Trophic resource partitioning between ungulates in the Bavarian Forest National Park

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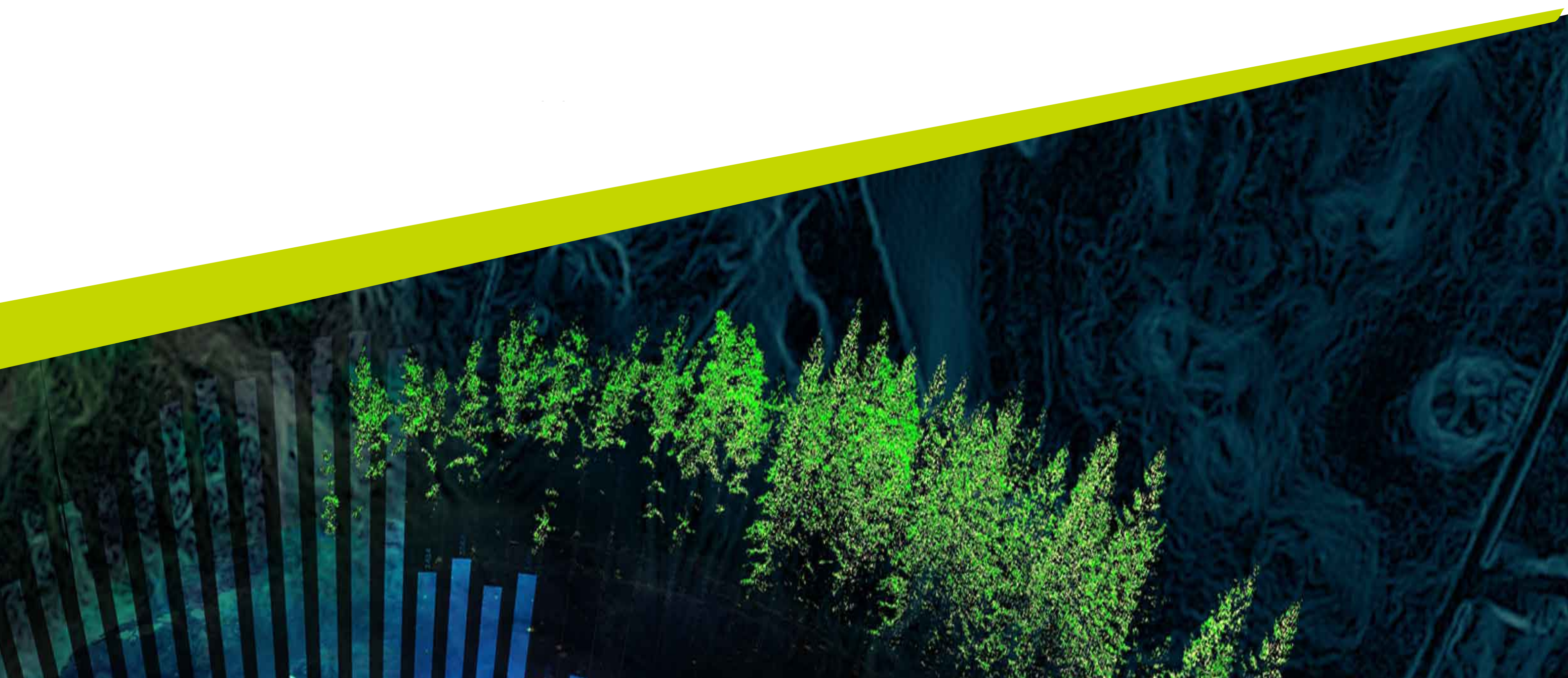

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Abstract

Ungulate populations in Europe have increased significantly in recent decades, leading to substantial impacts on ecosystem structure and composition. Understanding ungulate diets is crucial for managing their effects on ecosystems. This thesis used faecal DNA metabarcoding results to investigate the dietary patterns and resource partitioning of ungulate species in the Bavarian Forest National Park (BFNP). I tested three hypotheses: (1) diet diversity of red deer and roe deer increases with altitude, leading to less specialization; (2) roe deer alter their diet to include more head-up diet to lynx predation risk; and (3) both deer species select European silver fir as a food source. Results indicated that red deer diet diversity and evenness increased with altitude, while roe deer showed no significant altitudinal changes. Roe deer diets shifted towards shrubs and broadleaves in areas with high lynx presence, whereas red deer diets remained unaffected. Both species demonstrated a preference for Rosaceae and Vaccinium, with roe deer showing selective feeding on Abies. The findings illustrate the interactions between ungulate feeding behaviour, habitat conditions, and predator presence, providing essential insights for forest management and conservation strategies in the BFNP.

Keywords: Capreolus capreolus, *Cervus elaphus*, dietary patterns, diet DNA metabarcoding, diet selection, predation risk

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Introduction

Populations of ungulates, i.e. hoofed mammals with enamelled teeth (Cope 1882), have recovered rapidly across Europe in the past decades (Deinet et al. 2013); in many parts probably surpassing historical levels (Linnell et al. 2020). Historically, the extermination of large predators casing by human impacts has reduced ungulate predation pressure in Europe (Bradshaw et al. 2003; Milner et al. 2006; Apollonio et al. 2010; Ripple et al. 2014; Linnell et al. 2020). Reintroduction programs of large herbivorous and omnivorous ungulates like deer and wild boar, respectively, changes in land-use which provided high access to anthropogenic food sources such as crops or forest plantations coupled with warmer and less snow-rich winters due to climate change, have promoted an increase in ungulate numbers and expansion of species' ranges (Deinet et al. 2013; Spitzer et al. 2020).

Ungulates are important driving forces of terrestrial ecosystem structure, composition and development (GILL 1992; Ammer 1996; Fuller 2001; Apollonio et al. 2010; Möst et al. 2015; Ripple et al. 2015). They affect the ecosystems through grazing, browsing, physical disturbance of the soil surface, and seed dispersal (Bruinderink & Hazebroek 1996; Reimoser 2003; Pellerin et al. 2010). The impact of ungulates on ecosystems depends on their density, browsing intensity and forest management (Van Hees et al. 1996; Heckel et al. 2010; Pellerin et al. 2010). Studies have shown that high-density ungulate populations can affect vegetation, including species composition, ground vegetation and tree regeneration (Kuijper 2011; Bernes et al. 2018). Ramirez et al. (2018) illustrate that high grazing or browsing rates can have negative impacts on species of conservation concern and ecosystem productivity. For example, in Central Europe, increasing numbers of ungulates have been attributed to the poor regeneration of European silver fir (*Abies alba*) in the recent past (Motta 1996). European silver fir is one of the most historically and economically valuable conifers in Europe, it is a key indicator tree species for forest management and can increase the biodiversity of forest ecosystems (Dobrowolska et al. 2017). At the same time, it is one of the most heavily browsed tree species in Europe (Senn & Suter 2003), so understanding how ungulates select European silver fir is crucial for the survival of this valuable tree species.

Under such conditions, balancing ungulate impacts and ecosystem functioning becomes an enormous challenge to prevent shifts towards undesired states. Food choice is critical in addressing ungulate impacts on ecosystems as satisfying dietary needs is one of the strongest drivers of ungulate behaviour. Understanding what drives feeding choices and dietary changes is important because it is linked to the impact of ungulate communities on vegetation (Spitzer et al. 2020).

Space use is an important component in dietary choice (Stewart et al. 2002; Lowrey et al. 2018). An organism may choose what to eat in response to variations in habitat conditions (Senft et al. 1987; Apps et al. 2001; Ihl & Klein 2001). A strong natural gradient associated with habitat change is altitude. Studies have shown that food availability usually changes with altitudinal gradients (Terborgh 1977; Carothers et al. 1996; Martínez & Rechberger 2007). Altitudinal gradients could affect the animals' distribution and selection of resources. In mountainous areas, the species richness of vegetation decreases with increasing altitude (Gentry 1988; Grytnes & Vetaas 2002; Erschbamer et al. 2006). Consequently, (Pellissier et al. 2012) suggest that generalisation in the use of resources in herbivores increases at higher altitudes in response to the decrease in plant diversity. Dietary overlap can also be affected by spatial changes, as(Schwartz & Ellis 1981) have shown that diet overlap increases as forage resources become less available.

Ungulate prey species may also change their diet's composition in response to large carnivore predators' presence (Churski et al. 2021). Antipredator responses in ungulates can influence their foraging behaviour at different scales (Brown & Kotler 2004). Prey behaviour and foraging intensity may be influenced by escape choices. A common tactic used by ungulates to reduce predation risk is to increase vigilance while foraging (Brown et al. 1999). Churski et al. (2021) showed that red deer increase vigilance by changing their food plant selection, increasing broadleaved tree use and reducing the use of forb to keep their heads up to increase their chances of detecting approaching large predators. With the ongoing recovery of Europe's large carnivores since the latter half of the 20th century (Chapron et al. 2014), it is crucial to understand how large carnivores influence vegetation via effects on ungulate prey, which is relatively lacking in European systems.

Understanding the browsing patterns of ungulates is important for ecosystem productivity and is a prerequisite for increasing the survival of young European silver fir. In this thesis, I used faecal DNA metabarcoding-based diet data to investigate patterns of trophic resource use and partitioning in red deer (*Cervus elaphus*) and roe deer (*Capreolus capreolus*) in the Bavarian Forest National Park (BFNP) in Germany (Figure 1). I chose these species because they occur widely throughout the National Park (as well as Europe) and are of high management and conservation interest (Heurich et al. 2011).

Red deer and roe deer are both ruminants that digest structural carbohydrates (cellulose and hemicellulose) through microbial actions in the reticulo-rumen. Roe deer are commonly understood as browsers (Cederlund et al. 1980; Cornelis et al. 1999), dependent on high-quality, low-fibre food items (browse, forbs and fruits) due to their small body size (Gordon & Illius 1989). They are known for their preference for hardwood browse; with twigs often forming a large part of their diet (Tixier & Duncan 1996). Red deer are generally classified as intermediate feeders, with a mixed diet of grass, sedges and concentrated food, and should be characterised by seasonal variation (Hofmann 1989). Clauss et al. (2010) argued that ruminant species should be classified as 'moose-types' (for a typical 'browser', i.e. roe deer) and 'cattle-types' (well-adapted to digest graminoids, i.e. ‘grazer’ diets) based on anatomical/physiological features of the reticulo-rumen. The low stratified rumen content of ‘moose-type’ ruminants makes them poorly adapted to digest graminoids whereas there are no real restrictions on ‘cattle-type’ ruminants utilising browse.

Wild boar (*Sus scrofa*), an omnivorous hindgut fermenter, was also included in some of the analyses because it is a key component of the current European ungulate community and utilizes many of the same plant food resources as ruminants (Genov 1981).

Specifically, I tested the following hypotheses: (1) diet diversity of red deer and roe deer will increase with altitude and become less specialised; (2) roe deer diet will change to a ‘head-up’ vigilance diet including more shrubs and tree browse with lynx predation risk whereas red deer would not show such changes because roe deer are the preferred species of Eurasian lynx (*Lynx lynx*); (3) roe deer and red deer would select European silver fir as a food source.

Material and Method

Study area

The data used in this study is based on a long-term forest regeneration inventory in the Bavarian Forest National Park (BFNP) (Bässler et al. 2015). The national park is located in southeastern Germany (48°54' N, 13°29' E) situated in the German part of the Bohemian Forest and covering an area of 24235 ha. About 98 % of the national park is covered by forest at altitudes ranging from 300 to 1450 m a.s.l (Figure 1). The area belongs to the temperate zone and has predominantly acidic soils. The annual precipitation along the gradient ranges between 900 and 1800 mm. The mean annual temperature varies between 3.5°C at high elevations and 10.5°C at low elevations. The main tree species are European beech (*Fagus sylvatica*), silver fir (*Abies alba*), and Norway spruce (*Picea abies*). Through hundreds of years of forest management, the proportion of spruce increased artificially, while other tree species, such as silver fir, remained largely unchanged (Hilmers et al. 2018).

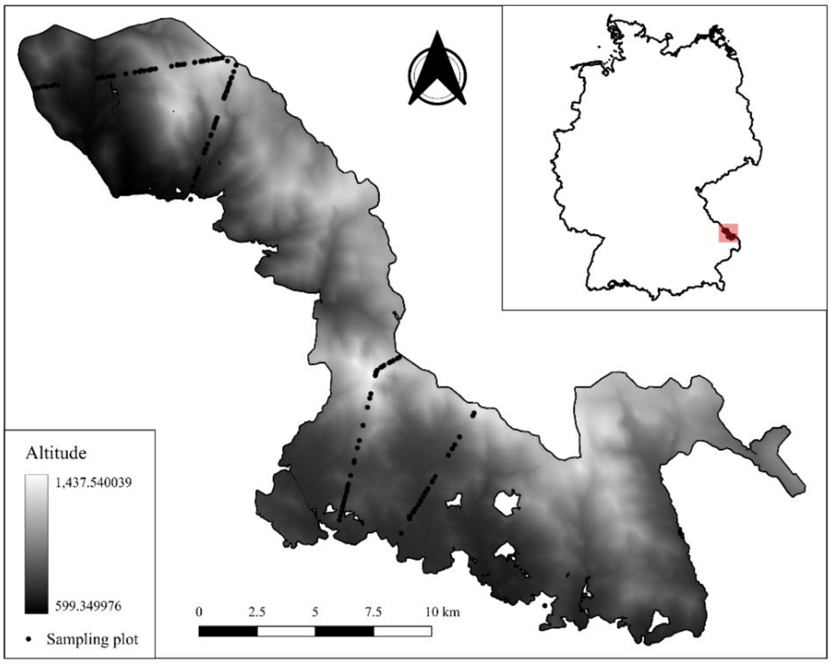

Figure 1. Location and altitude of the study area with sampling plots on which the faecal were collected. Some sampling points are not visible (overlayed) due to scaling.

Sample collection

A total of 331 plots were distributed along four straight transects across elevations ranging from 287 m a.s.l. to 1420 m a.s.l. The plots well represent the main plant communities of the National Park. On each plot, basic topographic information (e.g.

altitude, exposition) and forest structural variables (e.g. dead wood, tree species composition at different strata) were recorded. Plant species coverage was recorded at five different layers (grass, herb, shrub, tree1 and tree2). All taxonomic groups, methods and number of sampled plots (replications) are described in (Bässler et al. 2015).

Faecal samples from roe deer, red deer, and wild boar were collected on and in between the plots on the four transects during the period from May to October 2016 as described in (Spitzer et al. 2019).

DNA metabarcoding and diet composition

DNA metabarcoding was carried-out as described in (Spitzer et al. 2021). In short, DNA extraction and purification was performed on a QIASymphony SP platform using the DSP DNA minikit (Quiagen) according to the manufacturer's instructions. To confirm the deer species, we a universal primer for mammals (Mamm02 in (Taberlet et al. 2018). To determine the botanical diet composition, we used the universal primer pair Sper01_F & Sper01_R (Taberlet et al. 2018), which amplify the P6-loop of the trnL intron of chloroplasts.

Each fecal sample was run in three technical PCR replicates and all experiments included extraction controls, blanks, PCR negative and positive controls. PCR products were purified using the MinElute PCR purification kit and sequenced on an Illumina HiSeq 2500 platform using a paired-end approach (2 x 125 bp).

Sequence data were processed using OBITOOLS software (Boyer et al. 2016). Taxonomic annotation was based on a reference library of local plant species which was built by extracting the relevant parts of the EMBL nucleotide database, the NCBI taxonomy, and a database for arcto-boral plants and bryophytes (Sønstebø et al. 2010; Willerslev et al. 2014; Soininen et al. 2015). Annotated sequences were retained as molecular operational taxonomic units (MOTUs).

To confer the same weight to each sample, read abundances were converted into relative read abundances (RRA), representing the proportion of each MOTU in each sample. RRA is increasingly being used to describe the proportional composition of diets (Deagle et al. 2019; Kowalczyk et al. 2019; Churski et al. 2021) and has been shown to yield similar conclusions to quantification based on presence/absence (Kartzinel et al. 2015; Kowalczyk et al. 2019). Therefore, RRA was used as a measure of diet composition in the analyses.

Habitat suitability for lynx

The Eurasian lynx is the only large carnivore species that inhabits the study area permanently and occupies almost the entire range. As in most of Europe, the Eurasian lynx became extinct in Bavarian Forest National Park in the mid-19th century (Wölfl et al. 2001). The reintroduction began in the early 1970s, with the release of an uncertain number of animals (5 to >10) (Festetics 1980). The lynx population is stagnating at present (Müller et al. 2014), with estimated densities of 0.4 to 0.9 lynx/100 km² in the core area (Weingarth et al. 2012). The main prey of lynx is roe deer, with roe deer and red deer representing approximately 80% and 17% of lynx kills, respectively (Belotti et al. 2015).

The lynx habitat suitability map used in this thesis is based on (van Beeck Calkoen et al. 2022) (Figure 2), where the authors used the Maxent algorithm at a 30-m spatial resolution (Phillips et al. 2006) based on a total of 3679 lynx locations collected through GPS telemetry (2005–2012) from nine lynx individuals (four females and five males). The habitat suitability values predicted by Maxent (ranging from 0 to 1) reflect the relative rate of lynx occurrence (Merow et al. 2013), higher suitability values correspond to a higher predation risk for ungulates.

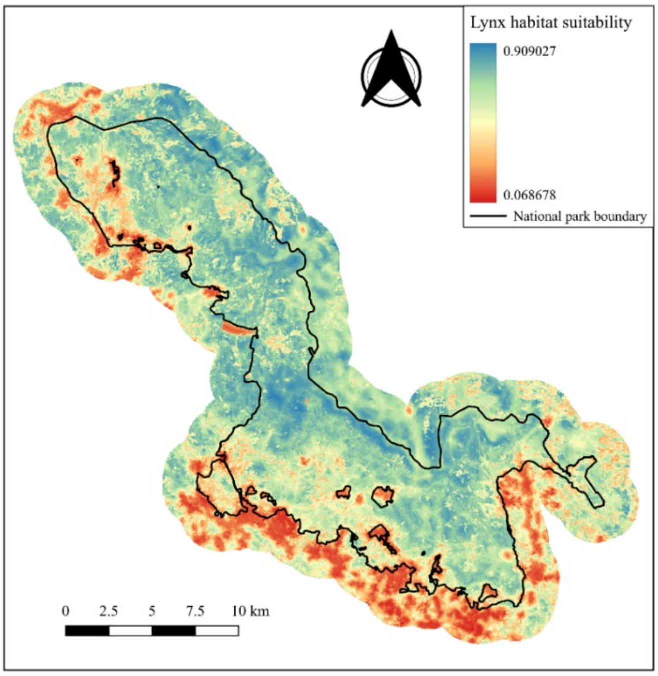

Figure 2. Lynx habitat suitability in the study area. Lynx habitat suitability values reflect the relative rate of lynx occurrence where higher suitability values correspond to a higher perceived risk of ungulate predation.

Statistical analysis

Data treatment

The dietary analysis was conducted on 153 red deer faecal pellets, 43 roe deer faecal pellets and, 82 wild boar faecal pellets samples collected in the national park. A total of 88 MOTUs were assigned at genus and higher taxonomic levels (Table S1). Additionally, MOTUs were also assigned to one of the following growth form categories: broadleaf, conifer, FLH (ferns, lycopods, horsetails), forb, graminoid, shrub, and other plants used in the subsequent analysis (Table S1). The metabarcoding markers used for the diet analysis in this study were restricted to plants so non-plant foods such as fungi, invertebrates and vertebrates could not be detected but often contribute substantially to wild boar diets (Ballari & Barrios-García 2014), so only the general diet analysis and the non-metric multidimensional scaling analysis included wild boar diets.

Dietary analysis

Morisita's index was used to quantify the niche overlap between roe deer and red deer because several studies have shown it to be the least biased overlap index (Wallace 1981; Smith & Zaret 1982). Morisita's index was calculated using the spaa package (Zhang & Zhang 2013) in R version 4.3.2 (R Core Team, 2024). The area above 1000 m a.s.l. was classified as high altitude, while the area below 1000 m a.s.l. was classified as low altitude.

For visualizing trophic resource partitioning between all three ungulate species, I first calculated Bray–Curtis dissimilarities between faecal samples based on the full taxonomic resolution of 88 MOTUs, and then ordinated these values using non-metric multidimensional scaling (NMDS) (Kartzinel et al. 2015; Pansu et al. 2019) with package vegan in R (Oksanen et al. 2013).

To obtain indices of diet diversity for red deer and roe deer diet, I calculated Shannon's diversity index and Pielou's evenness index using the vegan package in R (Oksanen et al. 2013). Generalized linear models (function glm() in R) were used to test the effects of altitude and lynx predation risk on red deer and roe deer diets. To linearize the data and improve model fit, log (y + 0.00001) transformation was used for the dependent variables graminoid, FLH, conifer, broadleaf. The assumption of normality was assessed through visual inspection of residuals in a normal quantile (QQ) plot.

Plant taxonomy groups representing ≥1% of the average diet for each ungulate species were considered key food items. As a measure of selectivity, I plotted the proportion of these key food items in deer diets against their relative abundance in the environment (i.e., their availability). The availability of each plant taxonomy group was calculated as the sum of the plot coverage in each layer (grass, herb and shrub layer) divided by the total plant coverage.

Results

General dietary information

Rosoideae and *Vaccinium* were the most dominant items in red deer diet (Figure 3). In roe deer diet, the most common dietary items belonged to *Vaccinium*, Rosoideae and *Rubus*. The most common dietary items for wild boar were *Fagus* and *Picea*.

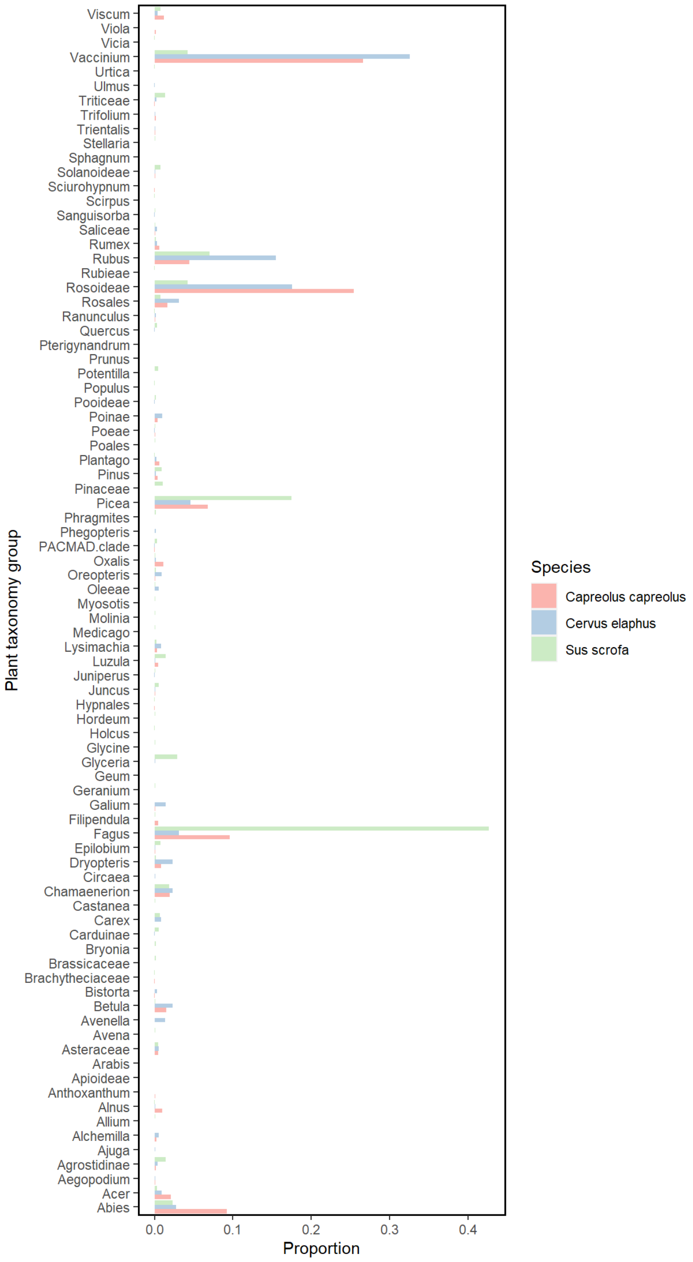

Figure 3. The average proportion of each plant taxonomy group in red deer, roe deer and wild boar diet.

Morisita's index for red deer and roe deer diet overlap for the study area was 0.93. The value in low altitude areas was 0.95 and 0.81 in high altitude areas. This suggests a large overlap between red deer and roe deer diets in the national park with some decrease in overlap with the growing altitude.

The result of NMDS indicated a clear separation of red deer and roe deer diets from wild boar diet, but – as already indicated by Morisita's index – there was a large overlap between red deer and roe deer diets (Figure 4). The stress value for the NMDS was 0.131, which, according to (Hair 2009), indicates that two dimensions were adequate to represent the variation in the measured data. The separation of wild boar diet from the deer diets was driven mainly by *Fagus* and *Picea*, while the main genera driving the clustering of red deer and roe deer diet included *Vaccinium* and *Rubus*.

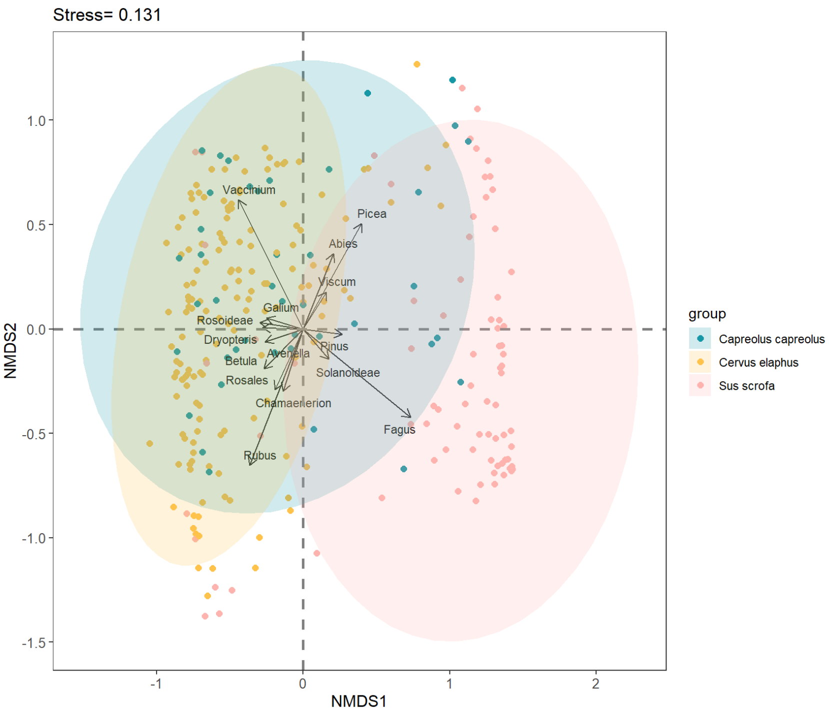

Figure 4. NMDS (non-metric multidimensional scaling) ordination plot based on Bray-Curtis dissimilarities. Arrows and labels indicate plant taxonomic groups driving the separation of points and the different colours denote the different ungulate species. The ellipses represent 95% Confidence interval.

The effect of altitudinal changes on deer diets

Both, diet diversity and diet evenness increased with altitude for red deer but not for roe deer (Figure 5, Table 1). Shrubs constituted the main food items for red deer and roe deer at high altitudes, supplemented by forbs (Figure 6). In contrast, at low altitudes, the main food item for roe deer was forbs, supplemented with conifers and shrubs. For both red deer and roe deer, the dietary proportion of forbs decreased with growing altitude, whereas the proportion of shrubs increased. The proportion of conifer in roe deer diet also decreased with the rising altitude.

*Table 1. Regression coefficients, p-values and R^2 for the relationship between Shannon index and evenness with altitude, detected using generalized linear models. The p-values are indicated with stars: ns: $p > 0.05$, *: $p <= 0.05$, **: $p <= 0.01$, ***: $p <= 0.001$*

Species	Index	p-value	R^2	Regression coefficient
Roe deer	Shannon	ns	-0.0211	-0.0002
	Evenness	ns	-0.0230	0.0000
Red deer	Shannon	***	0.0922	0.0005
	Evenness	***	0.1657	0.0003

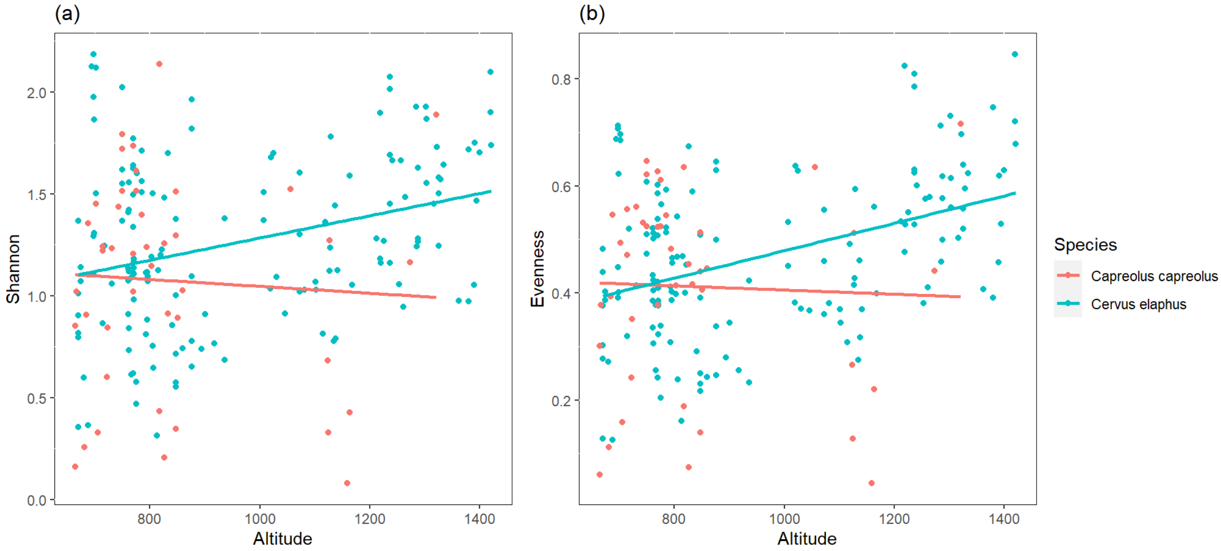

Figure 5. Relationship between a change in altitude and (a) the Shannon index (diet diversity) and (b) diet evenness. Each point represents a faecal sample; colours indicate the deer species.

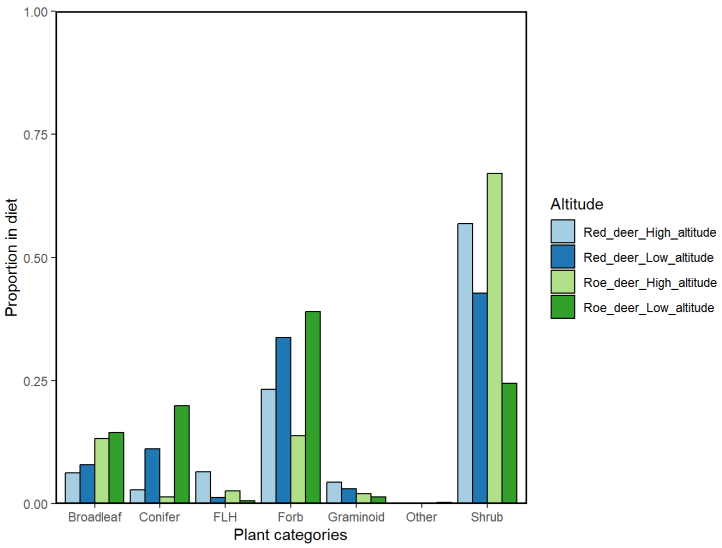

Figure 6. The average proportion of each plant category in red deer and roe deer in high (>1000 m a.s.l.) and low (<1000 m a.s.l.) altitudes respectively. FLH refers to ferns, lycopods, and horsetails.

The effect of lynx occurrence on deer diets

The diet of roe deer changed with the presence of lynx, whereas there was no big difference in red deer diet. The proportion of the forb in the roe deer diet decreased significantly ($P < 0.05$) with the lynx presence in the national park (Figure 7, Table 2). In areas with high lynx abundance, roe deer tended to eat more shrubs and broadleaves than forbs.

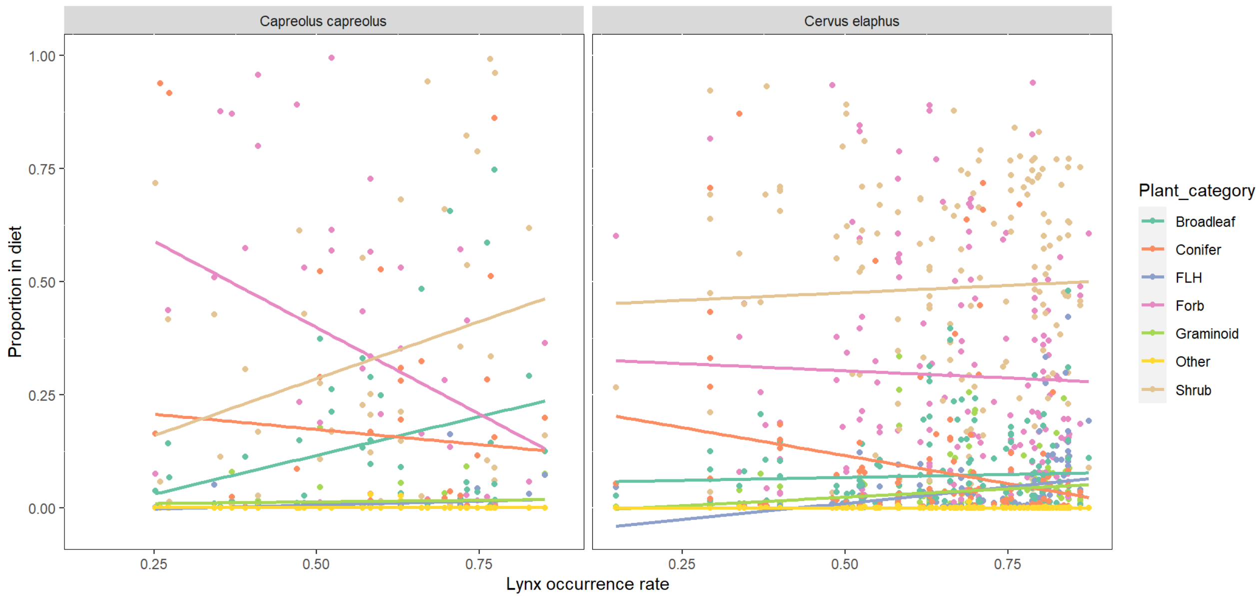

Figure 7. Relationship between the proportion of each food category in roe deer and red deer diet with lynx occurrence rate. Each point represents a faecal sample.

*Table 2. Effects of altitude and lynx occurrence rate on each plant category, detected using generalized linear models. The p-values are indicated with stars: ns: p > 0.05, *: p < = 0.05, **: p < = 0.01, ***: p < = 0.001*

	Lynx predation risk				Altitude			
Red deer	Estimate	Std. Error	t value	p-value	Estimate	Std. Error	t value	p-value
Graminoid	6.669	1.689	3.948	***	0.004	0.001	4.202	***
Forb	-0.063	0.123	-0.512	ns	0.000	0.002	-2.030	*
Shrub	0.066	0.127	0.520	ns	0.000	0.000	3.014	**
FLH	6.118	1.433	4.270	***	0.006	0.001	6.875	***
Broadleaf	-2.436	1.110	-2.194	*	-0.003	0.001	-4.772	***
Conifer	-5.881	1.304	-4.509	***	-0.006	0.001	-6.873	***
Roe deer								
Graminoid	3.765	3.101	1.214	ns	0.005	0.003	1.782	ns
Forb	-0.764	0.266	-2.876	**	0.000	0.000	-1.444	ns
Shrub	0.504	0.264	1.905	ns	0.001	0.000	3.619	***
FLH	0.070	2.992	0.023	ns	0.004	0.003	1.225	ns
Broadleaf	2.908	1.722	1.688	ns	-0.001	0.002	-0.839	ns
Conifer	2.307	2.544	0.907	ns	-0.004	0.002	-1.518	ns

Diet Selectivity

My results suggest selective use of several forages in the study area. Rosaceae and *Vaccinium* were preferred by both deer species, while *Avenella*, *Betula*, *Fagus* and *Picea* are avoided by red deer and roe deer. Red deer showed no preference for *Abies*, whereas roe deer preferred to eat *Abies* (Figure 8).

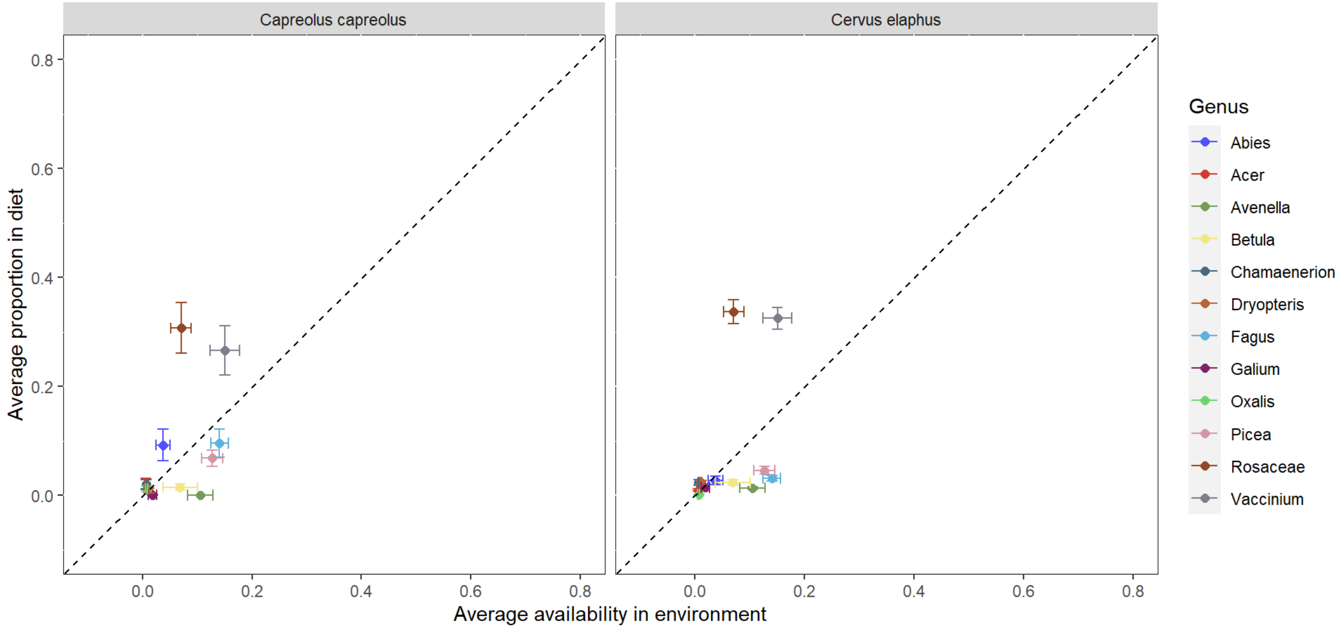

Figure 8. Scatterplot with 1:1 line (dashed) of the relationship between availability and proportion of plant taxonomy group in red deer and roe deer diets contributing > 1% to the average diet. The error bars indicate the standard deviation. Values above the 1:1 indicate larger proportions of a food item in the diet than what would be expected from its relative abundance in the environment (i.e., 'preference'), whereas values below the line indicate less-than-expected use ('avoidance'). Selection for items on the line is neutral.

Discussion

The result of this thesis suggests that deer diets were not constant throughout the national park. The diet composition of the two deer species was similar within the study area but varied among altitudes. Red deer diet diversity increases with altitude and roe deer diet changes significantly with predation risk.

Studies have shown that temperature was the most important factor explaining the variability in vascular plant community composition in the Bavarian Forest National Park, which changed discretely along the altitudinal gradient (Bässler et al. 2010). Studies have also shown that the number of plant species decreases with elevation in BFNP (Friess et al. 2018). Red deer and roe deer diets overlap more in high altitude areas. This result is similar to those niche studies, where overlap is greatest when food supply is abundant and least when food is limited and competition increased (Schoener 1982; Gordon & Illius 1989).

The diet evenness of the red deer diets increased with increasing altitude and plant species richness in the environment decreased, which agrees with my first hypothesis for altitudinal relationships. There was a reduction in diversity in red deer diet following the increase in altitude; this tendency was also detected in herbivores of Andean Precordillera (Puig et al. 2014). Red deer diet was more specialised (more diverse) at the lower altitudes, more enhanced opportunity for dietary selection, gives higher prevalence of the few preferred items that maintained acceptable availability and tenderness. As an intermediate feeder, red deer can switch between browse and grass dominated diets (Kerridge & Bullock 1991; Krojerová-Prokešová et al. 2010). Red deer are thought to be browsers when overall browse quality and availability are high and switch to a grass dominated diet in response to declines in concentrates (Dumont et al. 2005; Verheyden-Tixier et al. 2008). This agrees with my results that red deer tend to eat more forb at higher altitudes and more shrub at lower altitudes.

My results on roe deer diet shifts fit with the second hypothesis (Figure 7; Table 2). There was a significant increase in the proportion of shrub species and a decrease in the proportion of forb species in roe deer diet in high lynx occurrence areas. While red deer did not show similar shifts in diet, the proportions of graminoids, conifers and FLH changed significantly with lynx presence, with more graminoids and FLH being consumed and less conifers. Roe deer is the preferred prey species of lynx, while wolf is the main predator for adult red deer which are rarely predated by lynx in Bavarian Forest National Park (Belotti et al. 2015; Heurich et al. 2016). Studies have shown that large carnivores may create a landscape of fear for ungulates (Brown et al. 1999; Brown & Kotler 2004; Gaynor et al. 2019), but only a few have looked directly at changes in the diet of large ungulates in response to the risk of predation (Hernández & Laundré 2005; Christianson & Creel 2008; Barnier et al. 2014; Churski et al. 2021). Several studies have found that both roe deer and red deer respond to predator presence by increasing their vigilance (Eccard et al. 2017) or decreasing visitation duration (Wikenros et al. 2015; van Beeck Calkoen et al. 2021). Churski et al. (2021) suggest that deer increase vigilance by changing their foraging height upwards, i.e., increase browsing on taller food item such as shrubs and trees when vigilant and keeping their heads up. By foraging with their head-up, the prey species is able to maintain a good overview of their surroundings and have better chances of detecting approaching large predators (Kohl et al. 2018). To further support this pattern, future studies could also focus on how wolves affect the diet of red deer in the National Park, as wolves are the main predator of adult red deer.

One explanation for the trends in red deer diets could be due to lynx habitat modelling, only night-time positions of lynx were used for creating habitat suitability models (van Beeck Calkoen et al. 2022). Lynx prefers open habitats, such as meadows during night-time (Filla et al. 2017). There may be consistent differences in the availability of food plants between areas of high and low lynx occurrence areas. Another could be they have different strategies when facing lynx predation risk: red deer may show reduced visitation duration ('leave risky sites'), whereas roe deer become only alert ('stay but alert') when faced with a stalking predator. Previous studies suggested that red deer leave the site in the presence of lynx, which means red deer try to avoid areas where lynx are present (Wikenros et al. 2015).

For the third hypothesis that deer species would select European silver fir as a food source, roe deer agreed with the hypothesis, but red deer did not. The results of roe deer diet analyses showed their high preference for woody plants and forbs, similar patterns were also observed in other studies (Helle 1980; Jackson 1980; Barančeková et al. 2009). The result also suggests that roe deer actively search for silver fir (*Abies alba*) while red deer do not, hence, it is a preferred food plant for roe deer. The discrepancy in food choices between ungulate species may result from species-specific differences in the capacity to detoxify plant secondary compounds. Roe deer as a browser is adapted to detoxify high levels of secondary compounds (Hofmann 1989), red deer as an intermediate feeder may be less able to do so. European silver fir is the most heavily browsed conifer species in Central European mountain forests and is an important economically and ecologically species.

In Europe, the increasing numbers of ungulates have led to poor regeneration success of this species (e.g. Motta 1996). The major silvicultural concern of this species is the poor survival of young trees (Senn & Suter 2003). Pellerin et al. (2010) suggested that the number of saplings of silver fir can be negatively affected by deer browsing. It is speculated that in areas with high abundances of roe deer, forage might become limited, thus silver fir might be browsed more intensively. To reduce browsing pressure on silver fir, keeping roe deer density below carrying capacity is essential.

Understanding how predation risk and altitude influence deer diets can also help in developing strategies for forest management, to maintain healthy forest ecosystems and ensure the survival of important tree species like the European silver fir. However, in human-dominated landscapes, the increasingly large impact of human activities must also be considered. Higher human activity such as forestry, may modify the effects of natural gradients, which could result in changes to the composition of ungulate diets. (van Beeck Calkoen et al. 2022) suggest that deer species showed a stronger selection of tree species under higher human risk (close to human settlements and with hunting intensity). Moreover, studies have shown that the impact of human activity on ungulate behaviour can even exceed those of habitat and natural predators (Ciuti et al. 2012; van Beeck Calkoen et al. 2022). Therefore, further studies of how human activity affects ungulate diets are needed to ensure the protection of the national park.

References

Ammer, C. (1996). Impact of ungulates on structure and dynamics of natural regeneration of mixed mountain forests in the Bavarian Alps. *Forest Ecology and Management*, 88 (1), 43–53. https://doi.org/10.1016/S0378-1127(96)03808-X

Apollonio, M., Andersen, R. & Putman, R. (2010). *European Ungulates and Their Management in the 21st Century*. Cambridge University Press.

Apps, C.D., McLellan, B.N., Kinley, T.A. & Flaa, J.P. (2001). Scale-Dependent Habitat Selection by Mountain Caribou, Columbia Mountains, British Columbia. *The Journal of Wildlife Management*, 65 (1), 65–77. https://doi.org/10.2307/3803278

Ballari, S.A. & Barrios-García, M.N. (2014). A review of wild boar *Sus scrofa* diet and factors affecting food selection in native and introduced ranges. *Mammal Review*, 44 (2), 124–134. https://doi.org/10.1111/mam.12015

Barančeková, M., Krojerová-Prokešová, J., Šustr, P. & Heurich, M. (2009). Annual changes in roe deer (*Capreolus capreolus* L.) diet in the Bohemian Forest, Czech Republic/Germany. European Journal *of Wildlife Research*, 56 (3), 327–333. https://doi.org/10.1007/s10344-009-0321-0

Barnier, F., Valeix, M., Duncan, P., Chamaillé-Jammes, S., Barre, P., Loveridge, A.J., Macdonald, D.W. & Fritz, H. (2014). Diet quality in a wild grazer declines under the threat of an ambush predator. *Proceedings of the Royal Society B: Biological Sciences*, 281 (1785), 20140446. https://doi.org/10.1098/rspb.2014.0446

Bässler, C., Müller, J. & Dziock, F. (2010). Detection of Climate-Sensitive Zones and Identification of Climate Change Indicators: A Case Study from the Bavarian Forest National Park. *Folia Geobotanica*, 45 (2), 163–182. https://doi.org/10.1007/s12224-010-9059-4

Bässler, C., Seifert, L. & Müller, J. (2015). The BIOKLIM Project in the National Park Bavarian Forest: Lessons from a biodiversity survey. *Silva Gabreta*, 21.

van Beeck Calkoen, S.T.S., Deis, M.H., Oeser, J., Kuijper, D.P.J. & Heurich, M. (2022). Humans rather than Eurasian lynx (*Lynx lynx*) shape ungulate browsing patterns in a temperate forest. *Ecosphere*, 13 (2), e3931. https://doi.org/10.1002/ecs2.3931

van Beeck Calkoen, S.T.S., Kreikenbohm, R., Kuijper, D.P.J. & Heurich, M. (2021). Olfactory cues of large carnivores modify red deer behavior and browsing intensity. *Behavioral Ecology*, 32 (5), 982–992. https://doi.org/10.1093/beheco/arab071

Belotti, E., Weder, N., Bufka, L., Kaldhusdal, A., Küchenhoff, H., Seibold, H., Woelfing, B. & Heurich, M. (2015). Patterns of Lynx Predation at the Interface between Protected Areas and Multi-Use Landscapes in Central Europe. *PLOS ONE*, 10 (9), e0138139. https://doi.org/10.1371/journal.pone.0138139

Bernes, C., Macura, B., Jonsson, B.G., Junninen, K., Müller, J., Sandström, J., Lõhmus, A. & Macdonald, E. (2018). Manipulating ungulate herbivory in temperate and boreal forests: effects on vegetation and invertebrates. A systematic review. *Environmental Evidence*, 7 (1), 13. https://doi.org/10.1186/s13750-018-0125-3

Boyer, F., Mercier, C., Bonin, A., Le Bras, Y., Taberlet, P. & Coissac, E. (2016). obitools: a unix-inspired software package for DNA metabarcoding. *Molecular Ecology Resources*,

16 (1), 176–182. https://doi.org/10.1111/1755-0998.12428
Bradshaw, R.H.W., Hannon, G.E. & Lister, A.M. (2003). A long-term perspective on ungulate–vegetation interactions. *Forest Ecology and Management*, 181 (1), 267–280. https://doi.org/10.1016/S0378-1127(03)00138-5
Brown, J.S. & Kotler, B.P. (2004). Hazardous duty pay and the foraging cost of predation. *Ecology Letters*, 7 (10), 999–1014. https://doi.org/10.1111/j.1461-0248.2004.00661.x
Brown, J.S., Laundré, J.W. & Gurung, M. (1999). The Ecology of Fear: Optimal Foraging, Game Theory, and Trophic Interactions. *Journal of Mammalogy*, 80 (2), 385–399. https://doi.org/10.2307/1383287
Bruinderink, G.W.T.A.G. & Hazebroek, E. (1996). Wild boar (*Sus scrofa scrofa* L.) rooting and forest regeneration on podzolic soils in the Netherlands. *Forest Ecology and Management*, 88 (1), 71–80. https://doi.org/10.1016/S0378-1127(96)03811-X
Carothers, J., Jaksic, F. & Marquet, P. (1996). A model for species distributions along a gradient: Lizards as study systems. *Revista Chilena de Historia Natural*, 69, 301–307
Cederlund, G., Ljungqvist, H., Markgren, G., & Stalfelt, F. (1980). Foods of moose and roe-deer at Grimsö in central Sweden-results of rumen content analyses.
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 (6216), 1517–1519. https://doi.org/10.1126/science.1257553
Christianson, D. & Creel, S. (2008). Risk effects in elk: sex-specific responses in grazing and browsing due to predation risk from wolves. *Behavioral Ecology*, 19 (6), 1258–1266. https://doi.org/10.1093/beheco/arn079
Churski, M., Spitzer, R., Coissac, E., Taberlet, P., Lescinskaite, J., van Ginkel, H.A.L., Kuijper, D.P.J. & Cromsigt, J.P.G.M. (2021). How do forest management and wolf space-use affect diet composition of the wolf's main prey, the red deer versus a non-prey species, the European bison? *Forest Ecology and Management*, 479, 118620. https://doi.org/10.1016/j.foreco.2020.118620
Ciuti, S., Northrup, J.M., Muhly, T.B., Simi, S., Musiani, M., Pitt, J.A. & Boyce, M.S. (2012). Effects of Humans on Behaviour of Wildlife Exceed Those of Natural Predators in a Landscape of Fear. *PLOS ONE*, 7 (11), e50611. https://doi.org/10.1371/journal.pone.0050611
Clauss, M., Hume, I.D. & Hummel, J. (2010). Evolutionary adaptations of ruminants and their potential relevance for modern production systems. *animal*, 4 (7), 979–992. https://doi.org/10.1017/S1751731110000388
Cope, E.D. (1882). The Classification of the Ungulate Mammalia. *Proceedings of the American Philosophical Society*, 20 (112), 438–447

Cornelis, J., Casaer, J. & Hermy, M. (1999). Impact of season, habitat and research techniques on diet composition of roe deer (*Capreolus capreolus*): A review. *Journal of Zoology*, 248. https://doi.org/10.1111/j.1469-7998.1999.tb01196.x

Deagle, B.E., Thomas, A.C., McInnes, J.C., Clarke, L.J., Vesterinen, E.J., Clare, E.L., Kartzinel, T.R. & Eveson, J.P. (2019). Counting with DNA in metabarcoding studies: How should we convert sequence reads to dietary data? *Molecular Ecology*, 28 (2), 391–406. https://doi.org/10.1111/mec.14734

Deinet, S., Ieronymidou, C., McRae, L., Burfield, I.J., Foppen, R.P., Collen, B. & Böhm, M. (2013). *Wildlife Comeback in Europe: The recovery of selected mammal and bird species*. <bound method Organization.get_name_with_acronym of <Organization: Rewilding Europe>>. https://policycommons.net/artifacts/2677362/download-2013/3700562/ [2024-04-09]

Dobrowolska, D., Bončina, A., & Klumpp, R. (2017). Ecology and silviculture of silver fir (*Abies alba* Mill.): A review. *Journal of Forest Research*, 22(6), 326–335. https://doi.org/10.1080/13416979.2017.1386021

Dumont, B., Renaud, P.-C., Morellet, N., Mallet, C., Anglard, F. & Verheyden-Tixier, H. (2005). Seasonal variations of Red Deer selectivity on a mixed forest edge. *Animal Research*, 54 (5), 369–381. https://doi.org/10.1051/animres:2005030

Eccard, J.A., Meißner, J.K. & Heurich, M. (2017). European Roe Deer Increase Vigilance When Faced with Immediate Predation Risk by Eurasian Lynx. *Ethology*, 123 (1), 30–40. https://doi.org/10.1111/eth.12420

Erschbamer, B., Mallaun, M. & Unterluggauer, P. (2006). Plant diversity along altitudinal gradients in the Southern and Central Alps of South Tyrol and Trentino (Italy). 6

Festetics, A. (1980). Die Wiedereinbürgerung des Luchses in Europa. Der Luchs in Europa-Verbreitung, Wiedereinbürgerung, Räuber-Beute-Beziehung. Kilda Verlag, Greven.

Filla, M., Premier, J., Magg, N., Dupke, C., Khorozyan, I., Waltert, M., Bufka, L. & Heurich, M. (2017). Habitat selection by Eurasian lynx (*Lynx lynx*) is primarily driven by avoidance of human activity during day and prey availability during night. *Ecology and Evolution*, 7 (16), 6367–6381. https://doi.org/10.1002/ece3.3204

Friess, N., Bässler, C., Brandl, R., Hilmers, T., Müller, J. & Seifert, L. (2018). Biodiversity along an elevational gradient of the Bohemian Forest – the BIOKLIM project.

Fuller, R.J. (2001). Responses of woodland birds to increasing numbers of deer: a review of evidence and mechanisms. *Forestry: An International Journal of Forest Research*, 74 (3), 289–298. https://doi.org/10.1093/forestry/74.3.289

Gaynor, K.M., Brown, J.S., Middleton, A.D., Power, M.E. & Brashares, J.S. (2019). Landscapes of Fear: Spatial Patterns of Risk Perception and Response. *Trends in Ecology & Evolution*, 34 (4), 355–368. https://doi.org/10.1016/j.tree.2019.01.004

Genov, P. (1981). Food composition of wild boar in north-eastern and western Poland. *Acta Theriologica*, 26, 185–205. https://doi.org/10.4098/AT.arch.81-16

Gentry, A.H. (1988). Changes in Plant Community Diversity and Floristic Composition on Environmental and Geographical Gradients. *Annals of the Missouri Botanical Garden*, 75 (1), 1–34. https://doi.org/10.2307/2399464

GILL, R.M.A. (1992). A Review of Damage by Mammals in North Temperate Forests: 3. Impact on Trees and Forests. *Forestry: An International Journal of Forest Research*, 65 (4), 363–388. https://doi.org/10.1093/forestry/65.4.363-a

Gordon, I.J. & Illius, A.W. (1989). Resource partitioning by ungulates on the Isle of Rhum.

Oecologia, 79 (3), 383–389. https://doi.org/10.1007/BF00384318

Grytnes, J.A. & Vetaas, O.R. (2002). Species Richness and Altitude: A Comparison between Null Models and Interpolated Plant Species Richness along the Himalayan Altitudinal Gradient, Nepal. *The American Naturalist*, 159 (3), 294–304. https://doi.org/10.1086/338542

Hair, J. (2009). Multivariate Data Analysis. *Faculty and Research Publications*.

Heckel, C.D., Bourg, N.A., McShea, W.J. & Kalisz, S. (2010). Nonconsumptive effects of a generalist ungulate herbivore drive decline of unpalatable forest herbs. *Ecology*, 91 (2), 319–326. https://doi.org/10.1890/09-0628.1

Helle, P. (1980). Food composition and feeding habits of the roe deer in winter in central Finland. *Acta Theriologica*, 25, 395–402. https://doi.org/10.4098/AT.arch.80-36

Hernández, L. & Laundré, J.W. (2005). Foraging in the 'landscape of fear' and its implications for habitat use and diet quality of elk *Cervus elaphus* and bison *Bison bison*. *Wildlife Biology*, 11 (3), 215–220. https://doi.org/10.2981/0909-6396(2005)11[215:FITLOF]2.0.CO;2

Heurich, M., Baierl, F., Günther, S. & Sinner, K. (2011). Management and conservation of large mammals in the Bavarian Forest National Park. *Silva Gabreta*, 17, 1–18

Heurich, M., Zeis, K., Küchenhoff, H., Müller, J., Belotti, E., Bufka, L. & Woelfing, B. (2016). Selective Predation of a Stalking Predator on Ungulate Prey. *PLOS ONE*, 11 (8), e0158449. https://doi.org/10.1371/journal.pone.0158449

Hofmann, R.R. (1989). Evolutionary steps of ecophysiological adaptation and diversification of ruminants: a comparative view of their digestive system. *Oecologia*, 78 (4), 443–457. https://doi.org/10.1007/BF00378733

Ihl, C. & Klein, D.R. (2001). Habitat and Diet Selection by Muskoxen and Reindeer in Western Alaska. *The Journal of Wildlife Management*, 65 (4), 964–972. https://doi.org/10.2307/3803045

Jackson, J. (1980). The annual diet of the Roe deer (*Capreolus capreolus*) in the New Forest, Hampshire, as determined by rumen content analysis. *Journal of Zoology*, 192 (1), 71–83. https://doi.org/10.1111/j.1469-7998.1980.tb04220.x

Kartzinel, T.R., Chen, P.A., Coverdale, T.C., Erickson, D.L., Kress, W.J., Kuzmina, M.L., Rubenstein, D.I., Wang, W. & Pringle, R.M. (2015). DNA metabarcoding illuminates dietary niche partitioning by African large herbivores. *Proceedings of the National Academy of Sciences*, 112 (26), 8019–8024. https://doi.org/10.1073/pnas.1503283112

Kerridge, F.J. & Bullock, D.J. (1991). Diet and dietary quality of red deer and fallow deer in late summer. *Journal of Zoology*, 224 (2), 333–337. https://doi.org/10.1111/j.1469-7998.1991.tb04812.x

Kohl, M.T., Stahler, D.R., Metz, M.C., Forester, J.D., Kauffman, M.J., Varley, N., White, P.J., Smith, D.W. & MacNulty, D.R. (2018). Diel predator activity drives a dynamic landscape of fear. *Ecological Monographs*, 88 (4), 638–652. https://doi.org/10.1002/ecm.1313

Kowalczyk, R., Wójcik, J.M., Taberlet, P., Kamiński, T., Miquel, C., Valentini, A., Craine, J.M. & Coissac, E. (2019). Foraging plasticity allows a large herbivore to persist in a sheltering forest habitat: DNA metabarcoding diet analysis of the European bison. *Forest Ecology and Management*, 449, 117474. https://doi.org/10.1016/j.foreco.2019.117474

Krojerová-Prokešová, J., Barančeková, M., Šustr, P. & Heurich, M. (2010). Feeding patterns of red deer *Cervus elaphus* along an altitudinal gradient in the Bohemian Forest: effect of habitat and season. *Wildlife Biology*, 16 (2), 173–184. https://doi.org/10.2981/09-004

Kuijper, D.P.J. (2011). Lack of natural control mechanisms increases wildlife–forestry conflict in managed temperate European forest systems. *European Journal of Forest Research*, 130 (6), 895–909. https://doi.org/10.1007/s10342-011-0523-3

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. *Biological Conservation*, 244, 108500. https://doi.org/10.1016/j.biocon.2020.108500

Lowrey, B., Garrott, R.A., McWhirter, D.E., White, P.J., DeCesare, N.J. & Stewart, S.T. (2018). Niche similarities among introduced and native mountain ungulates. *Ecological Applications*, 28 (5), 1131–1142. https://doi.org/10.1002/eap.1719

Martínez, O. & Rechberger, J. (2007). Características de la avifauna en un gradiente altitudinal de un bosque nublado andino en La Paz, Bolivia. *Revista Peruana de Biología*, 14 (2), 225–236

Merow, C., Smith, M.J. & Silander Jr, J.A. (2013). A practical guide to MaxEnt for modeling species' distributions: what it does, and why inputs and settings matter. *Ecography*, 36 (10), 1058–1069. https://doi.org/10.1111/j.1600-0587.2013.07872.x

Milner, J.M., Bonenfant, C., Mysterud, A., Gaillard, J.-M., Csányi, S. & Stenseth, N.Chr. (2006). Temporal and spatial development of red deer harvesting in Europe: biological and cultural factors. *Journal of Applied Ecology*, 43 (4), 721–734. https://doi.org/10.1111/j.1365-2664.2006.01183.x

Möst, L., Hothorn, T., Müller, J. & Heurich, M. (2015). Creating a landscape of management: Unintended effects on the variation of browsing pressure in a national park. *Forest Ecology and Management*, 338, 46–56. https://doi.org/10.1016/j.foreco.2014.11.015

Motta, R. (1996). Impact of wild ungulates on forest regeneration and tree composition of mountain forests in the Western Italian Alps. *Forest Ecology and Management*, 88 (1), 93–98. https://doi.org/10.1016/S0378-1127(96)03814-5

Müller, J., Wölfl, M., Wölfl, S., Müller, D.W.H., Hothorn, T. & Heurich, M. (2014). Protected areas shape the spatial distribution of a European lynx population more than 20 years after reintroduction. *Biological Conservation*, 177, 210–217. https://doi.org/10.1016/j.biocon.2014.07.007

Oksanen, J., Blanchet, F.G., Kindt, R., Legendre, P., Minchin, P.R., O'hara, R.B., Simpson, G.L., Solymos, P., Stevens, M.H.H. & Wagner, H. (2013). Community ecology package. *R package version*, 2 (0), 321–326

Pansu, J., Guyton, J.A., Potter, A.B., Atkins, J.L., Daskin, J.H., Wursten, B., Kartzinel, T.R. & Pringle, R.M. (2019). Trophic ecology of large herbivores in a reassembling African ecosystem. *Journal of Ecology*, 107 (3), 1355–1376. https://doi.org/10.1111/1365-2745.13113

Pellerin, M., Saïd, S., Richard, E., Hamann, J.-L., Dubois-Coli, C. & Hum, P. (2010). Impact of deer on temperate forest vegetation and woody debris as protection of forest regeneration against browsing. *Forest Ecology and Management*, 260 (4), 429–437. https://doi.org/10.1016/j.foreco.2010.04.031

Pellissier, L., Fiedler, K., Ndribe, C., Dubuis, A., Pradervand, J.-N., Guisan, A. & Rasmann, S. (2012). Shifts in species richness, herbivore specialization, and plant resistance along elevation gradients. *Ecology and Evolution*, 2 (8), 1818–1825. https://doi.org/10.1002/ece3.296

Phillips, S.J., Anderson, R.P. & Schapire, R.E. (2006). Maximum entropy modeling of species geographic distributions. *Ecological Modelling*, 190 (3), 231–259. https://doi.org/10.1016/j.ecolmodel.2005.03.026

Puig, S., Rosi, M.I., Videla, F. & Mendez, E. (2014). Food selection by the guanaco (*Lama guanicoe*) along an altitudinal gradient in the Southern Andean Precordillera (Argentina). *Acta Theriologica*, 59 (4), 541–551. https://doi.org/10.1007/s13364-014-0191-z

Ramirez, J.I., Jansen, P.A. & Poorter, L. (2018). Effects of wild ungulates on the regeneration, structure and functioning of temperate forests: A semi-quantitative review. *Forest Ecology and Management*, 424, 406–419. https://doi.org/10.1016/j.foreco.2018.05.016

R Core Team, 2024. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria https://www.R-project.org/.

Reimoser, F. (2003). Steering the impacts of ungulates on temperate forests. *Journal for Nature Conservation*, 10 (4), 243–252. https://doi.org/10.1078/1617-1381-00024

Ripple, W.J., Estes, J.A., Beschta, R.L., Wilmers, C.C., Ritchie, E.G., Hebblewhite, M., Berger, J., Elmhagen, B., Letnic, M., Nelson, M.P., Schmitz, O.J., Smith, D.W., Wallach, A.D. & Wirsing, A.J. (2014). Status and Ecological Effects of the World's Largest Carnivores. *Science*, 343 (6167), 1241484. https://doi.org/10.1126/science.1241484

Ripple, W.J., Newsome, T.M., Wolf, C., Dirzo, R., Everatt, K.T., Galetti, M., Hayward, M.W., Kerley, G.I.H., Levi, T., Lindsey, P.A., Macdonald, D.W., Malhi, Y., Painter, L.E., Sandom, C.J., Terborgh, J. & Van Valkenburgh, B. (2015). Collapse of the world's largest herbivores. *Science Advances*, 1 (4), e1400103. https://doi.org/10.1126/sciadv.1400103

Schoener, T.W. (1982). The Controversy over Interspecific Competition: Despite spirited criticism, competition continues to occupy a major domain in ecological thought. *American Scientist*, 70 (6), 586–595

Schwartz, C.C. & Ellis, J.E. (1981). Feeding Ecology and Niche Separation in Some Native and Domestic Ungulates on the Shortgrass Prairie. *Journal of Applied Ecology*, 18 (2), 343–353. https://doi.org/10.2307/2402399

Senft, R.L., Coughenour, M.B., Bailey, D.W., Rittenhouse, L.R., Sala, O.E. & Swift, D.M. (1987). Large Herbivore Foraging and Ecological Hierarchies. *BioScience*, 37 (11), 789–799. https://doi.org/10.2307/1310545

Senn, J. & Suter, W. (2003). Ungulate browsing on silver fir (*Abies alba*) in the Swiss Alps: beliefs in search of supporting data. *Forest Ecology and Management*, 181 (1), 151–164. https://doi.org/10.1016/S0378-1127(03)00129-4

Smith, E.P. & Zaret, T.M. (1982). Bias in Estimating Niche Overlap. *Ecology*, 63 (5), 1248–1253. https://doi.org/10.2307/1938851

Soininen, E.M., Gauthier, G., Bilodeau, F., Berteaux, D., Gielly, L., Taberlet, P., Gussarova, G., Bellemain, E., Hassel, K., Stenøien, H.K., Epp, L., Schrøder-Nielsen, A., Brochmann, C. & Yoccoz, N.G. (2015). Highly Overlapping Winter Diet in Two Sympatric Lemming Species Revealed by DNA Metabarcoding. *PLOS ONE*, 10 (1), e0115335. https://doi.org/10.1371/journal.pone.0115335

Sønstebø, J.H., Gielly, L., Brysting, A.K., Elven, R., Edwards, M., Haile, J., Willerslev, E., Coissac, E., Rioux, D., Sannier, J., Taberlet, P. & Brochmann, C. (2010). Using next-generation sequencing for molecular reconstruction of past Arctic vegetation and climate. *Molecular Ecology Resources*, 10 (6), 1009–1018. https://doi.org/10.1111/j.1755-0998.2010.02855.x

Spitzer, R., Churski, M., Felton, A., Heurich, M., Kuijper, D.P.J., Landman, M., Rodriguez, E., Singh, N.J., Taberlet, P., van Beeck Calkoen, S.T.S., Widemo, F. & Cromsigt, J.P.G.M. (2019). Doubting dung: eDNA reveals high rates of misidentification in diverse European ungulate communities. *European Journal of Wildlife Research*, 65 (2), 28. https://doi.org/10.1007/s10344-019-1264-8

Spitzer, R., Coissac, E., Felton, A., Fohringer, C., Juvany, L., Landman, M., Singh, N.J., Taberlet, P., Widemo, F. & P.G.M. Cromsigt, J. (2021). Small shrubs with large importance? Smaller deer may increase the moose-forestry conflict through feeding competition over *Vaccinium* shrubs in the field layer. *Forest Ecology and Management*, 480, 118768. https://doi.org/10.1016/j.foreco.2020.118768

Spitzer, R., Felton, A., Landman, M., Singh, N.J., Widemo, F. & Cromsigt, J.P.G.M. (2020). Fifty years of European ungulate dietary studies: a synthesis. *Oikos*, 129 (11), 1668–1680. https://doi.org/10.1111/oik.07435

Stewart, K.M., Bowyer, R.T., Kie, J.G., Cimon, N.J. & Johnson, B.K. (2002). Temporospatial Distributions of Elk, Mule Deer, and Cattle: Resource Partitioning and Competitive Displacement. *Journal of Mammalogy*, 83 (1), 229–244. https://doi.org/10.1644/1545-1542(2002)083<0229:TDOEMD>2.0.CO;2

Taberlet, P., Bonin, A., Zinger, L. & Coissac, E. (2018). *Environmental DNA: For Biodiversity Research and Monitoring*. Oxford University Press.

Terborgh, J. (1977). Bird Species Diversity on an Andean Elevational Gradient. *Ecology*, 58 (5), 1007–1019. https://doi.org/10.2307/1936921

Tixier, H. & Duncan, P. (1996). Are European roe deer browsers? A review of variations in the composition of their diets. *Revue d'Écologie*, 51 (1), 3–17

Van Hees, A.F.M., Kuiters, A.T. & Slim, P.A. (1996). Growth and development of silver birch, pedunculate oak and beech as affected by deer browsing. *Forest Ecology and Management*, 88 (1), 55–63. https://doi.org/10.1016/S0378-1127(96)03809-1

Verheyden-Tixier, H., Renaud, P.-C., Morellet, N., Jamot, J., Besle, J.-M. & Dumont, B. (2008). Selection for nutrients by red deer hinds feeding on a mixed forest edge. *Oecologia*, 156 (3), 715–726. https://doi.org/10.1007/s00442-008-1020-3

Wallace, R.K. (1981). An Assessment of Diet-Overlap Indexes. *Transactions of the American Fisheries Society*, 110 (1), 72–76. https://doi.org/10.1577/1548-8659(1981)110<72:AAODI>2.0.CO;2

Weingarth, K., Heibl, C., Knauer, F., Zimmermann, F., Bufka, L. & Heurich, M. (2012). First estimation of Eurasian lynx (*Lynx lynx*) abundance and density using digital cameras and capture–recapture techniques in a German national park. *Animal Biodiversity and Conservation*, 35, 197–207. https://doi.org/10.32800/abc.2012.35.0197

Wikenros, C., Kuijper, D.P.J., Behnke, R. & Schmidt, K. (2015). Behavioural responses of ungulates to indirect cues of an ambush predator. *Behaviour*, 152 (7–8), 1019–1040. https://doi.org/10.1163/1568539X-00003266

Willerslev, E., Davison, J., Moora, M., Zobel, M., Coissac, E., Edwards, M.E., Lorenzen, E.D., Vestergård, M., Gussarova, G., Haile, J., Craine, J., Gielly, L., Boessenkool, S., Epp, L.S., Pearman, P.B., Cheddadi, R., Murray, D., Bråthen, K.A., Yoccoz, N., Binney, H., Cruaud, C., Wincker, P., Goslar, T., Alsos, I.G., Bellemain, E., Brysting, A.K., Elven, R., Sønstebø, J.H., Murton, J., Sher, A., Rasmussen, M., Rønn, R., Mourier, T., Cooper, A., Austin, J., Möller, P., Froese, D., Zazula, G., Pompanon, F., Rioux, D., Niderkorn, V., Tikhonov, A., Savvinov, G., Roberts, R.G., MacPhee, R.D.E., Gilbert, M.T.P., Kjær, K.H., Orlando, L., Brochmann, C. & Taberlet, P. (2014). Fifty thousand years of Arctic vegetation and megafaunal diet. *Nature*, 506 (7486), 47–51. https://doi.org/10.1038/nature12921

Wölfl, M., Bufka, L., Červený, J., Koubek, P., Heurich, M., Habel, H., Huber, T. & Poost, W. (2001). Distribution and status of lynx in the border region between Czech Republic, Germany and Austria. *Acta Theriologica*, 46 (2), 181–194. https://doi.org/10.1007/BF03192427

Zhang, J. & Zhang, M.J. (2013). Package ‘spaa’. *R package version*, 1 (1).

Popular science summary

In recent decades, populations of ungulates have rapidly recovered across Europe. Ungulate can significantly influence ecosystems through their feeding behaviors, impacting vegetation structure and biodiversity. Understanding the feeding patterns of ungulates is crucial for managing ecosystems, especially as their populations grow.

In this thesis, faecal samples of red deer, roe deer, and wild boar in the Bavarian Forest National Park were analyzed to determine their diets.

The result suggested that there was a high degree of overlap in the diets of red deer and roe deer, particularly at higher altitudes. Red deer showed increased diet diversity at higher altitudes, where the availability of plant species decreased. Roe deer, in contrast, did not significantly alter their diet diversity with altitude. The presence of lynx influenced roe deer feeding behavior. In areas with higher lynx activity, roe deer consumed more shrubs and broadleaved plants, likely as a strategy to remain vigilant and avoid predation. Red deer, being less preferred by lynx, did not show significant dietary changes in response to predation risk. Both deer species preferred certain plants like *Vaccinium* spp. and Rosaceae, while avoiding others such as *Avenella* spp. and *Betula* spp.. Roe deer showed a notable preference for European silver fir (*Abies alba*), which has crucial implications for the regeneration of this species in the context of forest management.

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Appendix 1

Table S1. Taxonomy of the MOTUs

MOTU	Genus_higher	Food categories
Abies	*Abies*	Conifer
Acer	*Acer*	Broadleaf
Aegopodium	Aegopodium	Forb
Agrostidinae	*Agrostidinae*	Graminoid
Ajuga	*Ajuga*	Forb
Alchemilla	*Alchemilla*	Forb
Allium	*Allium*	Forb
Alnus	*Alnus*	Broadleaf
Anthoxanthum.nipponicum	*Anthoxanthum*	Graminoid
Apioideae	Apioideae	Forb
Arabis.alpina	*Arabis*	Forb
Asteraceae	Asteraceae	Forb
Avena	*Avena*	Graminoid
Avenella.flexuosa	*Avenella*	Graminoid
Betula	*Betula*	Broadleaf
Bistorta	*Bistorta*	Forb
Brachytheciaceae	Brachytheciaceae	Other
Brassicaceae	Brassicaceae	Forb
Bryonia.dioica	*Bryonia*	Shrub
Carduinae	Carduinae	Forb
Carex	*Carex*	Forb
Carex.sylvatica	*Carex*	Forb
Castanea.sativa	*Castanea*	Broadleaf
Chamaenerion.angustifolium	*Chamaenerion*	Forb
Circaea	*Circaea*	Forb
Dryopteris	*Dryopteris*	FLH
Epilobium	*Epilobium*	Forb
Fagus.sylvatica	*Fagus*	Broadleaf
Filipendula.ulmaria	*Filipendula*	Forb
Galium	*Galium*	Forb
Geranium	*Geranium*	Forb
Geum	*Geum*	Forb
Glyceria	*Glyceria*	Graminoid
Glycine.max	*Glycine*	Graminoid
Holcus	*Holcus*	Graminoid
Hordeum	*Hordeum*	Graminoid
Hypnales	Hypnales	Other
Juncus	*Juncus*	Graminoid

Juniperus	*Juniperus*	Conifer
Luzula	*Luzula*	Graminoid
Luzula.pilosa	*Luzula*	Graminoid
Lysimachia.nemorum	*Lysimachia*	Forb
Medicago	*Medicago*	Forb
Molinia.caerulea	*Molinia*	Graminoid
Myosotis	*Myosotis*	Forb
Oleeae	Oleeae	Broadleaf
Oreopteris.limbosperma	*Oreopteris*	FLH
Oxalis.acetosella	*Oxalis*	Forb
PACMAD.clade	PACMAD.clade	Graminoid
Phegopteris.connectilis	*Phegopteris*	FLH
Phragmites.australis	*Phragmites*	Graminoid
Picea	*Picea*	Conifer
Pinaceae	Pinaceae	Conifer
Pinus	*Pinus*	Conifer
Plantago.lanceolata	*Plantago*	Forb
Poales	Poales	Graminoid
Poeae	Poeae	Graminoid
Poinae	Poinae	Graminoid
Pooideae	Pooideae	Graminoid
Populus	*Populus*	Broadleaf
Potentilla	*Potentilla*	Forb
Prunus	*Prunus*	Broadleaf
Pterigynandrum.filiforme	*Pterigynandrum*	FLH
Quercus	*Quercus*	Broadleaf
Ranunculus	*Ranunculus*	Forb
Rosales	Rosales	Forb
Rosoideae	Rosoideae	Forb
Rubieae	Rubieae	Forb
Rubus	*Rubus*	Shrub
Rumex	*Rumex*	Forb
Saliceae	Saliceae	Broadleaf
Sanguisorba.officinalis	*Sanguisorba*	Forb
Scirpus	*Scirpus*	Forb
Sciurohypnum	*Sciurohypnum*	Other
Solanoideae	Solanoideae	Forb
Sphagnum.russowii	*Sphagnum*	Other
Stellaria.alsine	*Stellaria*	Forb
Trientalis (Lysimachia)	*Trientalis*	Forb
Trifolium	*Trifolium*	Forb
Triticeae	*Triticeae*	Graminoid
Ulmus	*Ulmus*	Broadleaf
Urtica	*Urtica*	Forb

Vaccinium	*Vaccinium*	Shrub
Vaccinium.ovalifolium	*Vaccinium*	Shrub
Vaccinium.vitis.idaea	*Vaccinium*	Shrub
Vicia	*Vicia*	Forb
Viola	*Viola*	Forb
Viscum.album	*Viscum*	Shrub

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