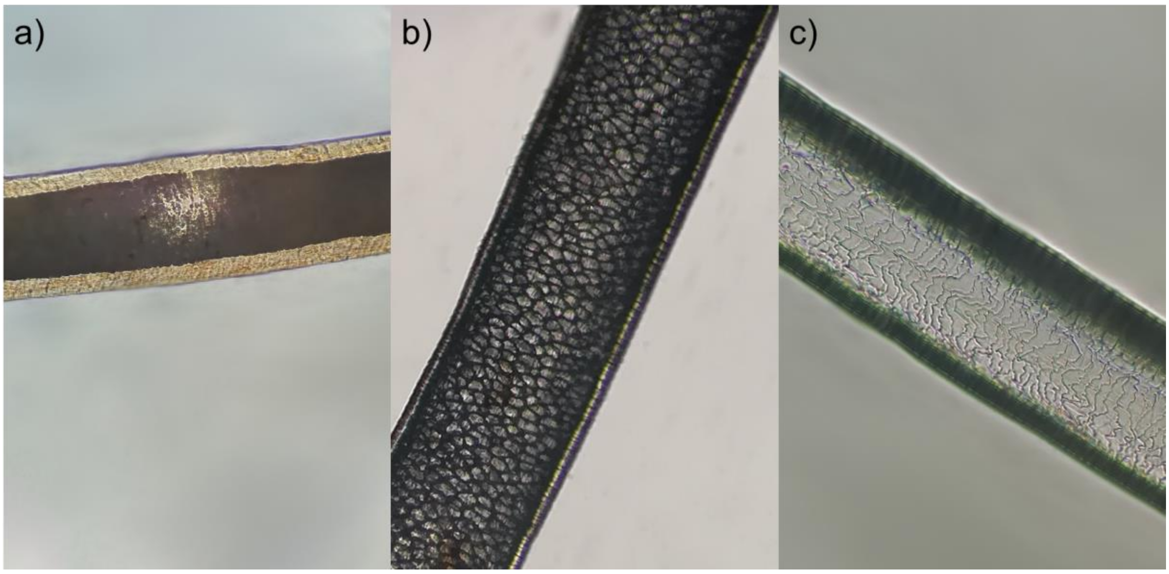


Figure 3 - Microscopic observation of the hair features (400x magnification). Medulla details of cattle (a) and roe deer (b). Cuticular pattern of the domestic goat's hair (c).

3.3. Data analysis

The samples with less than 20 hairs, and whose prey was impossible to identify, were discarded. The preys were separated in food categories for subsequent analysis. These food categories corresponded at the species level for meso-large mammals (*i.e*. wild and domestic ungulates, carnivores and meso-mammals); additionally, the following categories were considered: small carnivores and birds. For each food category, it was calculated the absolute and the relative frequency of species occurrence in diet (Lucherini and Crema, 1995). The absolute frequency of occurrence is obtained through the ratio between the number of occurrences of each category and the total number of scats analysed.

The relative frequency of occurrence is the ratio between the number of occurrences of each category and the total number of occurrences of all categories. The relative volume of each category, in each scat, was obtained visually considering the volumetric classes suggested by Kruuk and Parish (1981). For the analysis, the median point of each of these classes, *i.e*. 2.5%, 13%, 38%, 63%, 85.5% and 98% (Kruuk and Parish, 1981) was used to estimate mean volume of each category in diet (Lovari *et al*., 2015).

4. Results

A total of 73 genetically confirmed wolf scats were analysed to assess the diet of the Iberian wolf (figure 4).

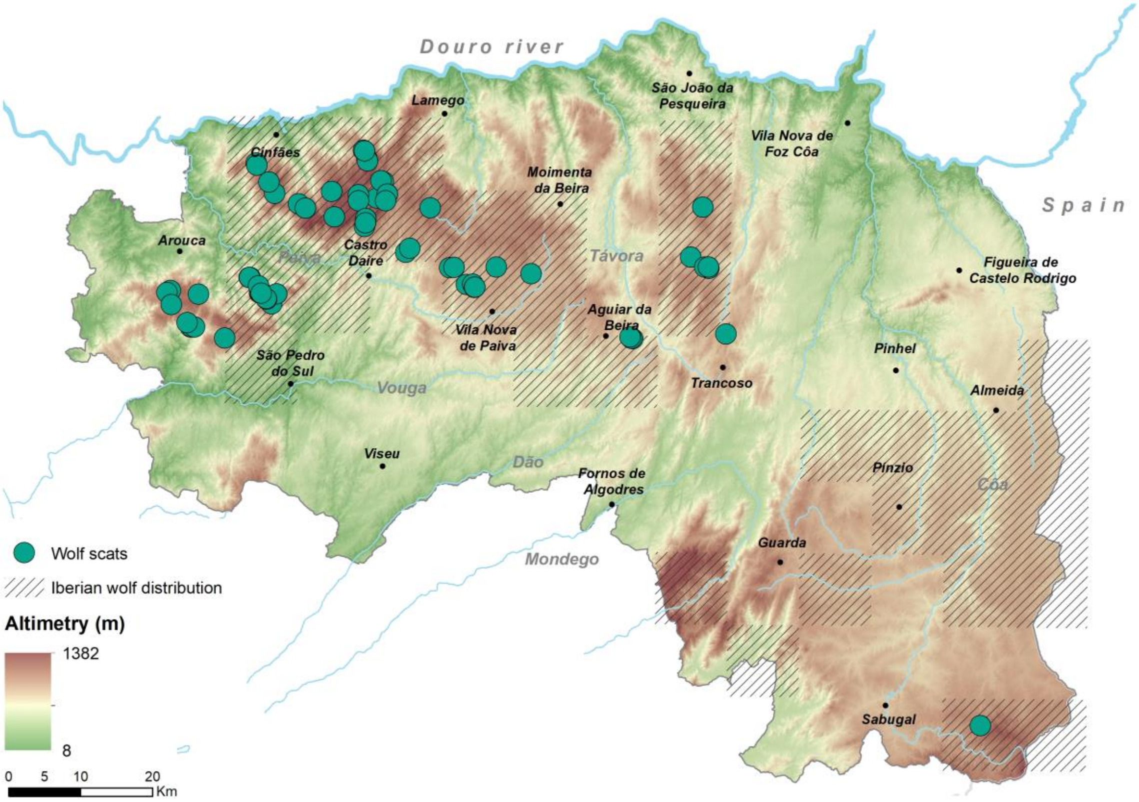


Figure 4 - Location of wolf scats collected and analysed in this study.

The Iberian wolf diet was mainly composed of goats, which showed an absolute occurrence of 0.52 and 52% of relative volume (figures 5-6). The wild boar was the second prey consumed with an absolute occurrence of 0.22 and a relative volume of 16% (figures 5-6). This prey was followed by birds that showed an absolute occurrence of 0.15 and relative volume of 8%. Hare and sheep had the same representation in the diet of Iberian wolf (absolute occurrence of 0.08 and relative volume 7.5%; figures 5-6). The remaining prey are donkey, cattle, roe deer, badger and small carnivores with little representation (absolute and relative occurrence of <0.06 and relative volume ≤4%, individually; figures 5-6). The only records of roe deer and sheep consumption were identified in samples collected in Freita, Arada and Montemuro mountains.

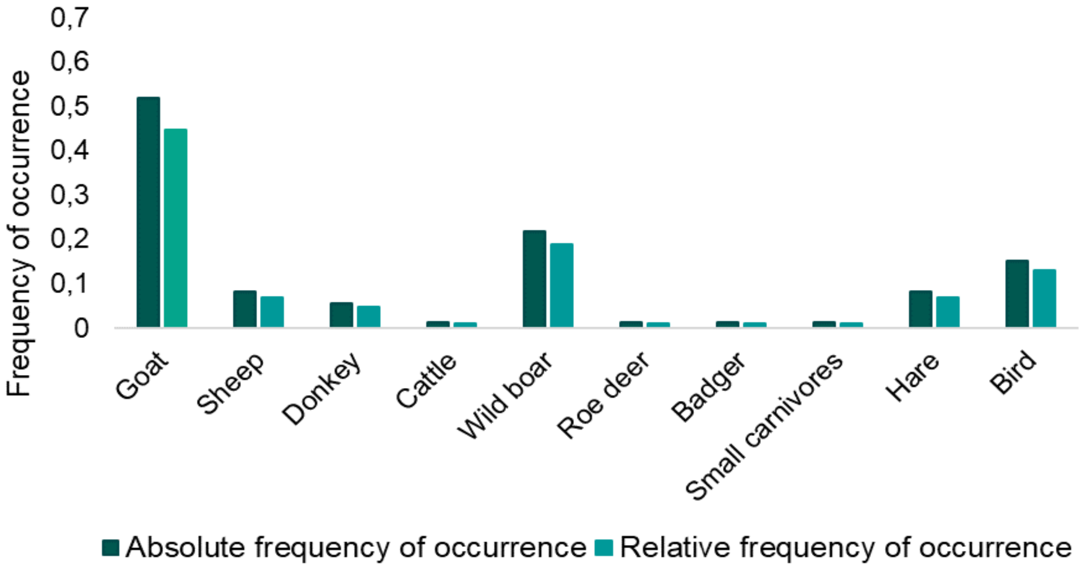


Figure 5 - Absolute and relative frequency of occurrence of food items in diet of wolves (N=73).

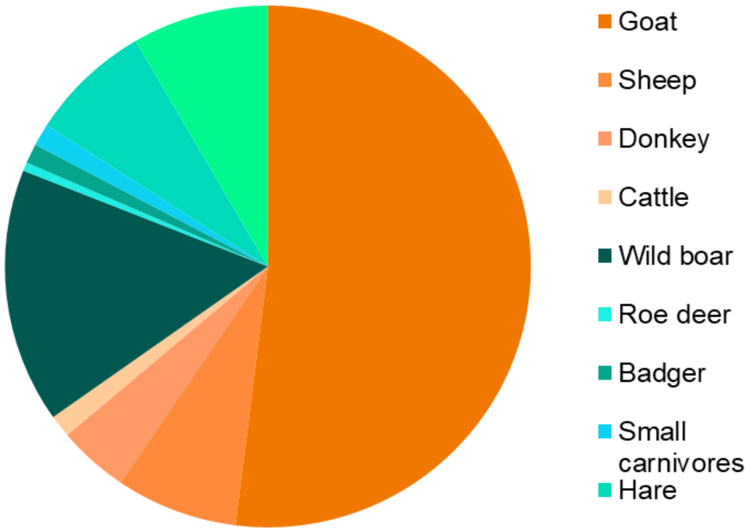


Figure 6 - Relative Volume of food items in diet of wolves (N=73).

5. Discussion

Human-Wolf conflicts are expected when predators feed on livestock (Khan *et al*., 2018). In southern Europe, the consumption of livestock by wolves is often recorded (Iliopoulos *et al*., 2009; Meriggi *et al*., 1996, 1991), which raises conservation and management concerns (Torres and Fonseca, 2016). In the present study, domestic prey is more consumed by Iberian wolf than wild prey. Some authors argue that this may be due to the low density of wild ungulates and ineffective protection measures of livestock (Ferretti *et al*., 2019; Torres *et al*., 2015b). This pattern had already been recorded in the study area (Torres *et al*., 2015b; Vos, 2000), but it was also described in other environmental scenarios. For instance, in central Iran, a study based on 132 samples, demonstrated that livestock are consumed in high proportions by grey wolf (*Canis lupus* Linnaeus, 1758; Hosseini-Zavarei *et al*., 2013). It has been shown that wolf predation is related to several factors such as availability/abundance, vulnerability and accessibility of prey (Ferretti *et al*., 2019; Mattioli *et al*., 2011; Meriggi and Lovari, 1996). The low density of wild ungulates (Torres *et al*., 2015a) and the greater accessibility and vulnerability of livestock species due to the husbandry system used (*i.e*. free-ranging during the day and kept in barns during the night, see Torres *et al*., 2015b), lead the wolf to consume more livestock than wild ungulates. The wild ungulate species most consumed by Iberian wolf was wild boar, whereas roe deer was recorded just once. The remaining preys were consumed as a supplement to the diet in order to survive in less favourable conditions (Meriggi *et al*., 1991).

Several studies have reported the higher consumption of goats by wolf (Papageorgiou *et al*., 1994) and other predators (*e.g.* common leopard *Panthera pardus*, Athreya *et al*., 2020; Khan *et al*., 2018) compared to other species. In Italy, despite the preference for wild ungulates, within the livestock species, the most preferred was the goat (Imbert *et al*., 2016; Meriggi *et al*., 2015). In the present study, the goat was the most consumed species, representing approximately half of this predator's diet. Previous studies in Western region had observed higher frequencies of occurrence than the reported here (52% versus ~62% frequency in Torres *et al*., 2015b; and ~98% frequency in Vos, 2000). This difference can be justified by the increase in the sampled area and the spatial variability in the abundance and accessibility of domestic goats. Among the livestock species, the goat was the most available prey for the wolf because they are usually in large flocks and spread out over the mountains (see Vos, 2000). In addition, goats use steeper areas than sheep being easily attacked by the wolf due to low human presence (Torres *et al*., 2015b). This may justify the higher presence of goats in the Iberian wolf's diet when compared to sheep in this study. Other studies have observed a lower than expected consumption of sheep (Iliopoulos *et al*., 2009; Torres *et al*., 2015b). Despite the great number of wolf attacks on sheep recorded by the government, Vos (2000) only detected goats on the wolf’s scats analysed. This differences can occur due to, the ease of finding sheep carcasses because of the proximity to the villages, the possible attack on the sheep without consuming them and miscrediting the attack to the wolf when a sheep dies from another cause (see Vos, 2000). These results are important since they demonstrate the differences in perception between the government and the data obtained by scientists through the study of the wolf's diet. This discrepancy may cause negative perception of the Iberian wolf and possible Human-Wolf conflicts.

The other livestock species present in the Iberian wolf’s diet on this study were cattle and donkey, with little representation. These species are larger and can serve as a deterrent to attack by the wolf due to the probability of suffering an injury (Meriggi *et al*., 1996). Additionally, cattle may have more effective protection measures (Torres *et al*., 2015b) due to their great economic importance (Iliopoulos *et al*., 2009). In this way, the consumption of these species is lower compared to goats. Predation of these species has had seasonal changes related to the presence in pastures (Meriggi *et al*., 1991; Milanesi *et al*., 2012), the season of calves (Iliopoulos *et al*., 2009) and breeding period of wolves and in the pups independence period (Roque *et al*., 2001). The small number of scats in this study area hampered a season scale study of the diet. Future work should take this analysis into account as it could be a source of knowledge applicable to the improvement of husbandry practices with the aim of reducing livestock consumption by wolves. For example, Pimenta *et al*. (2017) found that 90% of the attacks were associated with the presence of calves with less than 3 months old on the pasture and with the change of this husbandry practice cattle predation will decrease.

The wild animal most consumed by Iberian wolf was wild boar, corroborating previous studies in the study area (Torres *et al*., 2015b). Despite being a common prey in the wolf’s diet in several studies (Capitani *et al*., 2004; Ferretti *et al*., 2019; Figueiredo *et al*., 2020), its presence gains great relevance in studies similar to this, where the abundance of other wild ungulates such as roe deer and red deer is very low (Mattioli *et al*., 2011; Meriggi *et al*., 1996, 1991; Migli *et al*., 2005). Wild boar predation is usually associated with piglets or sick/old animals (Barja, 2009; Mattioli *et al*., 2011; Nores *et al*., 2008). Additionally, in several areas, wild boar is more preyed during the winter (Milanesi *et al*., 2012; Wagner *et al*., 2012) due to the vulnerability caused by severe climatic conditions and deep snow cover (Capitani *et al*., 2004). Healthy adults present active defence against predators (Ferretti *et al*., 2019), for this reason the wild boar is mostly predated when vulnerable (Mattioli *et al*., 2011). Therefore, its consumption by wolves is moderate (Meriggi *et al*., 1991; Torres *et al*., 2015b), even in areas where it occurs at high densities (Migli *et al*., 2005; Nores *et al*., 2008).

The consumption of secondary prey is associated with the scarcity of large wild ungulates (Meriggi *et al*., 1991). Here, this was observed through the consumption of birds, hares and small and meso-carnivores. Espirito-Santo (2007) suggested that, given the high presence of poultry farms in the districts of Aveiro and Viseu, the Iberian wolf could start feeding on this prey. The considerable consumption of birds in this area was first recorded in this study, however it was not possible to discriminate whether they are wild or domestic birds. Consumption of a higher percentage of hares was recorded several times (Chavez and Gese, 2005; Mattioli *et al*., 2011; Wagner *et al*., 2012). This study corroborates the idea that the low density of wild ungulates is reflected in a greater breadth of the wolf's diet (Meriggi *et al*., 1996; Meriggi and Lovari, 1996; Zlatanova *et al*., 2014). This predator in less favourable situations uses smaller prey, garbage or fruit (Meriggi *et al*., 1991). Thus, since the Iberian wolf south of the Douro river is in a less favourable conservation situation, due to the isolation and instability of the packs, together with the low density of wild prey, it is expected the high consumption of livestock and other secondary prey.

Several studies reported a high consumption of roe deer despite the presence of a diverse guild of wild ungulates (Barja, 2009; Figueiredo *et al*., 2020). In contrast, here, roe deer was recorded just once. Yet, this result is a good indicator of the success of previous reintroductions. In previous studies in this area, the absence of roe deer in the diet has been noted (Torres *et al*., 2015b; Vos, 2000) and a process of reintroduction has been initiated in Freita, Arada and Montemuro mountains from 2013 until now (table 1, Torres *et al*., 2015a). The sample in which the roe deer hair was detected was collected in the mountains where the referred reintroductions took place and after these reintroductions (sample dates from January 2018). In this way, we can confirm the predation of this reintroduced species and, therefore, the final goal of this process (*i.e.* to increase the availability of wild prey) is slowly being achieved.

6. Conclusions and management implications

The endangered Iberian wolf fed mainly on livestock species on its southern limit of Portugal. This consumption can lead to Human-Wolf conflicts. Thus, it is necessary to apply measures to prevent livestock consumption. We can expect that by promoting the availability of wild prey (*i.e.* roe deer) through reintroduction/population reinforcement processes, livestock consumption may be reduced (Imbert *et al*., 2016; Meriggi and Lovari, 1996). However, the restoration of the wild prey community should be simultaneous with the application of adequate husbandry practices to prevent wolf attacks (Pimenta *et al*., 2017). Both measures are of high importance and should be implemented together, otherwise it may not have the desired final result. For example, promoting the increase in wild species may not reduce livestock consumption (Patalano and Lovari, 1993; Poulle *et al*., 1997) and the application of strict measures to prevent livestock consumption may create instability in the wolf population by not having access to other food items. In this way, both population reinforcement processes and mitigation measures, such as the effective protection of livestock, are envisaged as useful tools to reduce the likelihood of Human-Wolf conflicts.

Chapter II

Environmental determinants of roe deer presence and abundance

Roe deer detected by camera trap.

1. Abstract

The conservation of a predator species should consider the constant monitoring of prey species. In the Southern edge of Iberian wolf distribution, the presence of roe deer is recognized, however there is no updated information on its population. The aim of this study is to estimate the density and relative abundance of roe deer, update its fine-scale distribution and study the environmental determinants of its presence and abundance. Roe deer density was estimated using pellet group counts in linear transects with distance sampling. This method was complemented with the calculation of relative abundance index (RAI) using camera trap. The fieldwork was carried out between June 2019 and June 2020. Regarding pellet group counts, 310 transects were surveyed, which corresponds to a sampling effort of 59,400 meters. For camera trap method, 330 cameras were used in the analysis, corresponding to 11,546 camera-trap days. The density of roe deer estimated only for the Côa valley was 2,2 individuals/100ha (CI: 1,4-3,5 individuals/100ha). The overestimation of population density is possible due to sampling constraints (*e.g.* low probability of detection). Relative abundance data showed that the roe deer population varies across the study area, decreasing from Southern to Northern and Eastern to Western locations. The environmental characteristics that positively influenced the presence and abundance of the roe deer were valleys with moderate slopes and native forests (broadleaved and coniferous). Water availability and mean altitude only had a positive significant effect on the relative abundance of roe deer. Habitat heterogeneity and anthropogenic presence had no significant effect on habitat selection. Locations of great environmental favourability and with low density of roe deer are potential sites to apply measures aimed at reinforcing current roe deer populations. These areas are strategically located to enhance the natural expansion of existing and established populations. Future population reinforcements designed in places of environmental favourability will be a useful tool in the medium-long term for the expansion of roe deer south of the Douro river. The selected locations also overlap the current Iberian wolf range and could contribute for the stability of existing wolf packs by increasing the wild prey availability.

2. Introduction

The conservation of a predator species relies, among other things, on the conservation and regular monitoring of wild prey species. Some authors have suggested that predators select the habitat mainly according to prey distribution (Eggermann *et al*., 2011; Kittle *et al*., 2017; Kunkel and Pletscher, 2001). Therefore, understanding the factors that shape the selection of habitat by wild preys allows to forecast possible areas of predator occupation. The habitat selection and distribution of wild prey species is influenced by several factors. Environmental characteristics generally have great impact on prey distribution because they are related to the most essential needs: food (Heinze *et al*., 2011; Zweifel-Schielly *et al*., 2009) and cover (Ciuti *et al*., 2005; Eom *et al*., 2018; Mysterud and Østbye, 1999). These, in turn, are related to the reproduction and survival of the species (Parker *et al*., 2009). In this way, characteristics associated with the presence of food such as marshes, natural pastures and agricultural lands are selected by the ungulates (Laforge *et al*., 2016; Sorensen *et al*., 2015), as well as the presence of a developed tree and shrub layer that provides cover (Ewald *et al*., 2014; Torres *et al*., 2012b). Other characteristics such as proximity to water points (Carvalho *et al*., 2018; Wallach *et al*., 2007) and climatic conditions (Coulson *et al*., 2000; Richard *et al*., 2014) are also relevant for ungulate species due to physiological constraints and the influence on food availability.

There are two types of interspecific competition that can also alter the distribution of the prey community. Competition for resources may decreases the availability of food (Odadi *et al*., 2011) and/or space (Bartos *et al*., 2002). Competition for interference involves physical interaction such as aggression between the two species (Bartoš *et al*., 1996; Ferretti, 2011). In this way, one species is forced to alter its behaviour to avoid the other through the partition of resources (Darmon *et al*., 2012; Hall *et al*., 2018; Hibert *et al*., 2010; Macandza *et al*., 2012). The presence of the predator substantially alters the behaviour of prey by creating a landscape of fear (Khan *et al*., 2018). This effect may cause changes in the use of space (Latombe *et al*., 2014; Lingle, 2002), the period of the day in which they are active (Rossa *et al*., 2021) or generate changes in the aggregation level of the species (Creel and Winnie, 2005) and the surveillance time (Périquet *et al*., 2010). Spatial avoidance of predators is widely reported in several species and is one of the most effective anti-predatory behaviours in ungulates (Basille *et al*., 2015; Laundré, 2010; Yang *et al*., 2018).

Human presence can be compared to the presence of a top predator as species of ungulates tend to avoid places inhabited by humans (Buuveibaatar *et al*., 2016; Fortin *et al*., 2015). In addition to its presence, humans are commonly related with habitat disturbance and greater associated risks. Pollution and habitat fragmentation are examples of disturbances caused by humans with an effect on ungulates (Ciach and Fröhlich, 2019; Ito *et al*., 2013; Rodríguez-Estival *et al*., 2011). For this reason, human settlements, infrastructure and roads are usually avoided by the ungulates (Bojarska *et al*., 2020; M'soka *et al*., 2017; Schuette *et al*., 2016; Skarin and Alam, 2017). Roads, for instance, fragment the habitat and constitute a perceived risk of ungulate-vehicle collision (*e.g.* Zuberogoitia *et al*., 2014).

In this study area, the Iberian wolf, is an endangered species requiring several conservation efforts. These efforts may include adequate legal protection (Chapron *et al*., 2014; Chapron and López-Bao, 2016), prevent habitat fragmentation and degradation (Crooks *et al*., 2011; Di Minin *et al*., 2016), promote human tolerance (Treves and Bruskotter, 2014), reduce the impact of human activities (Sévêque *et al*., 2020) and promote the increase of wild preys (Torres *et al*., 2018; Vidal *et al*., 2018). The latter allows the restoration of trophic chains and, consequently, the conservation of top predators (Bakker and Svenning, 2018). One of the potential wild prey of the Iberian wolf in the territory is the roe deer. This species is ideal for population reinforcements due to its ecology, continued efforts and widespread acceptance.

Roe deer had limited distribution and low densities in Portugal during the 19th and 20th century mainly due to anthropogenic pressure (Salazar, 2009). During the 20th century, the rural exodus allowed the restoration of natural habitat (Salazar, 2009). With a more pronounced presence to the north of the Douro river, several reintroductions of roe deer to the south of this river began during the late 20th century (Salazar, 2009; Torres *et al*., 2015a). These reintroductions were aimed at increasing the population of roe deer for hunting and as prey for the

Iberian wolf (see Torres and Fonseca, 2016). These processes together with the natural expansion of the Spanish populations have contributed to the increase in population and distribution of roe deer in border regions. In the Western region south of the Douro River, the absence of roe deer (Torres *et al*., 2015a) and the presence of wolf packs (see Torres and Fonseca, 2016) culminated in reintroduction processes in the Arada and Montemuro mountains starting in 2013 and with annual releases by 2018 (table 1, Torres *et al*., 2015a). Currently, the roe deer density and distribution throughout the study area south of the Douro River is unknown. Therefore, it is necessary to determine the current distribution of the species as well as to estimate its relative abundance. This information is relevant for the study and conservation of the Iberian wolf in this area as the distribution of prey is a determining factor for its presence (Eggermann *et al*., 2011) and stability (Imbert *et al*., 2016). In addition to the distribution and abundance data, it is equally important to understand the key factors of habitat selection for roe deer (table 2) and to determine the environmental favourability of this study area for the species. The identification of favourable habitats where roe deer is absent or occurs at low densities will be important for the application of ecosystem restoration measures, such as the reinforcement of local populations. These measures aim to increase the availability of Iberian wolf prey and, therefore, their application will contribute to the conservation of this endangered predator. The present study aims to produce tools to promote the conservation of roe deer population by estimating the density and relative abundance of roe deer, update its fine-scale distribution and study the environmental determinants of its presence and abundance.

Table 2 - Hypothesis, rationale, predictions potentially influence the species presence and abundance at macro scale habitat.

Hypothesis	Rationale	Prediction
H1 (Topography)	Topography is a very important factor in the movements of the roe deer (López-Martín *et al*., 2009). Due to energy saving, valleys with middle to lower slopes and moderate altitudes are generally selected by roe deer (Farfán *et al*., 2009).	Valleys with gentle slopes and moderate altitudes are favourable for roe deer.
H2 (Water availability)	The availability of water positively influences roe deer dispersion (Carvalho *et al*., 2018) as it constitutes a physiological and behavioural restriction (Wallach *et al*., 2007).	The presence of roe deer is positively related to the availability of water. Especially in the Northeast of the study area, an area characterized by harsh winters and summer droughts.
H3 (Land cover)	Native forest areas are often selected by roe deer (Morellet *et al*., 2011; Virgós and Tellería, 1998). However, this ungulate also uses meadows and agricultural areas (Acevedo *et al*., 2005). Native forests provide refuge and food for roe deer (Cargnelutii *et al*., 2002).	Native forest areas are preferably selected by the roe deer.
H4 (Landscape structure)	The roe deer has preference for heterogeneous habitats (Saïd and Servanty, 2005) with great edge effect (Hewison *et al*., 2001). In this diversity of habitats, the roe deer is able to trade-off between the need for open habitats where it feeds and closed areas of refuge.	Preference for heterogeneous habitats.
H5 (Anthropogenic pressure)	The areas inhabited by humans are avoided by roe deer (Torres *et al*., 2012a) due to the disturbance of human settlements and roads (Coulon *et al*., 2008). Its behaviour is also influenced by hunting activity, avoiding hunting areas (Bonnot *et al*., 2013).	Roads, human settlements and hunting areas will be avoided by roe deer.

3. Materials and Methods

3.1. Data collection

The fieldwork was carried out on a monthly basis between June 2019 and June 2020 and comprised 10 campaigns. Approximately 35 cameras (Browning Strike Force Pro X) were used per campaign in order to update the fine-scale distribution of wild ungulates present in the study area. A total of 355 locations were sampled, however, due to theft (n = 10), malfunction (n = 9) or problems with the external driver (n = 6), the subsequent analyses were performed with data from 330 cameras (figure 7). Due to logistic and time constraints it was impossible to sample the entire study area, therefore representative locations of the main habitat types, areas of known Iberian wolf presence and areas of potential connectivity among packs were selected. The study area was divided in a 1,5 x 1,5 km grid that allowed to follow a regular sampling design. Despite the difficulties imposed by topography, dense shrubs and several fenced private areas, each camera was installed at a minimum of 1000-1500 meters and placed 30-40 cm aboveground to increase the species detectability. The distance between cameras was defined based on the minimum home range of roe deer (summer: 409.64 ± 98.20 ha, winter 116.20 ± 17.90 ha, Carvalho *et al*., 2008). The selected location had a good visibility, was free from dense vegetation (to avoid false triggers) and roads/paths were avoided. Whenever possible, cameras were north-oriented to reduce the number of unidentifiable pictures caused by the direct sunlight. A GPS disposal (Garmin Etrex 10) was used to record the exact location of cameras. The habitat characteristics (*e.g*. the habitat type, the landscape diversity and the level of anthropogenic disturbance) were also registered. The camera traps were programmed to take three consecutive photographs, with a recovery time of 30 seconds. This choice represents a trade-off between increasing detectability of individuals and storage capacity of SD-cards. The cameras were approximately 28 days in the field before being moved to new locations. Meanwhile, SD-cards and batteries were checked, changed as needed and the photos stored for subsequent identification.

The density of roe deer in the study area was estimated by performing one transect *per* sample location. Transects were 200 meters long and the orientation of each one was randomly selected but limited by topographic and land use/cover constraints. A handheld Global Positioning System (GPS) was used to record the starting and end points. The roe deer pellets groups were searched over the transect (approximately 1-m width for each side of the transect line, Marques *et al*., 2001). The perpendicular distance from the transect line to the centre of the pellet group was measured. Three variables that are expected to influence the detectability of pellets (Marques *et al*., 2007) were also recorded: the size of pellet groups (medium, 10-40 pellets; large, > 40 pellets; groups with less than 10 pellets were discarded to avoid duplications of scattered groups), the dispersion of pellet groups (aggregated or scattered) and the micro-habitat.

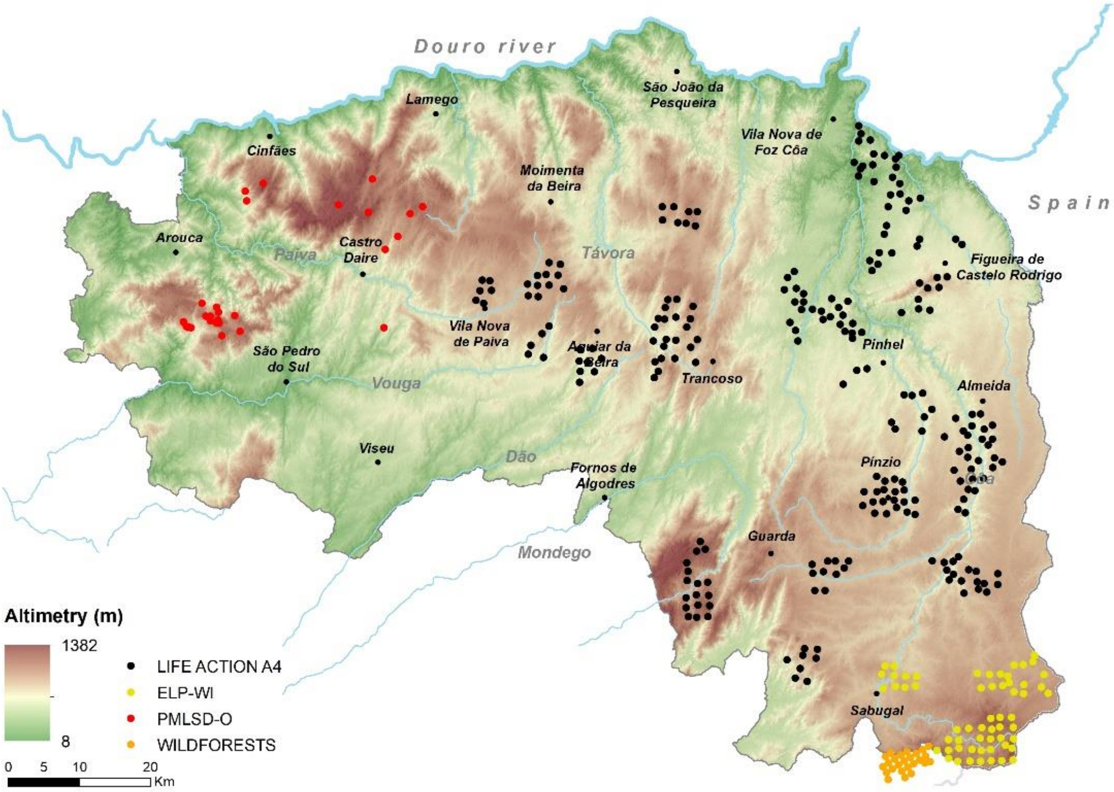


Figure 7 - Location of camera-traps and transects.

3.2. Data analysis

3.2.1. Roe deer density and distribution

From each photo, information regarding date, time, temperature, species, number of individuals and sex were recorded. All this information was organized and stored in a dataset. The photos of the same species in the same camera within a time window of 30 minutes (Monterroso *et al*., 2014; Torretta *et al*., 2017) were discarded. This procedure increases the likelihood of independence among records. The Relative Abundance Index (RAI) was calculated for each location. This index is calculated as the number of photographic records per species divided by the number of days the camera has been active. This value was standardized per unit of effort (x 100 camera days.) The values of RAI were used to map species distribution.

The pellet groups gathered from transects were analysed using the distance sampling method (Buckland *et al*., 2001). Distance sampling is a recommended and widely accepted approach to estimate species abundance and density from direct or indirect observation (*e.g.* pellets). Distance sampling allows to deal with uncertain detection by modelling the variation of detectability with distance through a detection function. The perpendicular distance from the pellet group to the centre of the transect are used to produce the detection function. This function allows to estimate the probability of locating pellet groups along the transect (Marques *et al*., 2001). The detection probability is used to calculate the density of pellet groups. The density is calculated using the number of pellet groups found, the probability of detection and the length of the sampled transect. Roe deer density is calculated by considering the density of pellet groups, the decay rate and the production rate of pellets. The decay rate is the number of days that a pellet group takes to disappear. In this study the value 176 ± 31 days, calculated by Torres *et al*. (2013) in Montesinho Natural Park, was used due to geographical proximity. The production rate is the number of pellet groups that an individual produces per day. The value used in this study was 20 groups per day (Mitchell *et al*., 1985). Data analysis was performed using the software Distance version 7.2 (Thomas *et al*., 2010). First, several models were fitted using different key functions and adjustment terms (Buckland *et al*., 2001). Second, the final detection function were selected by visually inspecting histograms and considering the Akaike Information Criterion (AIC, Akaike, 1974). The AIC ensures that the selected model is the most parsimonious.

3.2.2. Determinants of roe deer distribution and abundance

The favourability function has been used in studies where the probability of occurrence of an event (*e.g*. species) is analysed (Acevedo and Real, 2012). Species distribution models (SDMs) are one of the most common examples where the favourability function can be applied. SDMs combine species occurrences or abundance with a set of biotic/abiotic variables to describe the interactions between species distribution and environmental predictors (Carvalho *et al*., 2018; Elith and Leathwick, 2009). This tool became an important method to understand and manage ecological and conservation issues. Here, a continuous and spatially-explicit gradient of environmental favourability for roe deer was produced to identify potential areas for roe deer reintroduction or restocking. We selected predictor variables connected with the ecological requirements of the roe deer and known to influence the species' presence and abundance. These variables were grouped in a set of a hypothesis related to topographic features, water availability, land cover, landscape structure and anthropogenic pressure (table 3). Biotic variables accounting for intra and interspecific relationships (cattle, presence of other wild ungulate species) were discarded due to confounding effects (*e.g*. habitat), the low density of roe deer (Torres *et al*., 2015a) and its elusive behaviour. Predator effect was also discarded from the analysis due to low wolf presence in this area (Torres and Fonseca, 2016). Vegetation indexes, such as the Normalized Difference Vegetation Index (NDVI), were not included because the spectral response of tree canopy (that represents a significant proportion of the study area) does not reflect the quality and availability of understorey and accessible vegetation for roe deer. For modelling purposes, a total of 308 camera trapping locations (128 presences and 180 pseudoabsences) were considered. Based on fieldwork and species' home range (Carvalho *et al*., 2008), the environmental characteristics were retained considering a buffer of 800 meters

Table 3 – Hypothesis and variables connected with the ecology of roe deer and that potentially influence the species presence and abundance at macro scale habitat.

Hypothesis	Variable	Code	Pseudoabsence (mean)	Presence (mean)	Source
H1 (Topography)	Mean altitude (m)	AltMean	723	783	ASTER Global Digital Elevation Model platform
	Range altitude (m)	AltRange	142	119	
	Mean slope (º)	SlpMean	7.83	6.84	
	Range slope (º)	SlpRange	22.40	20.39	
	Mean topographic position index	TPIMean	0.50	0.50	
	Minimum topographic position index	TPIMin	0.26	0.26	
	Maximum topographic position index	TPIMax	0.74	0.75	
	Mean topographic ruggedness index	TRIMean	32.61	27.53	
	Minimum topographic ruggedness index	TRIMin	8.52	7.23	
	Maximum topographic ruggedness index	TRIMax	72.00	58.89	
	Outcrops (%)	OutcrPerc	46.28	40.86	Instituto Superior de Agronomia
	Terraces (%)	TerrPerc	11.98	12.88	
H2 (Water availability)	Streams density (km/km^2)	RivDens	0.82	0.85	Sistema Nacional de Informação de Recursos Hídricos
H3 (Land cover)	Agricultural areas (%)	AgrPerc	13.19	14.38	Carta de Uso e Ocupação do Solo - Direção-Geral do Território
	Native forests (%)	NativeForPerc	38.61	49.04	
	Eucalyptus stands (%)	ConifPerc	1.26	0.79	
	Orchards (%)	EucalPerc	6.50	2.88	
	Shrubs and herbaceous associations (%)	ShrubPerc	39.27	31.22	

	Burnt area (%)	BurntPerc	10.11	9.49	Instituto da Conservação da Natureza e das Florestas
H4 (Landscape structure)	Diversity of land cover patches (n)	PatchVar	4.79	4.56	Carta de Uso e Ocupação do Solo - Direção-Geral do Território
	Patches of native forests (n)	NForPatch	4.47	4.34	
	Mean area of forest patches (km^2)	MAForPatch	0.23	0.25	
H5 (Anthropogenic pressure)	Artificial surfaces (%)	HInfraPerc	2.91	4.07	
	Roads density (km/km^2)	RoadToDens	2.67	2.80	Geofabrik
	Primary roads density (km/km^2)	RoadPrDens	0.01	0.06	
	Secondary roads density (km/km^2)	RoadSeDens	0.11	0.14	
	Tertiary roads by square kilometer	RoadTeDens	0.26	0.30	
	Human population (inhab/km2)	PopDens	13.70	10.09	Eurostat
	Hunting areas (%)	HAreaPerc	88.29	87.31	Instituto da Conservação da Natureza e das Florestas

radius (area ≈ 2 km^2) from the sampling location. The study area was divided into a grid composed by individual units of 2,25 km^2 to project the model in space. The environmental domain of each individual grid was characterized.

First, an exploratory analysis was performed to assess the properties of data; the explanatory variables were examined to control for collinearity using a two-fold framework (Spearman's rank correlation test and variance inflation factors). The Spearman's rank correlation test assesses the collinearity among pairs of variables. Here, a coefficient of correlation of 0.8 was used. For multicollinearity, the variance inflation factors were used. Generalized linear models (GLM, McCullagh and Nelder, 1989) were used to describe the environmental determinants of roe deer presence and relative abundance. The roe deer presence is a binomial variable and a binomial GLM model was used. Relative abundance is a positive variable with several zeros and, therefore, Gamma distribution with a zero inflated function was used (Zuur *et al*., 2013). The explanatory variables were standardized to allow an easier comparison of effects sizes (Zuur *et al*., 2007). A forward–backward stepwise procedure based on the AIC was used to fit the final model. The data were split in two datasets: 70% of data was used to build the model and 30% to validate the model. The favourability function was used to make the probabilities of occurrence gathered from the GLM independent from the species' prevalence. By using the favourability function, it was possible to identify and map the regions that favour the species presence ($F \geq 0.5$) and the regions that do not present the favourable environmental characteristics for roe deer ($F < 0.5$).

For the micro-habitat characteristics analysis, the data collected in each location was used considering a buffer of 100 meters. Information regarding the type of habitat, ground cover (percentage of tree cover, shrub cover, herb cover and rocks cover), vegetation height (mean height of shrub and herbaceous layer: < 50cm, 50cm-1.5m and >1.5m) and anthropogenic pressure was used for the analysis. The type of habitat was classified as coniferous forests, eucalyptus, broadleaved forests, riparian forest, scrubland, agrosystem, unproductive areas and habitat mosaic. Anthropogenic pressure has been divided into 5 categories: recreational activities (hunting, tourism and BTT); agriculture (agriculture, resin collection, presence of tractors, and exploitation of eucalyptus); infrastructures (buildings and wind farms); presence of livestock and burnt areas. The selection of micro-habitat was performed using the same dataset used to fit the favourability function: roe deer presence (presence/absence data) and relative abundance (non-negative data). The collinearity control was performed using Spearman's rank correlation test and variance inflation factors. GLMs (McCullagh and Nelder, 1989) were also used to explain the roe deer presence and relative abundance through these micro-habitat characteristics. The GML analysis was performed using the previously applied tests for the macro-habitat, *i.e*. binomial GLM were used for presence/absence data and Gamma distribution with a zero inflated function for relative abundance data (Zuur *et al*., 2013). The final model was selected by analysing the AIC values.

Several reintroduction processes have occurred in this study area (table 1). For this reason, we hypothesized that the relative abundance of roe deer is related to the geographical proximity of the reintroduction sites. For this, the Euclidean distance between each location and the nearest reintroduction site was calculated. Additionally, the reintroduction sites were separated according to the date of reintroduction of the animals. Thus, the number of years after reintroduction was used as a factor in the analysis. In general, there are two groups of reintroductions: those performed 20-25 years ago and the most recent ones corresponding to Arada and Montemuro, which began 5 years ago and are still ongoing. Locations more than ~20 km distant from reintroduction sites and/or with severe geographical barriers were excluded from the reintroduction effect analysis. A GLM analysis was performed to assess the importance of reintroductions locations and antiquity on presence and relative abundance of roe deer.

4. Results

4.1. Roe deer density and distribution

Overall, a total of 1,686 detections of wild and domestic ungulates were recorded (roe deer: n = 789; wild boar: n = 657; red deer: n = 7; cattle: n = 94; sheep: n = 116; domestic goat: n = 23) for a sampling effort of 11,546 camera-trap days. The RAI for roe deer varies between 0 to 97.1 (figure 8). The higher values were recorded at the South-eastern region of the study area (Malcata). These results suggested that species abundance decreases toward Northern and Western locations. Roe deer detections in the West part were sporadic.

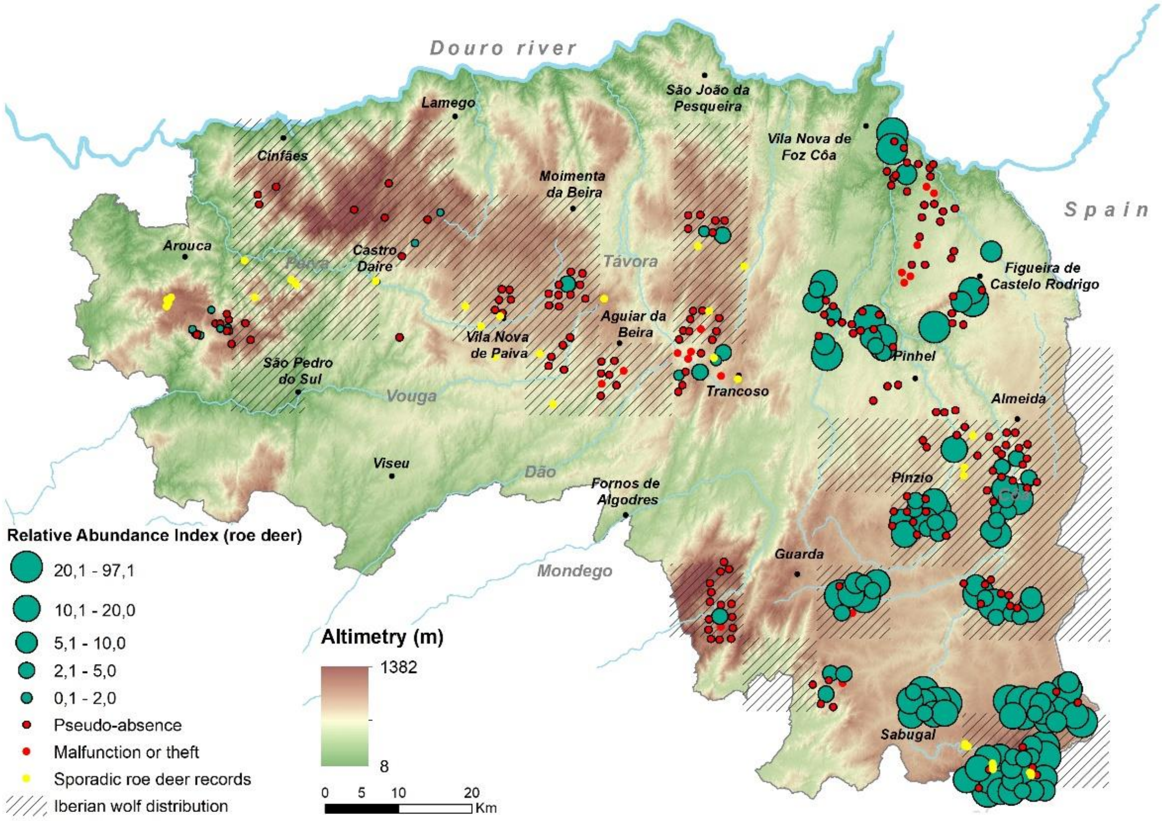


Figure 8 - Relative abundance index for roe deer based on camera trap data.

Over a total effort of 59,400 meters, 149 pellet groups were counted. The model that best fits the collected data was the half-normal with the cosine adjustment term (figure 9). The estimated roe deer population density was 2,2 individuals/100ha with a 95% confidence interval of 1,4-3,5 individuals/100ha and a coefficient of variation 24,2%. This value corresponds to the Côa valley, located in the eastern region of the study area. The low number of roe deer detections (direct or indirect records) in the central and Western areas did not allow to fill the minimum requirements to estimate the roe deer density for this area.

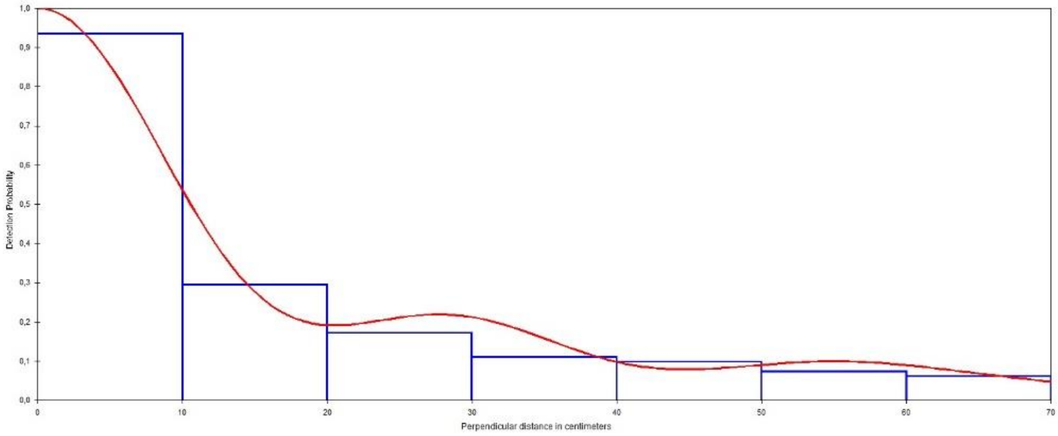


Figure 9 - Histogram of distance data considering a half-normal model with the cosine adjustment term. Observations were right-truncated to eliminate the largest 5 % portion.

4.2. Determinants of roe deer distribution and abundance

The final model retained 7 variables (AltMean, SlpMean, TPIMean, RoadTotDens, PopDens, NativeForPerc, PatchVar) from the initial set of 29 environmental predictors. The remained variables were excluded due to collinearity and/or to guarantee the parsimony of the fitted model. The most parsimonious model, *i.e.* with the lowest AIC, included variables related with topography, anthropogenic pressure, land cover and landscape structure, which indicated that roe deer presence is influenced by different environmental variables. Considering the statistically significant variables, the results supported H1 and H3 hypothesis, but there is no statistical evidence to support H2, H4 and H5 hypothesis (table 2). This analysis showed that roe deer presence is positively associated with the percentage of native forests (broadleaved and coniferous, βNativeForPerc = 0.30, SE = 0.13, z = 2.41, $p < 0.05$). The results also suggested that roe deer is more likely to occur in valleys (βTPIMean = -0.30, SE = 0.13, z = -2.25, $p < 0.05$) with middle or lower slopes (βSlpMean = -0.38, SE = 0.14, z = -2.66, $p < 0.01$). Neither mean altitude nor patch diversity played a significant role in roe deer presence. The variables accounting for the anthropogenic pressure (road and population density) were not significant (figure 10).

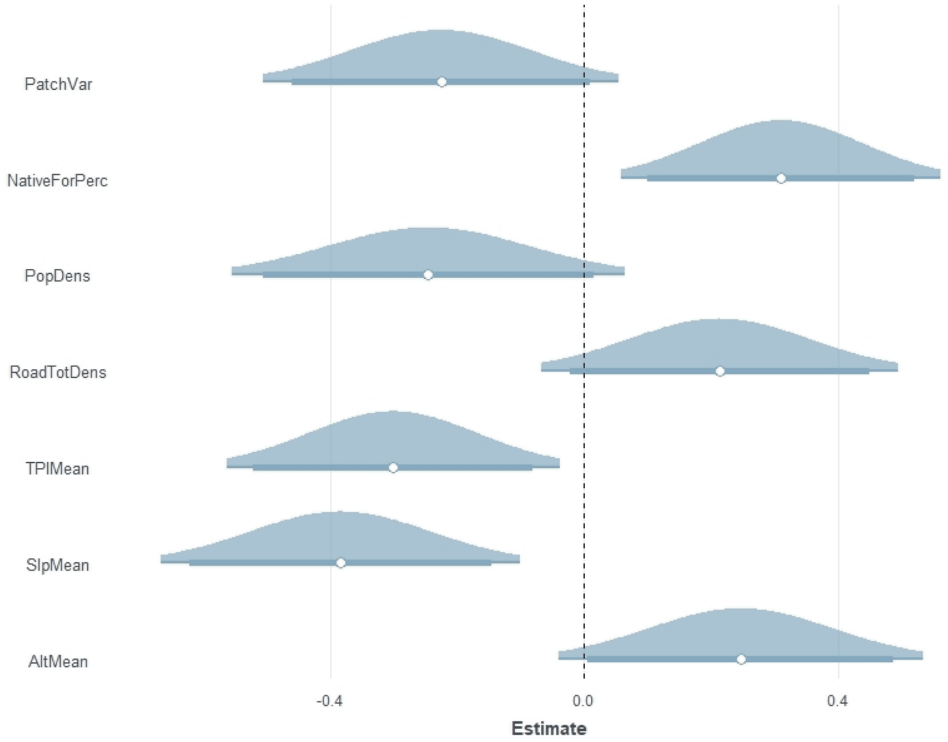


Figure 10 - Visual representation of variables coefficients that form the final GLM model.

Following the same approach, the analysis using the relative abundance index values allowed to obtain a final model with 8 variables (AltMean, SlpMean, TPIMean, RoadTotDens, PopDens, NativeForPerc, RivDens, PatchVar). Our results partially corroborate our hypothesis. The results supported H1, H2 and H3 hypothesis, but there is no statistical evidence to support H4 and H5 hypothesis (table 2). The relative abundance of roe deer is positively related to mean altitude (βAltMean = 0.14, SE = 0.03, z = 4.62, p < 0.001), the percentage of native forests (βNativeForPerc = 0.27, SE = 0.02, z = 11.25, p < 0.001) and the availability of water (βRivDens = 0.11, SE = 0.03, z = 3.66, p < 0.001). As for human disturbance, roads had a positive effect on the abundance of roe deer (βRoadTotDens = 0.09, SE = 0.03, z = 3.40, p < 0.001), while population density did not show a significant effect. The preference for valleys (βTPIMean = -0.35, SE = 0.03, z = -11.51, p < 0.05) with gentle slope (βSlpMean = -0.26, SE = 0.03, z = -9.15, p < 0.001) corroborated the previous analysis. Against our expectations, the heterogeneity of habitat had a negative effect on the abundance of roe deer (βPatchVar = -0.28, SE = 0.03, z = -10.43, p < 0.001).

The analysis of the microhabitat allowed us to corroborate the analysis of the macrohabitat and conclude that the roe deer occurs in broadleaved forests with a well-developed tree cover (βTreeCover = 0.27, SE = 0.02, z = 11.10, $p < 0.001$ characterised by an understorey composed of herbs (βHerbCover = 0.10, SE = 0.03, z = 3.13, $p < 0.01$).

Approximately 70% of the study area present favourable conditions ($F \geq 0.5$) for roe deer, but only 30% are highly favourable ($F \geq 0.7$). The figure 11 represents the spatial distribution of environmental favourableness for roe deer throughout the study area. The areas of high favourability are those located in the Central East and South-Eastern region, where roe deer density and abundance are already the highest throughout the study area (including some core areas previously identified in the LIFE WolFlux proposal). The Serra da Estrela and the Central region (Vila Nova de Paiva, Aguiar da Beira, Trancoso) also present continuous areas of high favourability (figure 11).

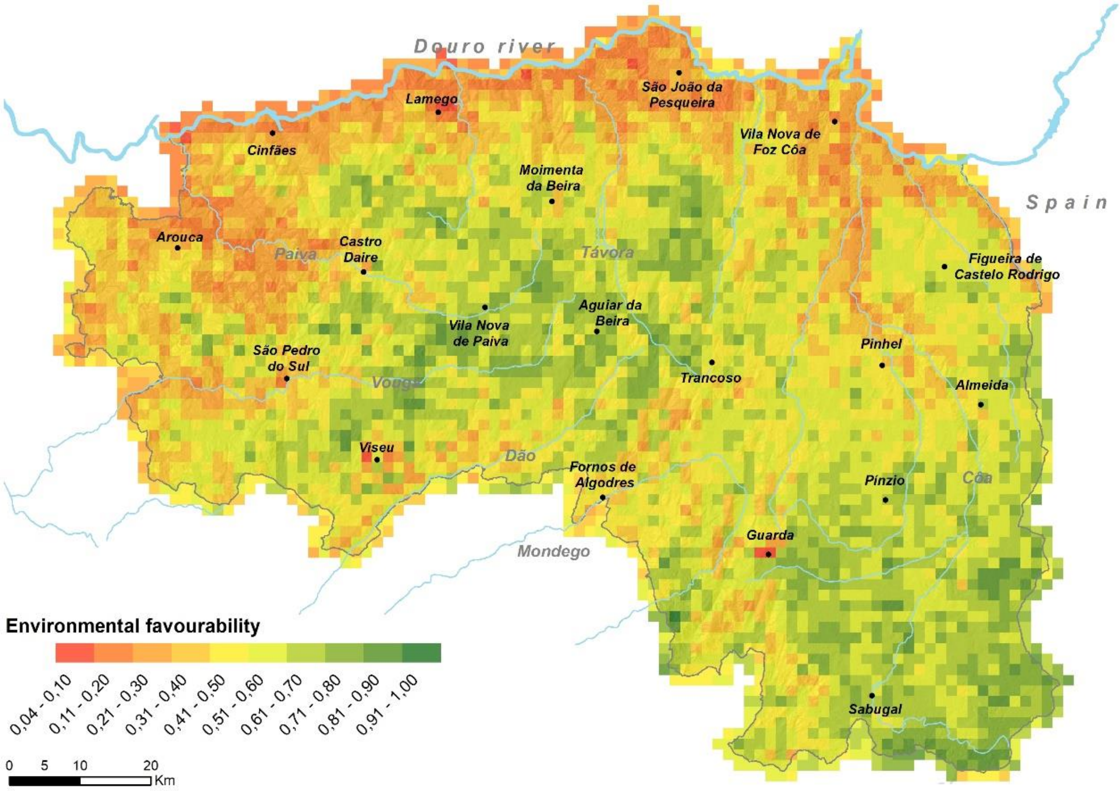


Figure 11 - Spatial gradient of environmental favourability for roe deer.

The distance to the reintroduction site had a negative effect on the abundance of roe deer (βDistReintr = -0.08, SE = 0.01 z = -15.70, p < 0.001), *i.e*. locations further away from the reintroduction areas had lower relative abundance index values. The years since the reintroduction process have had no significant effect on relative abundance.

5. Discussion

Less diverse ecosystems are more vulnerable to the propagation of disturbances along the trophic cascade (Calizza *et al*., 2019). Thus, the trophic chain south of the Douro river raises conservation concerns due to the low diversity of prey community and low density of wild prey populations (Torres and Fonseca, 2016). The constant assessment and monitoring of prey community is extremely important for the application of management and conservation actions. Information about the distribution, density and environmental favourability of roe deer is an essential tool for the conservation of roe deer populations and, consequently, its predator.

5.1. Roe deer density and distribution

In Portugal, several roe deer populations have high densities compared to the Côa valley population: Lousã mountain (3.10 ind./100 ha, CI95% = 1.6 – 5.9 ind./100 ha, Valente *et al*., 2017), Montesinho mountain (4.87 ind./100 ha, CI95% = 3.1 – 7.7 ind./100 ha, Valente *et al*., 2014) and Nogueira mountain (4.25 ind./100 ha, CI95% = 2.3 – 8,0 ind./100 ha, Valente *et al*., 2014). In this study, the density was only estimated for the Côa Valley area (North-eastern region of the study area) due to low detection of roe deer pellets in the Central and Western areas. This value must be analysed carefully due to a high set of factors. When few pellet groups are observed in relation to the number of transects surveyed and the sampled area, the density obtained must be interpreted with caution. The low probability of detection (25%) may leads to an overestimation of density (Valente *et al*., 2017). In this study, there is a great variation in the density of roe deer throughout the study area and, therefore, there are areas with higher densities and others with sporadic presence or absence of roe deer. The density calculation uses two conversion factors that can be sources of errors. The production rate used in this study was calculated for the United Kingdom (Mitchell *et al*., 1985) and is the only value available in the literature. The value of the decay rate was obtained in Montesinho (Torres *et al*., 2013), the geographically closest location with calculated values. Both rates are sources of error as they are not calculated in the same climatic conditions and both rates vary according to habitat. Despite the existence of these sources of error, the accuracy and precision of the density values can be improved through the sample design (Nomani *et al*., 2012). For this purpose, in this study, several short linear transects randomly distributed across the study area were used. Compared to few long transects, the use of several short transects reduces bias and increases the accuracy and precision of estimates (Nomani *et al*., 2012) and allow to reduce sampling dependence and maximize spatial coverage of the study area. Complementary to the study of density through distance sampling, a camera trap survey was used in order to understand the variation of roe deer abundance throughout the study area. Camera trap has been used to determine the density of animals using methods such as capture-recapture (Gardner *et al*., 2010; Heilbrun *et al*., 2006; Negrões *et al*., 2010), Random Encounter Model (REM; Gray, 2018; Romani *et al*., 2018; Rowcliffe *et al*., 2008) and an extension of this, the Random Encounter and Staying Time model (REST, Nakashima *et al*., 2020; Yokoyama *et al*., 2020). However, its application has been in areas smaller than this study area and with less sampling effort (*e.g*. Nakashima *et al*., 2020; Rovero and Marshall, 2009). Thus, Relative Abundance Index (RAI) is a good option as a complement to other methods (Foster and Harmsen, 2012; Pfeffer *et al*., 2018) and for large sampling areas. This method is easy to apply in the field and data processing is relatively simple even considering large amounts of data. Like all other methods that use camera trap, this method also has the disadvantage of not knowing the probability of detection (Jennelle *et al*., 2002; Sollmann *et al*., 2013) and its variation across the study area due to the characteristics of the habitat (Rovero and Zimmermann, 2016; Sollmann *et al*., 2013). The relative abundance should be interpreted with caution since sampling campaigns occurred at different locations and seasons. This can cause differences in roe deer behaviour due to site-specific conditions and life cycle (Fukuda *et al*., 2012). However, several studies have indicated a linear relationship between species density and RAI (Carbone *et al*., 2001; Rovero and Marshall, 2009; Rowcliffe *et al*., 2008). This makes RAI an easy method to apply and with significant results as a complement to other methods as in the present study. The results obtained through the RAI allowed to confirm an expansion of roe deer south of the Douro river. This is in agreement with the density data which indicate a high variability in the density of roe deer in this area. The roe deer population decreases towards Northern and Western areas. This is consistent with the presence of more abundant and stable populations in the East and South, due to the natural expansion of the Spanish population and populations reintroduced at the end of the 20th century (Torres *et al*., 2015a). More recently, reintroductions in the Western area, *i.e*. Freita and Montemuro mountains (Torres *et al*., 2015a), aim to counter the low density observed in this study.

5.2. Determinants of roe deer distribution and abundance

This study showed the importance of environmental variables such as land cover (H3), topography (H1), and water availability (H2) in the selection of habitat by roe deer. However, no statistical evidence was observed for the influence of anthropogenic pressure (H5) and landscape structure (H4) on the distribution of roe deer in this study area.

The forest plays a fundamental role at various levels in the life of roe deer: feeding (Gaudry *et al*., 2015; Moser *et al*., 2006), cover (Ewald *et al*., 2014) and bed sites (Bongi *et al*., 2008; Linnell *et al*., 1999). Several authors suggest that regardless of home range size, there is a minimum requirement for forest portion (Cargnelutii *et al*., 2002; Lovari and San José, 1997). Thus, it is expected that the forests are positively selected by roe deer as verified in this study. This pattern has also been reported in several studies (Acevedo *et al*., 2005; Farfán *et al*., 2009; Morellet *et al*., 2011). Moreover, the dispersion and genetic flow of roe deer are positively related to the presence of forests and their connectivity (Coulon *et al*., 2004). The vegetation species present in the forests also influence preference (Virgós and Tellería, 1998), for example native coniferous (Borkowski and Ukalska, 2008; López-Martín *et al*., 2009) and broadleaved forests (Oliveira and Carmo, 2000; Torres *et al*., 2011) are selected as noted in this study. However, even in altered landscapes such as pine plantations, roe deer shows preference for this type of forest (Hemami *et al*., 2004; Theuerkauf and Rouys, 2008), compensating for the anthropogenic pressure that may be subject.

Regarding the topography, valleys are the most positively selected feature by roe deer in this study area. However, only low to medium slopes are considered favourable. These results corroborate several studies that conclude that the roe deer avoids steep valleys (Coulon *et al*., 2008; López-Martín *et al*., 2009), with some reporting that these valleys are associated with falls and consequent mortality (Calenge *et al*., 2005). The roe deer use valleys and gentle slopes for natural expansion movements (Loro *et al*., 2016) or migration (Gaudry *et al*., 2015) probably as a way of saving energy (Cagnacci *et al*., 2011). Therefore, the difficulty of movement in steep valleys, associated with the risk of falling and energy savings seem to be the reasons for roe deer in this study area to select gentle valleys.

The altitude had no influence on the distribution of roe deer in this study area. In the Iberian Peninsula this trend had already been observed (Acevedo *et al*., 2005; Carvalho *et al*., 2018). However, in several situations where the seasonality of movements was studied, it was observed that in summer the roe deer tends to occupy higher areas which coincides with the increase in the home range and the possibility of being related to greater availability of food (Carvalho *et al*., 2008; Gaudry *et al*., 2015). Thus, the absence of a relationship between altitude and the distribution of roe deer can be justified by two factors. On the one hand, in our study area, the availability of food can be spatially distributed regardless of altitude. On the other hand, the presence of roe deer are not affected by some environmental factors such has altitude since they are able to use areas with less favourability (Acevedo *et al*., 2005). However, in places where roe deer is present, the mean altitude has a positive effect in explaining the relative abundance of roe deer. This may mean that when in expansion, the roe deer is not influenced by altitude, but when established it tends to occupy higher areas of the territory as moderate mountain ranges (López-Martín *et al*., 2009).

The roe deer usually avoids areas of strong human disturbance (Hewison *et al*., 2001; Oliveira and Carmo, 2000) namely structures such as buildings (Coulon *et al*., 2008; Torres *et al*., 2012a) and roads (Calenge *et al*., 2005; Torres *et al*., 2012b; Torres *et al*., 2011). However, in this study the effects of anthropogenic pressure on the distribution of roe deer were not observed, being in agreement with other studies (Acevedo *et al*., 2005; Carvalho *et al*., 2018; Torres *et al*., 2010). This pattern can be justified by the low human population density in the study area. In fact, Aragón *et al*. (1995) showed that roe deer was associated with areas of low human pressure.

Habitat heterogeneity such as mosaics of agricultural land and forests (Acevedo *et al*., 2005) or the use of clearings within forests (Kuijper *et al*., 2009) is commonly selected by this species of deer. The preference for agricultural fields or pastures is related to the greater availability of food (Bouyer *et al*., 2015; Farfán *et al*., 2009), however, these can be sources of human disturbance and, therefore, avoided by roe deer for this reason (Oliveira and Carmo, 2000). In this way, the heterogeneity of the landscape appears as a solution to the trade-off between availability of food offered by agricultural fields or pastures and forest areas that serve as a refuge. Several studies have revealed the importance and selection of forest edge areas (Hewison *et al*., 2001; Saïd and Servanty, 2005). The higher density of forest edge leads to greater contact between forest-open field and, therefore, better habitat for the roe deer (Saïd and Servanty, 2005). In this study the heterogeneity of habitat was not a decisive factor in the selection of roe deer habitat, corroborating the results of other studies (Torres *et al*., 2012b; Torres *et al*., 2010). This may be related to the fact that it is an expanding population and uses less favourable habitats. Acevedo *et al*. (2005) also observed that in expansion processes, the roe deer may slightly alter habitat preferences.

The availability of water is of great importance for the physiology and behaviour of roe deer (Wallach *et al*., 2007). In this way we expected it influences the habitat selection of the roe deer. This prediction was only partially confirmed: the availability of water had no significant influence on the distribution of roe deer, however, the relative abundance was positively correlated with the availability of water. This difference in habitat selection may be related to the quality of the surrounding habitat. Wallach *et al*. (2007) observed that in non-preferred habitats, roe deer did not change the amount of water consumed but did change the frequency of visitation of the habitat. The importance of water availability had already been observed in Portugal (Carvalho *et al*., 2018) and Spain even if related to precipitation (Acevedo *et al*., 2005). The study of climatic conditions and, specifically, precipitation and snow depth, is also relevant for the selection of habitat by roe deer (Acevedo *et al*., 2011; Aragón *et al*., 1995; Bouyer *et al*., 2015; Ewald *et al*., 2014; Farfán *et al*., 2009; Ossi *et al*., 2015). These conditions are related to the seasons, therefore, changes in habitat selection are expected depending on the seasons (Gaudry *et al*., 2015; Morellet *et al*., 2013, 2011; Mysterud *et al*., 1999). Unfortunately, both climatic conditions and seasonal patterns could not be analysed in this study. Future studies should use these variables in their analysis.

Environmental favourability was mapped using the habitat characteristics selected by roe deer (figure 12). This information is useful for wildlife management plans and future landscape conservation actions. Together with the knowledge of the relative abundance of roe deer, it is possible to select areas of low abundance and moderate-to-high environmental favourability. These areas are important for conservation and are possible sites for roe deer population reinforcement.

Distance to source populations is a determining factor when it comes to the expansion of a species (Acevedo *et al*., 2005). In this case, we studied the effect of the distance to the sites of roe deer reintroductions in relative abundance of roe deer (table 1, Torres *et al*., 2015a). The results have shown that there is a higher relative abundance of roe deer near the reintroduction sites. This pattern was observed in Spain where the proximity to the nearest known population had a significant effect on the relative abundance of roe deer (Acevedo *et al*., 2005; Torres *et al*., 2016). These results suggest that the reintroductions carried out in this study area were successful and the selected reintroduction sites have favourable conditions for roe deer (Cruz *et al*., 2014). In this way, this study demonstrates the importance and effectiveness of reintroduction processes in roe deer population. Therefore, future reintroductions or population reinforcements are useful tools from which we can expect results in the medium to long term.

6. Conclusions and management implications

Roe deer south of the Douro River is expanding and the relative abundance decreases toward Northern and Western locations. Native forests with undercover composed by herbs were selected as well as valleys with moderate slopes. The availability of water also positively influenced the relative abundance of roe deer. Thus, it was possible to perceive the selection of habitats with roe deer presence data, however we advise the use of more explanatory indexes, such as the relative abundance index. The latter is equally easy to apply and abundance data can illustrate the different degrees of preference (Fukuda *et al*., 2012) and, therefore, show influences that may be masked by the presence-absence data. Thus, relative abundance data has added essential knowledge for understanding roe deer habitat selection by supporting one more hypothesis (H2) than the presence-absence data.

The study of sites with favourable habitat allows the development of conservation measures and population reinforcement processes. The reintroduction processes carried out in this area of study so far have proved to be successful and the relative abundances of roe deer are higher near these sites. In this way, future population reinforcements designed in places of environmental favourability will be a useful tool in the medium-long term for the expansion of roe deer south of the Douro river and for increasing the availability of wild prey of Iberian wolf.

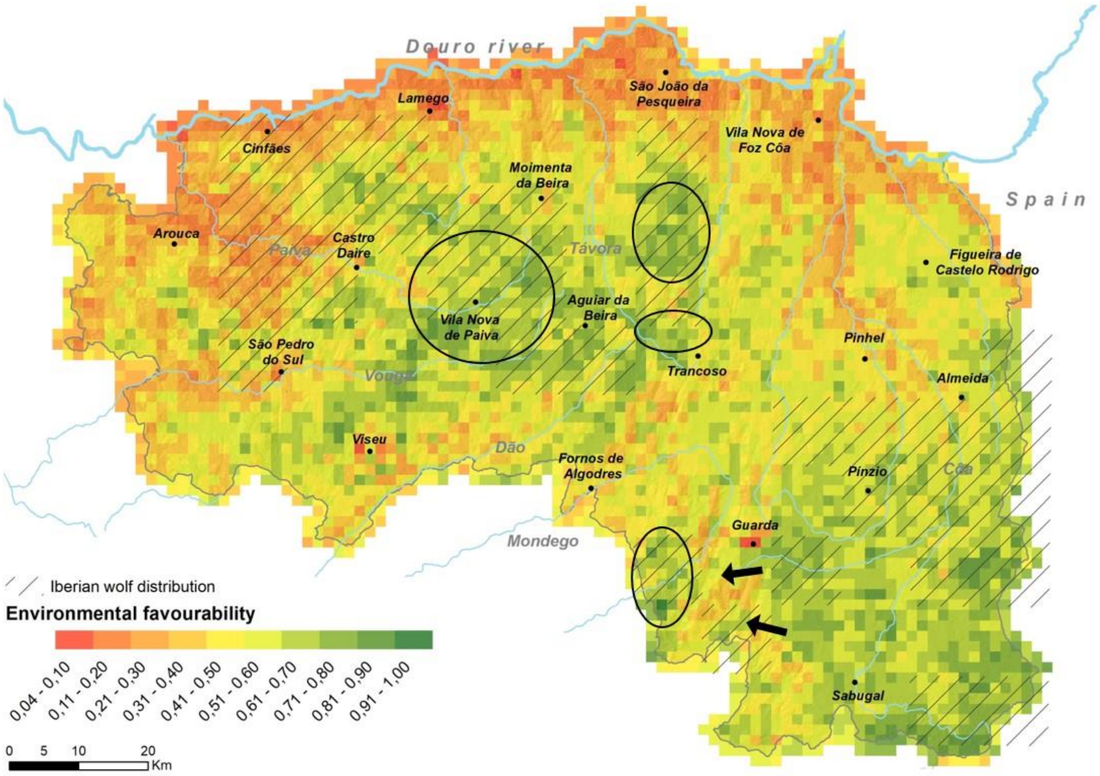


Figure 12 - Environmental favourability for roe deer with indication of potential areas for species reintroduction/restocking (black circles) and pathways that should be explored to foster the spread of existing population s and recently population nuclei (black arrows).

General Conclusions

The restoration of a trophic chain is a complex process that must cover several areas of knowledge and must be implemented over time without abruptly removing or providing a resource. We must knowledge the ecological, economic and social situation of the study area to make management measures effective and fit to conservation (IUCN/SSC, 2013). This study showed that the wolves south of the Douro river feed mainly on livestock, corroborating other studies carried out in altered and unbalanced ecosystems (Hosseini-Zavarei *et al*., 2013; Iliopoulos *et al*., 2009; Meriggi *et al*., 1996, 1991). This ecological unbalance in particular can lead to Human-Wolf conflicts (Anthony and Tarr, 2019). In Freita, Arada and Montemuro mountains, efforts to restore the trophic chain have already begun in 2013 through annual reintroductions of roe deer (Torres *et al*., 2015a). It was observed in the present study the consumption of roe deer, which indicates the success of reintroductions through the increase in the availability of wild prey. It is expected that the percentage of roe deer in the Iberian wolf's diet will increase with the slight increase in roe deer population.

Promoting the availability of prey in this area requires detailed knowledge of the distribution and relative abundance of prey species. With this study it was possible to verify that the roe deer population has a variable presence with greater abundance in the South and East of the study area. The selection of environmental characteristics such as valleys with moderate slope and native forests allowed the development of a map of environmental favourability for the roe deer. This map is a useful tool for the identification of areas of conservation importance and suitable locations for population reinforcements in order to promote the expansion of roe deer population. With this study it was also possible to see that sites of former reintroductions remain with moderate relative abundances indicating that they were successful processes.

Thus, in this work it was studied the possible challenges to the restoration of the trophic chain such as the high consumption of livestock in the region, to understand how the roe deer is distributed in the area and which habitat characteristics are selected. Useful tools for the management of the roe deer population were produced, such as I) maps of distribution and relative abundance that show the current state of roe deer population and II) environmental favourability maps for future conservation actions and population reinforcement.

The great opportunity shown in this study is the success of reintroductions. The present study indicated that the reintroduction processes carried out in this study area were successful through the presence of roe deer in the diet of wolves in Western region and the greater relative abundance of roe deer near the former reintroduction sites. In addition, the selection of reintroduction sites (see Cruz *et al*., 2014) has been adequate due to the presence of the roe deer near these sites after several years. In this way, we can expect that future population reinforcements will be a useful medium-long term conservation tool and that it will increase the availability of wild prey of Iberian wolf. With the gradual expansion and abundance of the roe deer population, it is expected to increase its consumption by Iberian wolf and possibly decrease the consumption of livestock (Imbert *et al*., 2016; Meriggi and Lovari, 1996). Thus, although this is the beginning of a long process, we can consider the restoration of the trophic chain as a viable solution to the present problem of conservation of the Iberian wolf south of the Douro river.

Next steps

The results of the diet should support the creation of management actions towards the Iberian wolf conservation. Measures to decrease livestock consumption should consist of promoting the increase and expansion of wild prey such as reintroduction/population reinforcement processes and effective livestock protection measures. Attacks can be mitigated through the use of fences, presence of a shepherd and/or guarding dog, gathering livestock overnight in barns. The simultaneous application of both measures should decrease the consumption of livestock in this study area. Through the map of environmental favourability, we can determine the appropriate sites for the population reinforcement processes. These sites are characterized by low density of roe deer and a set of habitat characteristics that the roe deer selected in this study area. In order to continue this process, it is necessary to have a debate between stakeholders such as local people, hunters, researchers and police makers.

The next challenge for the restoration of this trophic chain is related to the barriers to expansion and connectivity of the Iberian wolf population. South of the Douro river, the packs are isolated and have a small number of individuals, this is a source of reproductive instability due to the low gene flow (see Torres and Fonseca, 2016). To overcome this problem, it is not only necessary to restore the trophic chain by increasing prey population, but also to identify social and geographical barriers to the Iberian wolf. Social barriers are related to human's perception of the wolf (Behr *et al*., 2017). Through sociological studies we can identify areas according to the level of acceptance of the Iberian wolf (Behr *et al*., 2017). Several geographical barriers have been described and can be of natural (*e.g.* rivers, mountains; Blanco *et al*., 2005) or human origin (*e.g*. roads, infrastructures; Albayrak, 2011; Szewczyk *et al*., 2019). The identification of these barriers will serve as a tool for conservation projects to mitigate these barriers and promote connectivity among packs.

References

Acevedo, P., Delibes-Mateos, M., Escudero, M.A., Vicente, J., Marco, J., Gortazar, C., 2005. Environmental constraints in the colonization sequence of roe deer (Capreolus capreolus Linnaeus, 1758) across the Iberian Mountains, Spain. J. Biogeogr. 32, 1671–1680. https://doi.org/10.1111/j.1365-2699.2005.01310.x

Acevedo, P., Real, R., 2012. Favourability: concept, distinctive characteristics and potential usefulness. Naturwissenschaften 99, 515–522. https://doi.org/10.1007/s00114-012-0926-0

Acevedo, P., Real, R., Gortázar, C., 2011. Favorabilidad ecogeográfica para el corzo: distribución y abundancia. Pirineos 166, 9–27. https://doi.org/10.3989/pirineos.2011.166001

Akaike, H., 1974. A new look at the statistical model identification. IEEE Trans. Automat. Contr. 19, 716–723. https://doi.org/10.1109/TAC.1974.1100705

Albayrak, T., 2011. Anthropogenic barriers to the distribution of the Grey Wolf (Canis lupus Linnaeus, 1758) in the Beydağları Mountains area, Turkey. Zool. Middle East 52, 11–16. https://doi.org/10.1080/09397140.2011.10638474

Andersen, R., Linnell, J.D.C., Solberg, E.J., 2010. The future role of large carnivores in terrestrial trophic interactions: the northern temperate view, in: Danell, K., Bergstrom, R., Duncan, P., Pastor, J. (Eds.), Large Herbivore Ecology, Ecosystem Dynamics and Conservation. Cambridge University Press, Cambridge, pp. 413–448. https://doi.org/10.1017/CBO9780511617461.016

Andersone, Ž., Ozolinš, J., 2004. Public perception of large carnivores in Latvia. Ursus 15, 181–187. https://doi.org/10.2192/1537-6176(2004)015<0181:PPOLCI>2.0.CO;2

Anthony, B.P., Tarr, K., 2019. The wolves are back! Local attitudes towards the recently re-populated grey wolf and wolf management in Bükk National Park, Hungary. Acta Zool. Acad. Sci. Hungaricae 65, 195–214. https://doi.org/10.17109/AZH.65.2.195.2019

Aragón, S., Braza, F., San Jose, C., 1995. Socioeconomic, physiognomic, and climatic factors determining the distribution pattern of roe deer Capreolus capreolus in Spain. Acta Theriol. (Warsz). 40, 37–43. https://doi.org/10.4098/AT.arch.95-4

Athreya, V., Isvaran, K., Odden, M., Linnell, J.D.C., Kshettry, A., Krishnaswamy, J., Karanth, U.K., 2020. The impact of leopards (Panthera pardus) on livestock losses and human injuries in a human-use landscape in Maharashtra, India. PeerJ 8. https://doi.org/10.7717/peerj.8405

Athreya, V., Odden, M., Linnell, J.D.C., Krishnaswamy, J., Karanth, U., 2013. Big Cats in Our Backyards: Persistence of Large Carnivores in a Human Dominated Landscape in India. PLoS One 8. https://doi.org/10.1371/journal.pone.0057872

Aubry, T., 2009. 200 séculos da história do Vale do Côa: incursões na vida quotidiana dos caçadores-artistas do Paleolítico. IGESPAR.

Bakker, E.S., Svenning, J.-C., 2018. Trophic rewilding: impact on ecosystems under global change. Philos. Trans. R. Soc. B Biol. Sci. 373, 20170432. https://doi.org/10.1098/rstb.2017.0432

Barja, I., 2009. Prey and Prey-Age Preference by the Iberian Wolf Canis Lupus Signatus in a Multiple-Prey Ecosystem. Wildlife Biol. 15, 147–154. https://doi.org/10.2981/07-096

Bartos, L., Vankova, D., Miller, K. V., Siler, J., 2002. Interspecific Competition between White-Tailed, Fallow, Red, and Roe Deer. J. Wildl. Manage. 66, 522–527. https://doi.org/10.2307/3803185

Bartoš, L., Vaňková, D., Šiler, J., Losos, S., 1996. Fallow deer tactic to compete over food with red deer. Aggress. Behav. 22, 375–385. https://doi.org/10.1002/(SICI)1098-2337(1996)22:5<375::AID-AB6>3.0.CO;2-I

Basille, M., Fortin, D., Dussault, C., Bastille-Rousseau, G., Ouellet, J.-P., Courtois, R., 2015. Plastic response of fearful prey to the spatiotemporal dynamics of predator distribution. Ecology 96, 2622–2631. https://doi.org/10.1890/14-1706.1

Bautista, C., Revilla, E., Naves, J., Albrecht, J., Fernández, N., Olszańska, A., Adamec, M., Berezowska-Cnota, T., Ciucci, P., Groff, C., Härkönen, S., Huber, D., Jerina, K., Jonozovič, M., Karamanlidis, A.A., Palazón, S., Quenette, P.-Y., Rigg, R., Seijas, J., Swenson, J.E., Talvi, T., Selva, N., 2019. Large carnivore damage in Europe: Analysis of compensation and prevention programs. Biol. Conserv. 235, 308–316. https://doi.org/10.1016/j.biocon.2019.04.019

Behr, D.M., Ozgul, A., Cozzi, G., 2017. Combining human acceptance and habitat suitability in a unified socio-ecological suitability model: a case study of the wolf in Switzerland. J. Appl. Ecol. 54, 1919–1929. https://doi.org/10.1111/1365-2664.12880

Berger, K.M., Gese, E.M., Berger, J., 2008. Indirect effects and traditional trophic cascades: a test involving wolves, coyotes, and pronghorn. Ecology 89, 818–828. https://doi.org/10.1890/07-0193.1

Blanco, J.C., Cortés, Y., Virgós, E., 2005. Wolf response to two kinds of barriers in an agricultural habitat in Spain. Can. J. Zool. 83, 312–323. https://doi.org/10.1139/z05-016

Blondel, J., Aronson, J., 1999. Biology and wildlife of the Mediterranean region. Oxford University Press, USA.

Bojarska, K., Kurek, K., Śnieżko, S., Wierzbowska, I., Król, W., Zyśk-Gorczyńska, E., Baś, G., Widera, E., Okarma, H., 2020. Winter severity and anthropogenic factors affect spatial behaviour of red deer in the Carpathians. Mammal Res. 65, 815–823. https://doi.org/10.1007/s13364-020-00520-z

Bongi, P., Ciuti, S., Grignolio, S., Del Frate, M., Simi, S., Gandelli, D., Apollonio, M., 2008. Anti-predator behaviour, space use and habitat selection in female roe deer during the fawning season in a wolf area. J. Zool. 276, 242–251. https://doi.org/10.1111/j.1469-7998.2008.00481.x

Bonnot, N., Morellet, N., Verheyden, H., Cargnelutti, B., Lourtet, B., Klein, F., Hewison, A.J.M., 2013. Habitat use under predation risk: hunting, roads and human dwellings influence the spatial behaviour of roe deer. Eur. J. Wildl. Res. 59, 185–193. https://doi.org/10.1007/s10344-012-0665-8

Borkowski, J., Ukalska, J., 2008. Winter habitat use by red and roe deer in pine-dominated forest. For. Ecol. Manage. 255, 468–475. https://doi.org/10.1016/j.foreco.2007.09.013

Bosch, J., Peris, S., Fonseca, C., Martinez, M., Torre, A.D. la, Iglesias, I., Muñoz, M.J., 2012. Distribution, abundance and density of the wild boar on the Iberian Peninsula, based on the CORINE program and hunting statistics. Folia Zool. 61, 138–151. https://doi.org/10.25225/fozo.v61.i2.a7.2012

Bouyer, Y., Rigot, T., Panzacchi, M., Moorter, B. Van, Poncin, P., Beudels-Jamar, R., Odden, J., Linnell, J.D.C., 2015. Using Zero-Inflated Models to Predict the Relative Distribution and Abundance of Roe Deer Over Very Large Spatial

Scales. Ann. Zool. Fennici 52, 66–76. https://doi.org/10.5735/086.052.0206

Buckland, S.T., Anderson, D.R., Burnham, K.P., Laake, J.L., Borchers, D.L., Thomas, L., 2001. Introduction to distance sampling. Oxford University Press, Oxford.

Bump, J.K., Peterson, R.O., Vucetich, J.A., 2009. Wolves modulate soil nutrient heterogeneity and foliar nitrogen by configuring the distribution of ungulate carcasses. Ecology 90, 3159–3167. https://doi.org/10.1890/09-0292.1

Buuveibaatar, B., Mueller, T., Strindberg, S., Leimgruber, P., Kaczensky, P., Fuller, T.K., 2016. Human activities negatively impact distribution of ungulates in the Mongolian Gobi. Biol. Conserv. 203, 168–175. https://doi.org/10.1016/j.biocon.2016.09.013

Cagnacci, F., Focardi, S., Heurich, M., Stache, A., Hewison, A.J.M., Morellet, N., Kjellander, P., Linnell, J.D.C., Mysterud, A., Neteler, M., Delucchi, L., Ossi, F., Urbano, F., 2011. Partial migration in roe deer: migratory and resident tactics are end points of a behavioural gradient determined by ecological factors. Oikos 120, 1790–1802. https://doi.org/10.1111/j.1600-0706.2011.19441.x

Calenge, C., Maillard, D., Invernia, N., Gaudin, J.-C., 2005. Reintroduction of roe deer Capreolus capreolus into a Mediterranean habitat: female mortality and dispersion. Wildlife Biol. 11, 153–161. https://doi.org/10.2981/0909-6396(2005)11[153:RORDCC]2.0.CO;2

Calizza, E., Rossi, L., Careddu, G., Sporta Caputi, S., Costantini, M.L., 2019. Species richness and vulnerability to disturbance propagation in real food webs. Sci. Rep. 9. https://doi.org/10.1038/s41598-019-55960-8

Capitani, C., Bertelli, I., Varuzza, P., Scandura, M., Apollonio, M., 2004. A comparative analysis of wolf (Canis lupus) diet in three different Italian ecosystems. Mamm. Biol. 69, 1–10. https://doi.org/10.1078/1616-5047-112

Carbone, C., Christie, S., Conforti, K., Coulson, T., Franklin, N., Ginsberg, J.R., Griffiths, M., Holden, J., Kawanishi, K., Kinnaird, M., Laidlaw, R., Lynam, A., Macdonald, D.W., Martyr, D., McDougal, C., Nath, L., O'Brien, T., Seidensticker, J., Smith, D.J.L., Sunquist, M., Tilson, R., Shahruddin, W.N., 2001. The use of photographic rates to estimate densities of tigers and other cryptic mammals. Anim. Conserv. 4, 75–79.

https://doi.org/10.1017/S1367943001001081

Carey, M.P., Sanderson, B.L., Barnas, K.A., Olden, J.D., 2012. Native invaders – challenges for science, management, policy, and society. Front. Ecol. Environ. 10, 373–381. https://doi.org/10.1890/110060

Cargnelutii, B., Reby, D., Desneux, L., Angibault, J.-M., Joachim, J., Hewison, A.J.M., 2002. Space use by roe deer in a fragmented landscape some preliminary results. Rev. d'Ecologie (La Terre la Vie) 57, 29–37.

Carvalho, J., Torres, R.T., Acevedo, P., Santos, J.P. V., Barros, T., Serrano, E., Fonseca, C., 2018. Propagule pressure and land cover changes as main drivers of red and roe deer expansion in mainland Portugal. Divers. Distrib. 24, 551–564. https://doi.org/10.1111/ddi.12703

Carvalho, P., Nogueira, A.J.A., Soares, A.M.V.M., Fonseca, C., 2008. Ranging behaviour of translocated roe deer in a Mediterranean habitat: seasonal and altitudinal influences on home range size and patterns of range use. Mammalia 72, 89–94. https://doi.org/10.1515/MAMM.2008.019

Chapron, G., Kaczensky, P., Linnell, J.D.C., von Arx, M., Huber, D., Andrén, H., López-Bao, J.V., Adamec, M., Álvares, F., Anders, O., Balčiauskas, L., Balys, V., Bedő, P., Bego, F., Blanco, J.C., Breitenmoser, U., Brøseth, H., Bufka, L., Bunikyte, R., Ciucci, P., Dutsov, A., Engleder, T., Fuxjäger, C., Groff, C., Holmala, K., Hoxha, B., Iliopoulos, Y., Ionescu, O., Jeremić, J., Jerina, K., Kluth, G., Knauer, F., Kojola, I., Kos, I., Krofel, M., Kubala, J., Kunovac, S., Kusak, J., Kutal, M., Liberg, O., Majić, A., Männil, P., Manz, R., Marboutin, E., Marucco, F., Melovski, D., Mersini, K., Mertzanis, Y., Mysłajek, R.W., Nowak, S., Odden, J., Ozolins, J., Palomero, G., Paunović, M., Persson, J., Potočnik, H., Quenette, P.-Y., Rauer, G., Reinhardt, I., Rigg, R., Ryser, A., Salvatori, V., Skrbinšek, T., Stojanov, A., Swenson, J.E., Szemethy, L., Trajçe, A., Tsingarska-Sedefcheva, E., Váňa, M., Veeroja, R., Wabakken, P., Wölfl, M., Wölfl, S., Zimmermann, F., Zlatanova, D., Boitani, L., 2014. Recovery of large carnivores in Europe's modern human-dominated landscapes. Science. 346, 1517–1519. https://doi.org/10.1126/science.1257553

Chapron, G., López-Bao, J.V., 2016. Coexistence with Large Carnivores Informed by Community Ecology. Trends Ecol. Evol. 31, 578–580.

https://doi.org/10.1016/j.tree.2016.06.003

Chavez, A.S., Gese, E.M., 2005. Food habits of wolves in relation to livestock depredations in northwestern Minnesota. Am. Midl. Nat. 154, 253–263. https://doi.org/10.1674/0003-0031(2005)154[0253:FHOWIR]2.0.CO;2

Ciach, M., Fröhlich, A., 2019. Ungulates in the city: light pollution and open habitats predict the probability of roe deer occurring in an urban environment. Urban Ecosyst. 22, 513–523. https://doi.org/10.1007/s11252-019-00840-2

Ciuti, S., Bongi, P., Vassale, S., Apollonio, M., 2005. Influence of fawning on the spatial behaviour and habitat selection of female fallow deer (Dama dama) during late pregnancy and early lactation. J. Zool. 268, 97–107. https://doi.org/10.1111/j.1469-7998.2005.00003.x

Coulon, A., Cosson, J.F., Angibault, J.M., Cargnelutti, B., Galan, M., Morellet, N., Petit, E., Aulagnier, S., Hewinson, A.J.M., 2004. Landscape connectivity influences gene flow in a roe deer population inhabiting a fragmented landscape: an individual-based approach. Mol. Ecol. 13, 2841–2850. https://doi.org/10.1111/j.1365-294X.2004.02253.x

Coulon, A., Morellet, N., Goulard, M., Cargnelutti, B., Angibault, J.-M., Hewison, A.J.M., 2008. Inferring the effects of landscape structure on roe deer (Capreolus capreolus) movements using a step selection function. Landsc. Ecol. 23, 603–614. https://doi.org/10.1007/s10980-008-9220-0

Coulson, T., Milner–Gulland, E.J., Clutton–Brock, T., 2000. The relative roles of density and climatic variation on population dynamics and fecundity rates in three contrasting ungulate species. Proc. R. Soc. London. Ser. B Biol. Sci. 267, 1771–1779. https://doi.org/10.1098/rspb.2000.1209

Creel, S., Winnie, J.A., 2005. Responses of elk herd size to fine-scale spatial and temporal variation in the risk of predation by wolves. Anim. Behav. 69, 1181–1189. https://doi.org/10.1016/j.anbehav.2004.07.022

Cromsigt, J.P.G.M., te Beest, M., Kerley, G.I.H., Landman, M., le Roux, E., Smith, F.A., 2018. Trophic rewilding as a climate change mitigation strategy? Philos. Trans. R. Soc. B Biol. Sci. 373. https://doi.org/10.1098/rstb.2017.0440

Crooks, K.R., Burdett, C.L., Theobald, D.M., Rondinini, C., Boitani, L., 2011. Global patterns of fragmentation and connectivity of mammalian carnivore habitat. Philos. Trans. R. Soc. B Biol. Sci. 366, 2642–2651. https://doi.org/10.1098/rstb.2011.0120

Cruz, T., Fonseca, C., Carvalho, J., Oliveira, B., Torres, R., 2014. Roe deer reintroduction in central Portugal: a tool for Iberian wolf conservation. Galemys, Spanish J. Mammal. 22, 31–40. https://doi.org/10.7325/Galemys.2014.A3

Darmon, G., Calenge, C., Loison, A., Jullien, J.M., Maillard, D., Lopez, J.F., 2012. Spatial distribution and habitat selection in coexisting species of mountain ungulates. Ecography (Cop.). 35, 44–53. https://doi.org/10.1111/j.1600-0587.2011.06664.x

Daskalov, G.M., Grishin, A.N., Rodionov, S., Mihneva, V., 2007. Trophic cascades triggered by overfishing reveal possible mechanisms of ecosystem regime shifts. Proc. Natl. Acad. Sci. 104, 10518–10523. https://doi.org/10.1073/pnas.0701100104

De Marinis, A.M., Asprea, A., 2006a. Hair identification key of wild and domestic ungulates from southern Europe. Wildlife Biol. 12, 305–320. https://doi.org/10.2981/0909-6396(2006)12[305:HIKOWA]2.0.CO;2

De Marinis, A.M., Asprea, A., 2006b. How Did Domestication Change the Hair Morphology in Sheep and Goats? Hum. Evol. 21, 139–149. https://doi.org/10.1007/s11598-006-9010-0

Di Minin, E., Slotow, R., Hunter, L.T.B., Montesino Pouzols, F., Toivonen, T., Verburg, P.H., Leader-Williams, N., Petracca, L., Moilanen, A., 2016. Global priorities for national carnivore conservation under land use change. Sci. Rep. 6, 23814. https://doi.org/10.1038/srep23814

Eggermann, J., da Costa, G.F., Guerra, A.M., Kirchner, W.H., Petrucci-Fonseca, F., 2011. Presence of Iberian wolf (Canis lupus signatus) in relation to land cover, livestock and human influence in Portugal. Mamm. Biol. 76, 217–221. https://doi.org/10.1016/j.mambio.2010.10.010

Elith, J., Leathwick, J.R., 2009. Species Distribution Models: Ecological Explanation and Prediction Across Space and Time. Annu. Rev. Ecol. Evol. Syst. 40, 677–697. https://doi.org/10.1146/annurev.ecolsys.110308.120159

Eom, T.-K., Hwang, H.-S., Lee, J.-K., Rhim, S.-J., 2018. Ecological factors influencing winter field sign abundance of Korean water deer Hydropotes inermis argyropus in a temperate forest in South Korea. Folia Zool. 67, 173–178. https://doi.org/10.25225/fozo.v67.i3-4.a5.2018

Espirito-Santo, C., 2007. Human dimensions in Iberian wolf management in Portugal: attitudes and beliefs of interest groups and the public toward a fragmented wolf population (Master’s Thesis).

Estes, J.A., Terborgh, J., Brashares, J.S., Power, M.E., Berger, J., Bond, W.J., Carpenter, S.R., Essington, T.E., Holt, R.D., Jackson, J.B.C., Marquis, R.J., Oksanen, L., Oksanen, T., Paine, R.T., Pikitch, E.K., Ripple, W.J., Sandin, S.A., Scheffer, M., Schoener, T.W., Shurin, J.B., Sinclair, A.R.E., Soulé, M.E., Virtanen, R., Wardle, D.A., 2011. Trophic Downgrading of Planet Earth. Science. 333, 301–306. https://doi.org/10.1126/science.1205106

Ewald, M., Dupke, C., Heurich, M., Müller, J., Reineking, B., 2014. LiDAR Remote Sensing of Forest Structure and GPS Telemetry Data Provide Insights on Winter Habitat Selection of European Roe Deer. Forests 5, 1374–1390. https://doi.org/10.3390/f5061374

Farfán, M.Á., Duarte, J., Vargas, J.M., Real, R., 2009. El corzo en la región más meridional de su área de distribución. Las zonas más favorables para su caza. Galemys 21, 245–262.

Fernandez, F.A.S., Rheingantz, M.L., Genes, L., Kenup, C.F., Galliez, M., Cezimbra, T., Cid, B., Macedo, L., Araujo, B.B.A., Moraes, B.S., Monjeau, A., Pires, A.S., 2017. Rewilding the Atlantic Forest: Restoring the fauna and ecological interactions of a protected area. Perspect. Ecol. Conserv. 15, 308–314. https://doi.org/10.1016/j.pecon.2017.09.004

Ferretti, F., 2011. Interspecific aggression between fallow and roe deer. Ethol. Ecol. Evol. 23, 179–186. https://doi.org/10.1080/03949370.2011.554883

Ferretti, F., Lovari, S., Mancino, V., Burrini, L., Rossa, M., 2019. Food habits of wolves and selection of wild ungulates in a prey-rich Mediterranean coastal area. Mamm. Biol. 99, 119–127. https://doi.org/10.1016/j.mambio.2019.10.008

Figueiredo, A.M., Valente, A.M., Barros, T., Carvalho, J., Silva, D.A.M., Fonseca, C., Carvalho, L.M. de, Torres, R.T., 2020. What does the wolf eat? Assessing the diet of the endangered Iberian wolf (Canis lupus signatus) in northeast Portugal. PLoS One 15. https://doi.org/10.1371/journal.pone.0230433

Fonseca, C., Migueis, D., Fernandes, T., Carvalho, H., Loureiro, A., Carvalho, J., Torres, R.T., 2017. The return of the Iberian wild goat Capra pyrenaica to Portugal: From reintroduction to recolonization. J. Nat. Conserv. 38, 56–61. https://doi.org/10.1016/j.jnc.2017.05.006

Fortin, D., Buono, P.-L., Schmitz, O.J., Courbin, N., Losier, C., St-Laurent, M.-H., Drapeau, P., Heppell, S., Dussault, C., Brodeur, V., Mainguy, J., 2015. A spatial theory for characterizing predator–multiprey interactions in heterogeneous landscapes. Proc. R. Soc. B Biol. Sci. 282, 20150973. https://doi.org/10.1098/rspb.2015.0973

Foster, R.J., Harmsen, B.J., 2012. A critique of density estimation from camera-trap data. J. Wildl. Manage. 76, 224–236. https://doi.org/10.1002/jwmg.275

Fritts, S.H., Mech, L.D., 1981. Dynamics, movements, and feeding ecology of a newly protected wolf population in northwestern Minnesota. Wildl. Monogr. 80, 3–79.

Fukuda, S., Mouton, A.M., De Baets, B., 2012. Abundance versus presence/absence data for modelling fish habitat preference with a genetic Takagi–Sugeno fuzzy system. Environ. Monit. Assess. 184, 6159–6171. https://doi.org/10.1007/s10661-011-2410-2

Gardner, B., Reppucci, J., Lucherini, M., Royle, J.A., 2010. Spatially explicit inference for open populations: estimating demographic parameters from camera-trap studies. Ecology 91, 3376–3383. https://doi.org/10.1890/09-0804.1

Gaudry, W., Saïd, S., Gaillard, J.-M., Chevrier, T., Loison, A., Maillard, D., Bonenfant, C., 2015. Partial migration or just habitat selection? Seasonal movements of roe deer in an Alpine population. J. Mammal. 96, 502–510. https://doi.org/10.1093/jmammal/gyv055

Gervasi, V., Nilsen, E.B., Odden, J., Bouyer, Y., Linnell, J.D.C., 2013. The spatio-temporal distribution of wild and domestic ungulates modulates lynx kill rates in a multi-use landscape. J. Zool. 292, 175–183. https://doi.org/10.1111/jzo.12088

Gray, T.N.E., 2018. Monitoring tropical forest ungulates using camera-trap data. J. Zool. 305, 173–179. https://doi.org/10.1111/jzo.12547

Hall, L.K., Larsen, R.T., Knight, R.N., McMillan, B.R., 2018. Feral horses influence both spatial and temporal patterns of water use by native ungulates in a semi-arid environment. Ecosphere 9. https://doi.org/10.1002/ecs2.2096

Hebblewhite, M., Pletscher, D.H., 2002. Effects of elk group size on predation by wolves. Can. J. Zool. 80, 800–809. https://doi.org/10.1139/z02-059

Heilbrun, R.D., Silvy, N.J., Peterson, M.J., Tewes, M.E., 2006. Estimating bobcat abundance using automatically triggered cameras. Wildl. Soc. Bull. 34, 69–73. https://doi.org/10.2193/0091-7648(2006)34[69:EBAUAT]2.0.CO;2

Heinze, E., Boch, S., Fischer, M., Hessenmöller, D., Klenk, B., Müller, J., Prati, D., Schulze, E.-D., Seele, C., Socher, S., Halle, S., 2011. Habitat use of large ungulates in northeastern Germany in relation to forest management. For. Ecol. Manage. 261, 288–296. https://doi.org/10.1016/j.foreco.2010.10.022

Hemami, M.., Watkinson, A.., Dolman, P.., 2004. Habitat selection by sympatric muntjac (Muntiacus reevesi) and roe deer (Capreolus capreolus) in a lowland commercial pine forest. For. Ecol. Manage. 194, 49–60. https://doi.org/10.1016/j.foreco.2004.01.049

Hewison, A.J., Vincent, J.P., Joachim, J., Angibault, J.M., Cargnelutti, B., Cibien, C., 2001. The effects of woodland fragmentation and human activity on roe deer distribution in agricultural landscapes. Can. J. Zool. 79, 679–689. https://doi.org/10.1139/z01-032

Hibert, F., Calenge, C., Fritz, H., Maillard, D., Bouché, P., Ipavec, A., Convers, A., Ombredane, D., de Visscher, M.N., 2010. Spatial avoidance of invading pastoral cattle by wild ungulates: Insights from using point process statistics. Biodivers. Conserv. 19, 2003–2024. https://doi.org/10.1007/s10531-010-9822-0

Holdo, R.M., Sinclair, A.R.E., Dobson, A.P., Metzger, K.L., Bolker, B.M., Ritchie, M.E., Holt, R.D., 2009. A Disease-Mediated Trophic Cascade in the Serengeti and its Implications for Ecosystem C. PLoS Biol. 7. https://doi.org/10.1371/journal.pbio.1000210

Holtgrieve, G.W., Schindler, D.E., Jewett, P.K., 2009. Large predators and biogeochemical hotspots: brown bear (Ursus arctos) predation on salmon alters nitrogen cycling in riparian soils. Ecol. Res. 24, 1125–1135. https://doi.org/10.1007/s11284-009-0591-8

Hosseini-Zavarei, F., Farhadinia, M.S., Beheshti-Zavareh, M., Abdoli, A., 2013. Predation by grey wolf on wild ungulates and livestock in central Iran. J. Zool.

290, 127–134. https://doi.org/10.1111/jzo.12022

Iliopoulos, Y., Sgardelis, S., Koutis, V., Savaris, D., 2009. Wolf depredation on livestock in central Greece. Acta Theriol. (Warsz). 54, 11–22.

Imbert, C., Caniglia, R., Fabbri, E., Milanesi, P., Randi, E., Serafini, M., Torretta, E., Meriggi, A., 2016. Why do wolves eat livestock? Biol. Conserv. 195, 156–168. https://doi.org/10.1016/j.biocon.2016.01.003

Ito, T.Y., Lhagvasuren, B., Tsunekawa, A., Shinoda, M., Takatsuki, S., Buuveibaatar, B., Chimeddorj, B., 2013. Fragmentation of the Habitat of Wild Ungulates by Anthropogenic Barriers in Mongolia. PLoS One 8. https://doi.org/10.1371/journal.pone.0056995

IUCN/SSC, 2013. Guidelines for Reintroductions and Other Conservation Translocations, Version 1. ed. Gland, Switzerland. https://doi.org/10.1016/j.biocon.2015.07.030

Jarvie, S., Svenning, J.-C., 2018. Using species distribution modelling to determine opportunities for trophic rewilding under future scenarios of climate change. Philos. Trans. R. Soc. B Biol. Sci. 373. https://doi.org/10.1098/rstb.2017.0446

Jedrzejewski, W., Schmidt, K., Theuerkauf, J., Jedrzejewska, B., Selva, N., Zub, K., Szymura, L., 2002. Kill Rates and Predation by Wolves on Ungulate Populations in Bialowieza Primeval Forest (Poland). Ecology 83, 1341–1356. https://doi.org/10.2307/3071948

Jennelle, C.S., Runge, M.C., MacKenzie, D.I., 2002. The use of photographic rates to estimate densities of tigers and other cryptic mammals: a comment on misleading conclusions. Anim. Conserv. 5, 119–120. https://doi.org/10.1017/S1367943002002160

Johnson, B.E., Cushman, J.H., 2007. Influence of a Large Herbivore Reintroduction on Plant Invasions and Community Composition in a California Grassland. Conserv. Biol. 21, 515–526. https://doi.org/10.1111/j.1523-1739.2006.00610.x

Johnson, C.N., Prior, L.D., Archibald, S., Poulos, H.M., Barton, A.M., Williamson, G.J., Bowman, D.M.J.S., 2018. Can trophic rewilding reduce the impact of fire in a more flammable world? Philos. Trans. R. Soc. B Biol. Sci. 373.

https://doi.org/10.1098/rstb.2017.0443

Kagata, H., Ohgushi, T., 2006. Bottom-up trophic cascades and material transfer in terrestrial food webs. Ecol. Res. 21, 26–34. https://doi.org/10.1007/s11284-005-0124-z

Khan, U., Lovari, S., Ali Shah, S., Ferretti, F., 2018. Predator, prey and humans in a mountainous area: loss of biological diversity leads to trouble. Biodivers. Conserv. 27, 2795–2813. https://doi.org/10.1007/s10531-018-1570-6

Kittle, A.M., Anderson, M., Avgar, T., Baker, J.A., Brown, G.S., Hagens, J., Iwachewski, E., Moffatt, S., Mosser, A., Patterson, B.R., Reid, D.E.B., Rodgers, A.R., Shuter, J., Street, G.M., Thompson, I.D., Vander Vennen, L.M., Fryxell, J.M., 2017. Landscape-level wolf space use is correlated with prey abundance, ease of mobility, and the distribution of prey habitat. Ecosphere 8. https://doi.org/10.1002/ecs2.1783

Kruuk, H., Parish, T., 1981. Feeding specialization of the European Badger (Meles meles) in Scotland. J. Anim. Ecol. 50, 773–788.

Kuijper, D.P.J., Cromsigt, J.P.G.M., Churski, M., Adam, B., Jędrzejewska, B., Jędrzejewski, W., 2009. Do ungulates preferentially feed in forest gaps in European temperate forest? For. Ecol. Manage. 258, 1528–1535. https://doi.org/10.1016/j.foreco.2009.07.010

Kunkel, K., Pletscher, D.H., 2001. Winter hunting patterns of wolves in and near Glacier National Park, Montana. J. Wildl. Manage. 65, 520–530.

Laforge, M.P., Michel, N.L., Wheeler, A.L., Brook, R.K., 2016. Habitat selection by female moose in the Canadian prairie ecozone. J. Wildl. Manage. 80, 1059–1068. https://doi.org/10.1002/jwmg.21095

Latombe, G., Fortin, D., Parrott, L., 2014. Spatio-temporal dynamics in the response of woodland caribou and moose to the passage of grey wolf. J. Anim. Ecol. 83, 185–198. https://doi.org/10.1111/1365-2656.12108

Laundré, J.W., 2010. Behavioral response races, predator–prey shell games, ecology of fear, and patch use of pumas and their ungulate prey. Ecology 91, 2995–3007. https://doi.org/10.1890/08-2345.1

Leahy, L., Legge, S.M., Tuft, K., McGregor, H.W., Barmuta, L.A., Jones, M.E., Johnson, C.N., 2015. Amplified predation after fire suppresses rodent populations in Australia's tropical savannas. Wildl. Res. 42, 705–716. https://doi.org/10.1071/WR15011

Levi, T., Kilpatrick, A.M., Mangel, M., Wilmers, C.C., 2012. Deer, predators, and the emergence of Lyme disease. Proc. Natl. Acad. Sci. 109, 10942–10947. https://doi.org/10.1073/pnas.1204536109

Lingle, S., 2002. Coyote predation and habitat segregation of white-tailed deer and mule deer. Ecology 83, 2037–2048. https://doi.org/10.1890/0012-9658(2002)083[2037:CPAHSO]2.0.CO;2

Linnell, J.D.C., Cretois, B., Nilsen, E.B., Rolandsen, C.M., Solberg, E.J., Veiberg, V., Kaczensky, P., Van Moorter, B., Panzacchi, M., Rauset, G.R., Kaltenborn, B., 2020. The challenges and opportunities of coexisting with wild ungulates in the human-dominated landscapes of Europe's Anthropocene. Biol. Conserv. 244. https://doi.org/10.1016/j.biocon.2020.108500

Linnell, J.D.C., Nijhuis, P., Teurlings, I., Andersen, R., 1999. Selection of bed-sites by roe deer Capreolus capreolus fawns in a boreal forest landscape. Wildlife Biol. 5, 225–231. https://doi.org/10.2981/wlb.1999.027

Linnell, J.D.C., Odden, J., Mertens, A., 2012. Mitigation methods for conflicts associated with carnivore depredation on livestock, in: Carnivore Ecology and Conservation. Oxford University Press, pp. 314–332. https://doi.org/10.1093/acprof:oso/9780199558520.003.0014

Lizarralde, M., Escobar, J.M., Deferrari, G., 2004. Invader species in Argentina: A review about the beaver (Castor canadensis) population situation on Tierra del Fuego ecosystem. Interciencia 29, 352–356.

López-Martín, J.M., Martínez-Martínez, D., Such, À., 2009. Supervivencia, dispersión y selección de recursos de corzos Capreolus capreolus (Linnaeus, 1758) reintroducidos en un hábitat mediterráneo. Galemys 21, 143–164.

Loro, M., Ortega, E., Arce, R.M., Geneletti, D., 2016. Assessing landscape resistance to roe deer dispersal using fuzzy set theory and multicriteria analysis: a case study in Central Spain. Landsc. Ecol. Eng. 12, 41–60. https://doi.org/10.1007/s11355-015-0275-1

Lovari, S., Boesi, R., Minder, I., Mucci, N., Randi, E., Dematteis, A., Ale, S.B., 2009. Restoring a keystone predator may endanger a prey species in a human-altered ecosystem: the return of the snow leopard to Sagarmatha National Park. Anim. Conserv. 12, 559–570. https://doi.org/10.1111/j.1469-1795.2009.00285.x

Lovari, S., Pokheral, C.P., Jnawali, S.R., Fusani, L., Ferretti, F., 2015. Coexistence of the tiger and the common leopard in a prey-rich area: the role of prey partitioning. J. Zool. 295, 122–131. https://doi.org/10.1111/jzo.12192

Lovari, S., San José, C., 1997. Wood dispersion affects home range size of female roe deer. Behav. Processes 40, 239–241. https://doi.org/10.1016/S0376-6357(97)00019-3

Lucherini, M., Crema, G., 1995. Seasonal variation in the food habits of Badgers in an Alpine Valley. Hystrix 7, 165–171. https://doi.org/10.4404/hystrix-7.1-2-4067

Lute, M.L., Carter, N.H., López-Bao, J. V., Linnell, J.D.C., 2020. Conservation professionals' views on governing for coexistence with large carnivores. Biol. Conserv. 248. https://doi.org/10.1016/j.biocon.2020.108668

M'soka, J., Creel, S., Becker, M.S., Murdoch, J.D., 2017. Ecological and anthropogenic effects on the density of migratory and resident ungulates in a human-inhabited protected area. Afr. J. Ecol. 55, 618–631. https://doi.org/10.1111/aje.12398

Macandza, V.A., Owen-Smith, N., Cain, J.W., 2012. Habitat and resource partitioning between abundant and relatively rare grazing ungulates. J. Zool. 287, 175–185. https://doi.org/10.1111/j.1469-7998.2012.00900.x

Marjakangas, E.-L., Genes, L., Pires, M.M., Fernandez, F.A.S., de Lima, R.A.F., de Oliveira, A.A., Ovaskainen, O., Pires, A.S., Prado, P.I., Galetti, M., 2018. Estimating interaction credit for trophic rewilding in tropical forests. Philos. Trans. R. Soc. B Biol. Sci. 373. https://doi.org/10.1098/rstb.2017.0435

Markandya, A., Taylor, T., Longo, A., Murty, M.N., Murty, S., Dhavala, K., 2008. Counting the cost of vulture decline—An appraisal of the human health and other benefits of vultures in India. Ecol. Econ. 67, 194–204. https://doi.org/10.1016/j.ecolecon.2008.04.020

Marques, F.F.C., Buckland, S.T., Goffin, D., Dixon, C.E., Borchers, D.L., Mayle, B.A., Peace, A.J., 2001. Estimating deer abundance from line transect surveys of dung: sika deer in southern Scotland. J. Appl. Ecol. 38, 349–363. https://doi.org/10.1046/j.1365-2664.2001.00584.x

Marques, T.A., Thomas, L., Fancy, S.G., Buckland, S.T., 2007. Improving Estimates of Bird Density Using Multiple- Covariate Distance Sampling. Auk 124, 1229–1243. https://doi.org/10.1093/auk/124.4.1229

Mattioli, L., Capitani, C., Gazzola, A., Scandura, M., Apollonio, M., 2011. Prey selection and dietary response by wolves in a high-density multi-species ungulate community. Eur. J. Wildl. Res. 57, 909–922. https://doi.org/10.1007/s10344-011-0503-4

McCullagh, P., Nelder, J.A., 1989. Generalized Linear Models, 2nd Editio. ed. Chapman and Hall.

Meireles, C., Pinto-Gomes, C., 2012. Guia de campo do itinerário geobotânico à Serra da Estrela. Folia Bot. Extrem. 6, 106–170.

Meriggi, A., Brangi, A., Matteucci, C., Sacchi, O., 1996. The feeding habits of wolves in relation to large prey availability in northern Italy. Ecography (Cop.). 19, 287–295.

Meriggi, A., Dagradi, V., Dondina, O., Perversi, M., Milanesi, P., Lombardini, M., Raviglione, S., Repossi, A., 2015. Short-term responses of wolf feeding habits to changes of wild and domestic ungulate abundance in Northern Italy. Ethol. Ecol. Evol. 27, 389–411. https://doi.org/10.1080/03949370.2014.986768

Meriggi, A., Lovari, S., 1996. A Review of Wolf Predation in Southern Europe: Does the Wolf Prefer Wild Prey to Livestock? J. Appl. Ecol. 33, 1561. https://doi.org/10.2307/2404794

Meriggi, A., Rosa, P., Brangi, A., Matteucci, C., 1991. Habitat use and diet of the wolf in northern Italy. Acta Theriol. (Warsz). 36, 141–151. https://doi.org/10.4098/AT.arch.91-11

Migli, D., Youlatos, D., Iliopoulos, Y., 2005. Winter food habits of wolves in central Greece. J. Biol. Res. 4, 217–220.

Milanesi, P., Meriggi, A., Merli, E., 2012. Selection of wild ungulates by wolves Canis lupus (L. 1758) in an area of the Northern Apennines (North Italy). Ethol. Ecol. Evol. 24, 81–96. https://doi.org/10.1080/03949370.2011.592220

Mills, M.G.L., 1990. Kalahari Hyenas, Kalahari Hyenas. Springer Netherlands, Dordrecht. https://doi.org/10.1007/978-94-011 -1244-4

Mitchell, B., Rowe, J.J., Ratcliffe, P., Hinge, M., 1985. Defecation frequency in Roe deer (Capreolus capreolus) in relation to the accumulation rates of faecal deposits. J. Zool. 207, 1–7. https://doi.org/10.1111/j.1469-7998.1985.tb04910.x

Monteiro-Henriques, T., Martins, M.J., Cerdeira, J.O., Silva, P., Arsénio, P., Silva, Á., Bellu, A., Costa, J.C., 2016. Bioclimatological mapping tackling uncertainty propagation: application to mainland Portugal. Int. J. Climatol. 36, 400–411. https://doi.org/10.1002/joc.4357

Monterroso, P., Alves, P.C., Ferreras, P., 2014. Plasticity in circadian activity patterns of mesocarnivores in Southwestern Europe: implications for species coexistence. Behav. Ecol. Sociobiol. 68, 1403–1417. https://doi.org/10.1007/s00265-014-1748-1

Morellet, N., Bonenfant, C., Börger, L., Ossi, F., Cagnacci, F., Heurich, M., Kjellander, P., Linnell, J.D.C., Nicoloso, S., Sustr, P., Urbano, F., Mysterud, A., 2013. Seasonality, weather and climate affect home range size in roe deer across a wide latitudinal gradient within Europe. J. Anim. Ecol. 82, 1326–1339. https://doi.org/10.1111/1365-2656.12105

Morellet, N., Van Moorter, B., Cargnelutti, B., Angibault, J.-M., Lourtet, B., Merlet, J., Ladet, S., Hewison, A.J.M., 2011. Landscape composition influences roe deer habitat selection at both home range and landscape scales. Landsc. Ecol. 26, 999–1010. https://doi.org/10.1007/s10980-011-9624-0

Moser, B., Schütz, M., Hindenlang, K.E., 2006. Importance of alternative food resources for browsing by roe deer on deciduous trees: The role of food availability and species quality. For. Ecol. Manage. 226, 248–255. https://doi.org/10.1016/j.foreco.2006.01.045

Mueller, M.A., Drake, D., Allen, M.L., 2018. Coexistence of coyotes (Canis latrans) and red foxes (Vulpes vulpes) in an urban landscape. PLoS One 13. https://doi.org/10.1371/journal.pone.0190971

Mysterud, A., Larsen, P.K., Ims, R.A., Østbye, E., 1999. Habitat selection by roe deer and sheep: does habitat ranking reflect resource availability? Can. J. Zool. 77, 776–783. https://doi.org/10.1139/z99-025

Mysterud, A., Østbye, E., 1999. Cover as a habitat element for temperate ungulates: effects on habitat selection and demography. Wildl. Soc. Bull. 27, 385–394.

Nakashima, Y., Hongo, S., Akomo-Okoue, E.F., 2020. Landscape-scale estimation of forest ungulate density and biomass using camera traps: Applying the REST model. Biol. Conserv. 241, 108381. https://doi.org/10.1016/j.biocon.2019.108381

Negrões, N., Sarmento, P., Cruz, J., Eira, C., Revilla, E., Fonseca, C., Sollmann, R., Tôrres, N.M., Furtado, M.M., Jácomo, A.T.A., Silveira, L., 2010. Use of Camera-Trapping to Estimate Puma Density and Influencing Factors in Central Brazil. J. Wildl. Manage. 74, 1195–1203. https://doi.org/10.2193/2009-256

Newsome, T.M., Ripple, W.J., 2015. A continental scale trophic cascade from wolves through coyotes to foxes. J. Anim. Ecol. 84, 49–59. https://doi.org/10.1111/1365-2656.12258

Nomani, S.Z., Oli, M.K., Carthy, R.R., 2012. Line Transects by Design: The Influence of Study Design, Spatial Distribution and Density of Objects on Estimates of Abundance. Open Ecol. J. 5, 25–44. https://doi.org/10.2174/1874213001205010025

Nores, C., Llaneza, L., Álvarez, Á., 2008. Wild boar Sus scrofa mortality by hunting and wolf Canis lupus predation: an example in northern Spain. Wildlife Biol. 14, 44–51. https://doi.org/10.2981/0909-6396(2008)14[44:WBSSMB]2.0.CO;2

Odadi, W.O., Karachi, M.K., Abdulrazak, S.A., Young, T.P., 2011. African Wild Ungulates Compete with or Facilitate Cattle Depending on Season. Science. 333, 1753–1755. https://doi.org/10.1126/science.1208468

Oliveira, T., Carmo, P., 2000. Distribuição das Principais Presas Selvagens do Lobo Ibérico (Canis lupus signatus Cabrera 1907) a Norte do Rio Douro. Galemys 12, 257–268.

Ossi, F., Gaillard, J.-M., Hebblewhite, M., Cagnacci, F., 2015. Snow sinking depth and forest canopy drive winter resource selection more than supplemental feeding in an alpine population of roe deer. Eur. J. Wildl. Res. 61, 111–124. https://doi.org/10.1007/s10344-014-0879-z

Papageorgiou, N., Vlachos, C., Sfougaris, A., Tsachalidis, A., 1994. Status and diet of wolves in Greece. Acta Theriol. (Warsz). 39, 411–416. https://doi.org/10.4098/AT.arch.94-48

Parker, K.L., Barboza, P.S., Gillingham, M.P., 2009. Nutrition integrates environmental responses of ungulates. Funct. Ecol. 23, 57–69. https://doi.org/10.1111/j.1365-2435.2009.01528.x

Patalano, M., Lovari, S., 1993. Food habits and throphic niche overlap of the wolf (Canis lupus, L. 1758) and the red fox (Vulpes Vulpes, L. 1758) in a mediterranean mountain area. Rev. d’Ecologie 48, 279–294.

Peñuelas, J., Sardans, J., Filella, I., Estiarte, M., Llusià, J., Ogaya, R., Carnicer, J., Bartrons, M., Rivas-Ubach, A., Grau, O., Peguero, G., Margalef, O., Pla-Rabés, S., Stefanescu, C., Asensio, D., Preece, C., Liu, L., Verger, A., Barbeta, A., Achotegui-Castells, A., Gargallo-Garriga, A., Sperlich, D., Farré-Armengol, G., Fernández-Martínez, M., Liu, D., Zhang, C., Urbina, I., Camino-Serrano, M., Vives-Ingla, M., Stocker, B., Balzarolo, M., Guerrieri, R., Peaucelle, M., Marañón-Jiménez, S., Bórnez-Mejías, K., Mu, Z., Descals, A., Castellanos, A., Terradas, J., 2017. Impacts of Global Change on Mediterranean Forests and Their Services. Forests 8, 463. https://doi.org/10.3390/f8120463

Périquet, S., Valeix, M., Loveridge, A.J., Madzikanda, H., Macdonald, D.W., Fritz, H., 2010. Individual vigilance of African herbivores while drinking: the role of immediate predation risk and context. Anim. Behav. 79, 665–671. https://doi.org/10.1016/j.anbehav.2009.12.016

Pfeffer, S.E., Spitzer, R., Allen, A.M., Hofmeester, T.R., Ericsson, G., Widemo, F., Singh, N.J., Cromsigt, J.P.G.M., 2018. Pictures or pellets? Comparing camera trapping and dung counts as methods for estimating population densities of ungulates. Remote Sens. Ecol. Conserv. 4, 173–183. https://doi.org/10.1002/rse2.67

Pimenta, V., Barroso, I., Álvares, F., Correia, J., Ferrão da Costa, G., Moreira, L., Nascimento, J., Petrucci-Fonseca, F., Roque, S., Santos, E., 2005. Situação populacional do Lobo em Portugal: resultados do Censo Nacional 2002/2003. Lisboa.

Pimenta, V., Barroso, I., Boitani, L., Beja, P., 2017. Wolf predation on cattle in Portugal: Assessing the effects of husbandry systems. Biol. Conserv. 207, 17–26. https://doi.org/10.1016/j.biocon.2017.01.008

Pires, A.P.F., Marino, N.A.C., Srivastava, D.S., Farjalla, V.F., 2016. Predicted rainfall changes disrupt trophic interactions in a tropical aquatic ecosystem.

Ecology 97, 2750–2759. https://doi.org/10.1002/ecy.1501

Pohja-Mykrä, M., Kurki, S., 2014. Strong community support for illegal killing challenges wolf management. Eur. J. Wildl. Res. 60, 759–770. https://doi.org/10.1007/s10344-014-0845-9

Poulle, M.L., Carles, L., Lequette, B., 1997. Significance of ungulates in the diet of recently settled wolves in the Mercantour mountains (southeastern France). Rev. d'Ecologie (La Terre la Vie) 52, 357–368.

Redpath, S.M., Young, J., Evely, A., Adams, W.M., Sutherland, W.J., Whitehouse, A., Amar, A., Lambert, R.A., Linnell, J.D.C., Watt, A., Gutiérrez, R.J., 2013. Understanding and managing conservation conflicts. Trends Ecol. Evol. 28, 100–109. https://doi.org/10.1016/j.tree.2012.08.021

Reimoser, F., Putman, R., 2011. Impacts of wild ungulates on vegetation: costs and benefits, in: Putman, R., Apollonio, M., Andersen, R. (Eds.), Ungulate Management in Europe. Cambridge University Press, Cambridge, pp. 144–191. https://doi.org/10.1017/CBO9780511974137.007

Richard, J.H., Wilmshurst, J., Côté, S.D., 2014. The effect of snow on space use of an alpine ungulate: recently fallen snow tells more than cumulative snow depth. Can. J. Zool. 92, 1067–1074. https://doi.org/10.1139/cjz-2014-0118

Ripple, W.J., Beschta, R.L., Painter, L.E., 2015. Trophic cascades from wolves to alders in Yellowstone. For. Ecol. Manage. 354, 254–260. https://doi.org/10.1016/j.foreco.2015.06.007

Ripple, W.J., Estes, J.A., Schmitz, O.J., Constant, V., Kaylor, M.J., Lenz, A., Motley, J.L., Self, K.E., Taylor, D.S., Wolf, C., 2016. What is a Trophic Cascade? Trends Ecol. Evol. 31, 842–849. https://doi.org/10.1016/j.tree.2016.08.010

Rodríguez-Estival, J., Martinez-Haro, M., Monsalve-González, L., Mateo, R., 2011. Interactions between endogenous and dietary antioxidants against Pb-induced oxidative stress in wild ungulates from a Pb polluted mining area. Sci. Total Environ. 409, 2725–2733. https://doi.org/10.1016/j.scitotenv.2011.04.010

Romani, T., Giannone, C., Mori, E., Filacorda, S., 2018. Use of track counts and camera traps to estimate the abundance of roe deer in North-Eastern Italy: are they effective methods? Mammal Res. 63, 477–484.

https://doi.org/10.1007/s13364-018-0386-9

Roque, S., Álvares, F., Petrucci-Fonseca, F., 2001. Utilización espacio-temporal y hábitos alimentarios de un grupo reproductor de lobos en el noroeste de Portugal. Galemys 13, 179–198.

Rossa, M., Lovari, S., Ferretti, F., 2021. Spatiotemporal patterns of wolf, mesocarnivores and prey in a Mediterranean area. Behav. Ecol. Sociobiol. 75, 32. https://doi.org/10.1007/s00265-020-02956-4

Rovero, F., Marshall, A.R., 2009. Camera trapping photographic rate as an index of density in forest ungulates. J. Appl. Ecol. 46, 1011–1017. https://doi.org/10.1111/j.1365-2664.2009.01705.x

Rovero, F., Zimmermann, F., 2016. Camera trapping for Wildlife Research, Pelagic Publishing. Pelagic Publishing, Exeter.

Rowcliffe, J.M., Field, J., Turvey, S.T., Carbone, C., 2008. Estimating animal density using camera traps without the need for individual recognition. J. Appl. Ecol. 45, 1228–1236. https://doi.org/10.1111/j.1365-2664.2008.01473.x

Saïd, S., Servanty, S., 2005. The Influence of Landscape Structure on Female Roe Deer Home-range Size. Landsc. Ecol. 20, 1003–1012. https://doi.org/10.1007/s10980-005-7518-8

Salazar, D.C., 2009. Distribuição e Estatuto do Veado e Corço em Portugal. Biol. Dep. Universidade de Aveiro.

Schmitz, O.J., Krivan, V., Ovadia, O., 2004. Trophic cascades: the primacy of trait-mediated indirect interactions. Ecol. Lett. 7, 153–163. https://doi.org/10.1111/j.1461-0248.2003.00560.x

Schuette, P., Creel, S., Christianson, D., 2016. Ungulate distributions in a rangeland with competitors, predators and pastoralists. J. Appl. Ecol. 53, 1066–1077. https://doi.org/10.1111/1365-2664.12610

Sévêque, A., Gentle, L.K., López-Bao, J. V., Yarnell, R.W., Uzal, A., 2020. Human disturbance has contrasting effects on niche partitioning within carnivore communities. Biol. Rev. 95, 1689–1705. https://doi.org/10.1111/brv.12635

Sidorovich, V., Schnitzler, A., Schnitzler, C., Rotenko, I., Holikava, Y., 2017. Responses of wolf feeding habits after adverse climatic events in central-western Belarus. Mamm. Biol. 83, 44–50.

https://doi.org/10.1016/j.mambio.2016.11.012

Skarin, A., Alam, M., 2017. Reindeer habitat use in relation to two small wind farms, during preconstruction, construction, and operation. Ecol. Evol. 7, 3870–3882. https://doi.org/10.1002/ece3.2941

Sollmann, R., Mohamed, A., Samejima, H., Wilting, A., 2013. Risky business or simple solution – Relative abundance indices from camera-trapping. Biol. Conserv. 159, 405–412. https://doi.org/10.1016/j.biocon.2012.12.025

Sorensen, A.A., van Beest, F.M., Brook, R.K., 2015. Quantifying overlap in crop selection patterns among three sympatric ungulates in an agricultural landscape. Basic Appl. Ecol. 16, 601–609. https://doi.org/10.1016/j.baae.2015.05.001

Soulsbury, C.D., White, P.C.L., 2015. Human–wildlife interactions in urban areas: a review of conflicts, benefits and opportunities. Wildl. Res. 42, 541–553. https://doi.org/10.1071/WR14229

Strickland, M.S., Hawlena, D., Reese, A., Bradford, M.A., Schmitz, O.J., 2013. Trophic cascade alters ecosystem carbon exchange. Proc. Natl. Acad. Sci. 110, 11035–11038. https://doi.org/10.1073/pnas.1305191110

Strong, D.R., Frank, K.T., 2010. Human Involvement in Food Webs. Annu. Rev. Environ. Resour. 35, 1–23. https://doi.org/10.1146/annurev-environ-031809-133103

Szewczyk, M., Nowak, S., Niedźwiecka, N., Hulva, P., Špinkytė-Bačkaitienė, R., Demjanovičová, K., Bolfíková, B.Č., Antal, V., Fenchuk, V., Figura, M., Tomczak, P., Stachyra, P., Stępniak, K.M., Zwijacz-Kozica, T., Mysłajek, R.W., 2019. Dynamic range expansion leads to establishment of a new, genetically distinct wolf population in Central Europe. Sci. Rep. 9. https://doi.org/10.1038/s41598-019-55273-w

Tambling, C.J., Minnie, L., Meyer, J., Freeman, E.W., Santymire, R.M., Adendorff, J., Kerley, G.I.H., 2015. Temporal shifts in activity of prey following large predator reintroductions. Behav. Ecol. Sociobiol. 69, 1153–1161. https://doi.org/10.1007/s00265-015-1929-6

Teerink, B.J., 1991. Hairs of west European mammals. Cambridge University Press, Cambridge.

Terborgh, J., Feeley, K., Silman, M., Nuñez, P., Balukjian, B., 2006. Vegetation dynamics of predator-free land-bridge islands. J. Ecol. 94, 253–263. https://doi.org/10.1111/j.1365-2745.2006.01106.x

Theuerkauf, J., Rouys, S., 2008. Habitat selection by ungulates in relation to predation risk by wolves and humans in the Białowieża Forest, Poland. For. Ecol. Manage. 256, 1325–1332. https://doi.org/10.1016/j.foreco.2008.06.030

Thomas, L., Buckland, S.T., Rexstad, E.A., Laake, J.L., Strindberg, S., Hedley, S.L., Bishop, J.R.B., Marques, T.A., Burnham, K.P., 2010. Distance software: design and analysis of distance sampling surveys for estimating population size. J. Appl. Ecol. 47, 5–14. https://doi.org/10.1111/j.1365-2664.2009.01737.x

Torres, R.T., Brotas, G., Fonseca, C., 2018. Roe deer reintroduction in central Portugal: a tool for Iberian wolf conservation, in: Soorae, P.S. (Ed.), Global Reintroduction Perspectives: 2018. Case Studies from around the Globe. IUCN/SSC Reintroduction Specialist Group, Gland, Switzerland and Environment Agency, Abu Dhabi, UAE, pp. 139–143. https://doi.org/10.2305/iucn.ch.2018.08.en

Torres, R.T., Carvalho, J., Fonseca, C., Serrano, E., López-Martín, J.M., 2016. Long-term assessment of roe deer reintroductions in North-East Spain: A case of success. Mamm. Biol. 81, 415–422. https://doi.org/10.1016/j.mambio.2016.05.002

Torres, R.T., Carvalho, J.C., Panzacchi, M., Linnell, J.D.C., Fonseca, C., 2011. Comparative use of forest habitats by roe deer and moose in a human-modified landscape in southeastern Norway during winter. Ecol. Res. 26, 781–789. https://doi.org/10.1007/s11284-011-0837-0

Torres, R.T., Fonseca, C., 2016. Perspectives on the Iberian wolf in Portugal: population trends and conservation threats. Biodivers. Conserv. 25, 411–425. https://doi.org/10.1007/s10531-016-1061-6

Torres, R.T., Miranda, J., Carvalho, J., Fonseca, C., 2015a. Expansion and Current Status of Roe Deer (Capreolus capreolus) at the Edge of Its Distribution in Portugal. Ann. Zool. Fennici 52, 339–352. https://doi.org/10.5735/086.052.0508

Torres, R.T., Santos, J., Fonseca, C., 2013. Persistence of roe (Capreolus

capreolus) and red (Cervus elaphus) deer pellet-groups in a Mediterranean mosaic landscape. Wildl. Biol. Pract. 9, 7–18. https://doi.org/10.2461/wbp.2013.ibeun.2

Torres, R.T., Santos, J., Linnell, J.D.C., Virgós, E., Fonseca, C., 2010. Factors affecting roe deer occurrence in a Mediterranean landscape, Northeastern Portugal. Mamm. Biol. 76, 491–497. https://doi.org/10.1016/j.mambio.2010.10.013

Torres, R.T., Silva, N., Brotas, G., Fonseca, C., 2015b. To Eat or Not To Eat? The Diet of the Endangered Iberian Wolf (Canis lupus signatus) in a Human-Dominated Landscape in Central Portugal. PLoS One 10, e0129379. https://doi.org/10.1371/journal.pone.0129379

Torres, Rita Tinoco, Virgós, E., Panzacchi, M., Linnell, J.D.C., Fonseca, C., 2012. Life at the edge: Roe deer occurrence at the opposite ends of their geographical distribution, Norway and Portugal. Mamm. Biol. 77, 140–146. https://doi.org/10.1016/j.mambio.2011.11.002

Torres, Rita T., Virgós, E., Santos, J., Linnell, J.D.C., Fonseca, C., 2012. Habitat use by sympatric red and roe deer in a Mediterranean ecosystem. Anim. Biol. 62, 351–366. https://doi.org/10.1163/157075612X631213

Torretta, E., Mosini, A., Piana, M., Tirozzi, P., Serafini, M., Puopolo, F., Saino, N., Balestrieri, A., 2017. Time partitioning in mesocarnivore communities from different habitats of NW Italy: insights into martens' competitive abilities. Behaviour 154, 241–266. https://doi.org/10.1163/1568539X-00003420

Treves, A., Bruskotter, J., 2014. Tolerance for Predatory Wildlife. Science. 344, 476–477. https://doi.org/10.1126/science.1252690

Valente, A., Valente, J., Fonseca, C., Torres, R., 2017. The success of species reintroductions: a case study of red deer in Portugal two decades after reintroduction. Int. J. Biodivers. Sci. Ecosyst. Serv. Manag. 13, 134–138. https://doi.org/10.1080/21513732.2016.1277265

Valente, A.M., Acevedo, P., Figueiredo, A.M., Fonseca, C., Torres, R.T., 2020. Overabundant wild ungulate populations in Europe: management with consideration of socio-ecological consequences. Mamm. Rev. 50, 353–366. https://doi.org/10.1111/mam.12202

Valente, A.M., Fonseca, C., Marques, T.A., Santos, J.P., Rodrigues, R., Torres, R.T., 2014. Living on the Edge: Roe Deer (Capreolus capreolus) Density in the Margins of Its Geographical Range. PLoS One 9. https://doi.org/10.1371/journal.pone.0088459

Valente, A.M., Rocha, R.G., Delgado, E., Ferreira, J.P., Fonseca, C., 2015. Atlas dos Pelos dos Mamíferos Terrestres Ibéricos. Edições Afrontamento.

van der Knaap, W.O., van Leeuwen, J.F.., 1995. Holocene vegetation succession and degradation as responses to climatic change and human activity in the Serra de Estrela , Portugal. Rev. Palaeobot. Palynol. 89, 153–211. https://doi.org/10.1016/0034-6667(95)00048-0

Vidal, F., Arias, E., Garrido, F., Parra, Y., Espinoza, J., 2018. Experimental reintroduction of South Andean huemul and Guanaco in the Huilo Huilo Chilean Private Reserve, Chile, in: Soorae, P.S. (Ed.), Global Reintroduction Perspectives: 2018. Case Studies from around the Globe. IUCN/SSC Reintroduction Specialist Group, Gland, Switzerland and Environment Agency, Abu Dhabi, UAE, pp. 125–129.

Virgós, E., Tellería, J.L., 1998. Roe deer habitat selection in Spain: constraints on the distribution of a species. Can. J. Zool. 76, 1294–1299. https://doi.org/10.1139/z98-065

Vos, J., 2000. Food habits and livestock depredation of two Iberian wolf packs (Canis lupus signatus) in the north of Portugal. J. Zool. 251, 457–462. https://doi.org/10.1017/S0952836900008050

Wagner, C., Holzapfel, M., Kluth, G., Reinhardt, I., Ansorge, H., 2012. Wolf (Canis lupus) feeding habits during the first eight years of its occurrence in Germany. Mamm. Biol. 77, 196–203. https://doi.org/10.1016/j.mambio.2011.12.004

Wallach, A.D., Inbar, M., Scantlebury, M., Speakman, J.R., Shanas, U., 2007. Water requirements as a bottleneck in the reintroduction of European roe deer to the southern edge of its range. Can. J. Zool. 85, 1182–1192. https://doi.org/10.1139/Z07-098

Wirsing, A.J., Cameron, K.E., Heithaus, M.R., 2010. Spatial responses to predators vary with prey escape mode. Anim. Behav. 79, 531–537. https://doi.org/10.1016/j.anbehav.2009.12.014

Yang, H., Zhao, X., Han, B., Wang, T., Mou, P., Ge, J., Feng, L., 2018. Spatiotemporal patterns of Amur leopards in northeast China: Influence of tigers, prey, and humans. Mamm. Biol. 92, 120–128. https://doi.org/10.1016/j.mambio.2018.03.009

Yokoyama, Y., Nakashima, Y., Yajima, G., Miyashita, T., 2020. Simultaneous estimation of seasonal population density, habitat preference and catchability of wild boars based on camera data and harvest records. R. Soc. Open Sci. 7, 200579. https://doi.org/10.1098/rsos.200579

Zlatanova, D., Ahmed, A., Valasseva, A., Genov, P., 2014. Adaptive diet strategy of the wolf (Canis lupus L.) in Europe: A review. Acta Zool. Bulg. 66, 439–452.

Zuberogoitia, I., del Real, J., Torres, J.J., Rodríguez, L., Alonso, M., Zabala, J., 2014. Ungulate Vehicle Collisions in a Peri-Urban Environment: Consequences of Transportation Infrastructures Planned Assuming the Absence of Ungulates. PLoS One 9. https://doi.org/10.1371/journal.pone.0107713

Zuur, A.F., Hilbe, J.M., Ieno, E.N., 2013. A Beginner's Guide to GLM and GLMM with R, Beginner's Guide Series. Highland Statistics Ltd.

Zuur, A.F., Ieno, E.N., Smith, G.M., 2007. Analysing Ecological Data. Springer Verlag.

Zweifel-Schielly, B., Kreuzer, M., Ewald, K.C., Suter, W., 2009. Habitat selection by an Alpine ungulate: the significance of forage characteristics varies with scale and season. Ecography (Cop.). 32, 103–113. https://doi.org/10.1111/j.1600-0587.2008.05178.x