

## Article

# Heritage Trees as an Important Sanctuary for Saproxylic Beetles in the Central European Landscape: A Case Study from Litovelské Pomoraví, Czech Republic

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**Abstract:** Intensive forest and agroforestry management has greatly reduced the biodiversity of saproxylic organisms. Large trees are one of the most important refuges of saproxylic beetles. These large trees that grow outside and inside the forest are declining in the wider landscape. Heritage trees are one of the essential groups of beneficial trees in the landscape. We investigated saproxylic beetles associated with 35 selected oak heritage trees in Litovelské Pomoraví in the eastern Czech Republic. The study aimed to investigate the distribution of saproxylic beetles on trees growing inside or at the edge of forest stands, or on free-growing heritage trees. The other studied variables were the height, DBH, and light condition (sunny or shady) of heritage trees. The results showed that sunny habitats were the only significant factor found for all saproxylic species. However, the significance of increasing tree trunk dimension was found for the endangered species. Diversity indices  $q = 0$  (species richness) and  $q = 1$  (exponential of Shannon entropy index) were also higher for sunny trees, while solitary trees showed a high Shannon index value despite the low number of samples. Redundancy analysis of saproxylic species showed that the preferred habitats of most species were sunny massive solitary oaks. The results indicated that strictly protected heritage trees scattered in the landscape are crucial sanctuaries for many species—especially in landscapes where there are not enough suitable habitats for saproxylic beetles. Finding, conserving, and protecting these rare types of massive trees in the landscape has a significant impact on the conservation of saproxylic beetle biodiversity.

**Keywords:** biodiversity; coleoptera; large tree; open canopy; beetle



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## 1. Introduction

Large, aged trees are in decline throughout the landscape [1,2]. The main reason for this is due to the intensive human use of the landscape. Massive trees are valuable from a biological, cultural and aesthetic point of view [3]. One type of massive trees in the landscape is heritage trees, which grow in historically significant places, at road junctions, or have served as landmarks in the landscape [4,5]. These are usually solitary trees of massive dimensions that have reached a very old age and carry large numbers of microhabitats [6,7], much more numerous than trees that grow in a denser spacing [8]. Therefore, they are one of the important factors in the biodiversity of saproxylic beetles [1,9,10]. Saproxylic beetles are the most studied group of forest organisms [11], and at the same time, they are one of the most threatened groups [12,13]. Saproxylic beetles depend on deadwood of all types and sizes and also on other organisms living on deadwood, e.g., mycetophages on tree fungi [14], and also microhabitats, e.g., tree cavities are very important [15,16]. Saproxylic beetles are an essential component in the decomposition of deadwood [17].

Oak (*Quercus* spp.) is the most frequently occurring of solitary tree species [18]. From this point of view, heritage trees are most often oaks. The most massive and oldest heritage

trees in Europe are mainly oaks [5]. Oak is considered the most important tree species for saproxylic beetles [19–21]. Massive oak trees, especially free-growing ones, are known to be the only hosts of a number of large or rare species [22,23]. In general, the diversity of saproxylic beetles increases with the amount of deadwood [24]. Standing deadwood hosts more beetles than lying deadwood [25,26], as well as lichens, birds, and bats [27,28]. In general, massive trees are preserved mainly in old game hunting areas, parks, pond dams, and orchards [29], or in agroforestry areas [30], and as prominent features in the landscape, such as heritage trees [5].

These types of landscape management, along with large heritage trees, excel in their natural and cultural value [31], and are often considered the best features for the life and conservation of saproxylic beetles [32]. In recent years, locations with scattered old trees have seen a significant increase in scientific interest [18,30,31,33–37]. Traditional management of scattered massive trees has been abandoned, which naturally leads to a gradual shading of the grounds and a subsequent loss of biodiversity [38,39]. The newly formed environment then fails to meet the demands of sunny-habitat species, which leads to the homogenization of invertebrate communities [40], and, at the same time, brings about the faster death rate of large trees [10] and the failure of other ecosystem functions [41]. Old trees are threatened by felling and by the absence of active support for the future generation of solitary trees [12,13,42,43].

When massive solitary trees are declared heritage trees, law protection in the Czech Republic comes into play. The protection of these heritage trees also has secondary effects from a biodiversity viewpoint. For these reasons, this paper deals with the importance of large heritage trees growing independently in the landscape in connection with beetle biodiversity. Our selected heritage trees are under the management of the conservation authority Nature Conservation Agency of the Czech Republic (Agentura ochrany přírody a krajiny České republiky, AOPK). The trees are listed in the detailed database of the AOPK. The aim of this paper was to determine whether the diameter and height of a heritage tree are important elements for saproxylic beetle diversity. We also focused on the consequences of large shade trees and whether heritage trees growing in open countryside host higher numbers of species than trees growing in a forest or at a forest edge. The group we studied included beetles, especially the group of bioindicator saproxylic beetles and endangered species.

## 2. Materials and Methods

### 2.1. Study Area

The study area lies in the north of Central Moravia, in the Czech Republic. The model area is called Litovelské Pomoraví (49.697° N, 17.135° E). The site is a protected area of 96 km<sup>2</sup> established in 1990 to protect the naturally meandering course of the Morava River and the surrounding oak floodplain forests. There are also thermophilous oak forests and oak-hornbeam and beech forests. A natural species composition prevails in the forests. The most frequently occurring soil types are luvisols and fluvisols. The location belongs to the warm climate zone T2. The average annual air temperature is 8.4 °C, and the average annual precipitation is 586 mm. Biogeographically, Litovelské Pomoraví can be classified in the Hercynian sub-province part of the province of Central European deciduous forests.

### 2.2. Studied Groups

Beetles were identified at the lowest taxonomic level (species) in the following families according to [44]. Staphylinidae were the only family not identified at the species level due to the lack of available experts. Individual species were included in the group of saproxylic beetles by [42,45,46]. We organized all saproxylic species according to their functional groups during larval development into the guilds. Types of inhabited microhabitats were classified into three cohorts: (1) wood-bark, (2) wood decay fungus, (3) cavities. The adult saproxylic beetles were separated into flower-visiting species and non-flower-visiting species; see [42]. The species taxonomy corresponded to Zicha O. (ed.) (2022) BioLib.

<http://www.biolib.cz> (accessed on 1 January 2022). The species were classified according to the Red List of Endangered Species of Invertebrates of the Czech Republic [47].

### 2.3. Method of Collection

Standard window flight interception traps (Figure 1) hung on the trunks of heritage trees (Figures 2 and 3) were used for the collection. Currently, these are the most frequently used traps for monitoring saproxylic beetles [48] and for entomological studies, cf. [49–54]. The traps consisted of a roof, plexiglass barrier, funnel, and collection container (Figure 1). When assembled, the passive barrier traps were 95–105 cm tall. A total of 35 traps were set (one trap per tree, south direct). The trapping devices were installed on 15 April 2010. Acetic acid 8% was poured into the containers, serving as a preservative, with a drop of surfactant to disturb the surface tension used. The traps were emptied six times during the season. The contents of the traps were extracted on 7 May, 22 May, 22 June, 18 July, 7 August, and 4 September 2010, when the traps were dismantled. The traps were extracted at 3 to 4-week intervals.

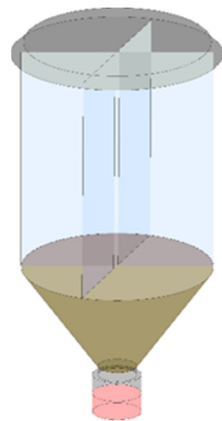


Figure 1. Window flight interception traps.



Figure 2. Forest-interior growing heritage oak tree.



**Figure 3.** Free-growing heritage oak tree.

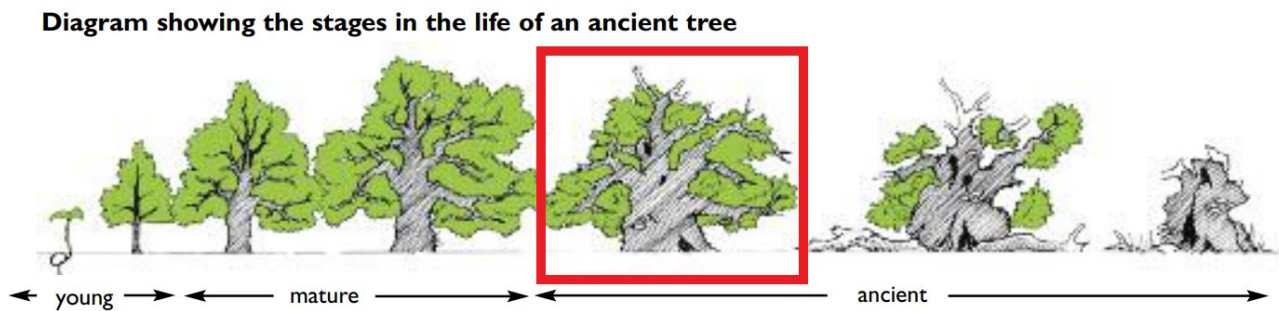
#### 2.4. Study Variables

Heritage trees are one group of large and important trees in the landscape (Figure 4). All of the examined variables were obtained from 35 oak heritage trees in the growth phase (Figure 5). The species of saproxylic beetles and Red List species were selected as dependent variables. A total of four environmental (independent) variables were selected. These included two continuous variables: the diameter of breast height (DBH) of the trunk of the heritage tree (in cm), measured at 1.3 m above the ground (min. = 96, mean = 131, max. = 220), and the height of the tree values obtained from the heritage tree database (min. = 17, mean = 28, max. = 33) measured in 2010. The two categorical variables were: degree of solar exposure (light condition) of the tree (trap): sunny (n = 16) and shady (n = 19). We divided the sites (locations) into three groups: (1) Trees growing inside a closed forest, (2) trees growing on the edge of the forest or in a forest clearing, and (3) trees completely outside the forest. The degree of sun exposure was evaluated by the location of the tree, with sunny trees either growing as a solitaire, near the forest edge, or on it. The shaded trees on which the traps were placed grew in a closed forest.



**Figure 4.** Significant trees according to Ancient Tree Forum [4].





**Figure 5.** The course of tree growth according to Ancient Tree Forum [4] with the selected studied phase of the growth framed.

### 2.5. Statistical Analysis

We used the stepwise generalized linear model  $\chi^2$  test (log link function, Poisson distribution) according to the lowest Akaike's information criterium (AIC) to find important variables, where comparison of individual environmental variables with the total variation was explained by the model in which the variable was omitted. The model was used for two dependent variables—species richness of all saproxylic beetles and endangered species in each sample (number of species per trap over the whole season) to search for important independent variables. The significance of the variables was tested at  $p < 0.05$ . Statistical analyses were performed in the Statistica 13 software (StatSoft, Inc., Tulsa, OK, USA).

A group of categorical variables was selected for curves of diversity indices. Rarefaction, an extrapolation approach that estimates the rate of increase in the number of species per number of the samples, was used. We used the iNext program [55] to analyze the species richness and diversity curves. Sample-based rarefaction was used to generate the species richness curve, where the estimated richness profile (200 bootstraps, incidence data) was calculated. Diversity indices were based on [56], with Hill numbers:  $q = 0$  (species richness) and  $q = 1$  (exponential of Shannon entropy index) used. Hill numbers offer some distinct advantages over other diversity indices [56]. The method of non-overlapping confidence intervals was used for the significance of these curves [57].

Ordination diagrams were plotted, showing the preferences of recorded species to variables of heritage trees. Constrained linear redundancy analysis (RDA) with log transformation was used. Two approaches in the form of RDA were used—species individual preferences and richness (number of species) in samples summarized to independent variables, similar to [58]. A test of all canonical axes was computed for all species. Species individual preference diagrams were plotted for the 40 best-fitted species. We summarized the compositional community of sunny and shady variables in heritage trees using principal coordinates analysis (PCoA). All ordination analysis was computed with unrestricted 4999 Monte Carlo permutations. Calculations were performed in the CANOCO 5 program for multivariate statistical analysis using ordination methods [59,60].

A similar analysis of inhabited habitats was performed by Weiss et al. [61]. Non-parametric ANOVA Kruskal–Wallis was used to detect differences between the studied microhabitat groups. The normality of data was checked by the Shapiro–Wilk test. The Dunnett post-hoc test was used to compare differences between groups. Log  $\chi^2$  linear model tests (Poisson distribution) were fitted as the model for all independent factors. Preference microhabitat guilds were verified by multivariate space using redundancy analysis (RDA) (as in the previous RDA analysis) to find the important variables for the richness of microhabitat guilds as the dependent variable.

## 3. Results

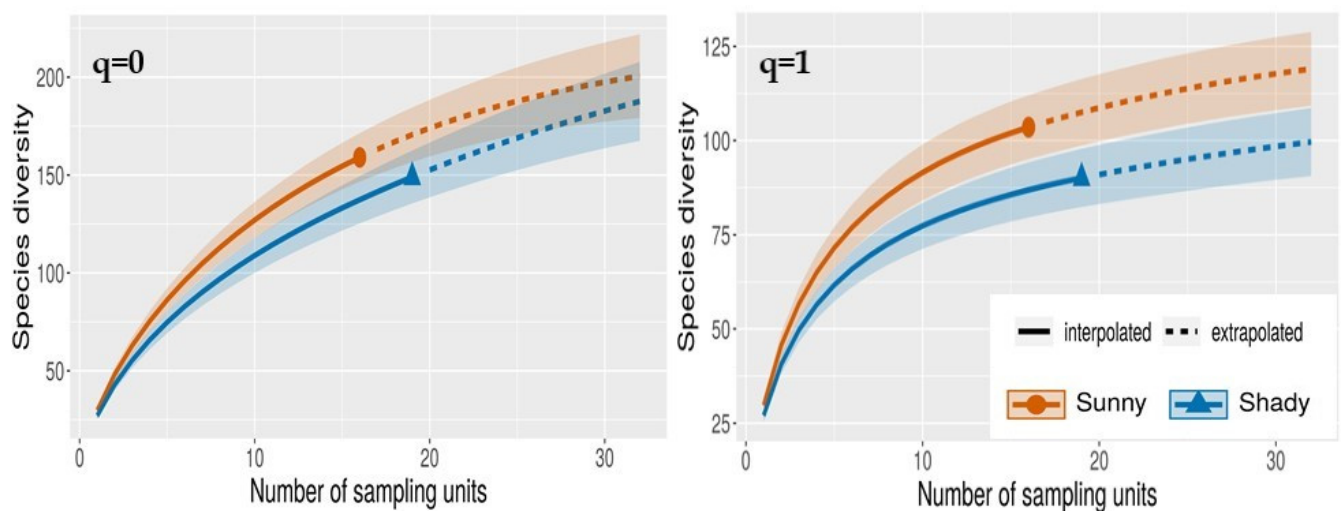
The dataset of saproxylic beetles included 206 species and 4493 saproxylic individuals (Supplementary Material). Thirty-nine species were Red List species. The most abundant species was *Cryptarcha strigata* (Fabricius, 1787), with 1103 individuals, which was almost 25% of the recorded saproxylic beetles.

### 3.1. Species Richness

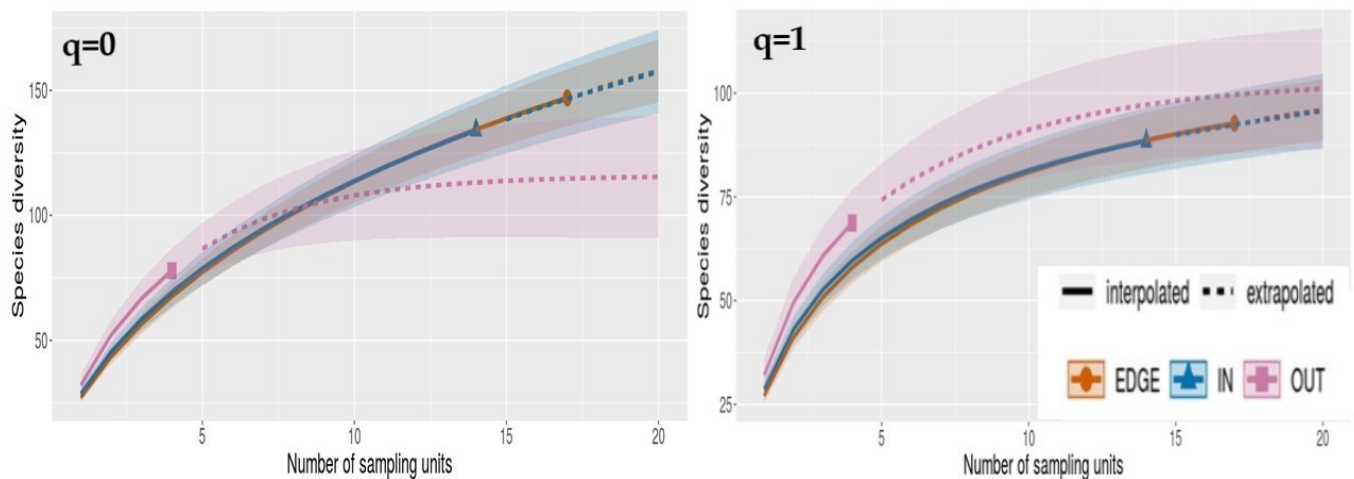
The highest species richness of saproxylic beetles was on sunny trees, and the diameter and locality of the heritage trees were near to the significance border (Table 1). Species curves indicated higher species richness on sunny trees ( $q = 0$ ) with a borderline significance. In contrast, the Shannon diversity index ( $q = 1$ ) showed statistical significance for sunny trees (Figure 6). In terms of management, the curves showed that the solitary tree had a higher onset of species accumulation, and the Shannon index outweighed the other sites despite the extrapolated values (no significance; Figure 7). The visualization of saproxylic species preferences showed a univariate tendency, where the beetles found suitable living conditions—a sunny massive solitary oak tree (RDA analysis; Figure 8). In contrast, endangered species were statistically significantly associated with tree diameter (Table 1). Similarity/dissimilarity of shaded and sunny trees using PCoA showed trees aggregated by insolation had different beetle communities (Figure 9).

**Table 1.** Species richness of saproxylic beetles in the study location. Stepwise (the lowest AIC) generalized linear model (Poisson distribution) was used. Results of whole model are with the name of dependent variables and estimate of variables in model are shown below. Value (+) corresponded to positive result of variable. Omitted variables during the process of stepwise model are marked (–). Statistically significant effects are marked in bold.

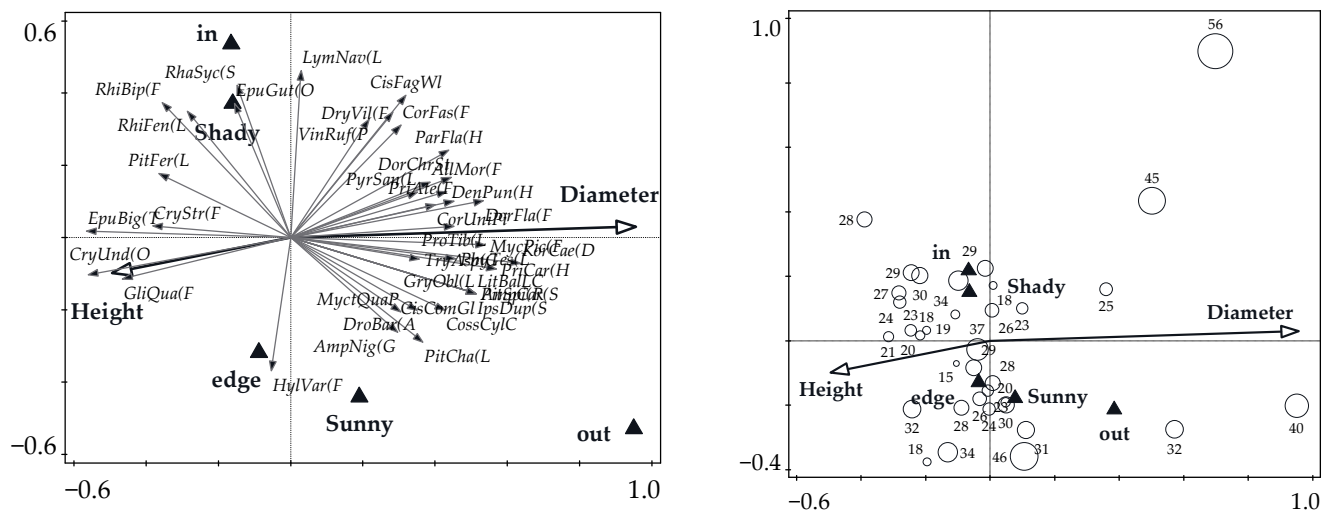
All Saproxylic Species (AIC 265.56; $\chi^2$ 10.25; $p = 0.036$ )				Endangered Species (AIC 172.00; $\chi^2$ 5.20; $p = 0.023$ )		
Variables	Value	$\chi^2$	$p$	Value	$\chi^2$	$p$
Height		–	–		–	–
Diameter		2.04	0.153	+	<b>5.20</b>	<b>0.023</b>
Locality		5.61	0.060		–	–
Light condition (sunny)	+	<b>5.62</b>	<b>0.018</b>		–	–



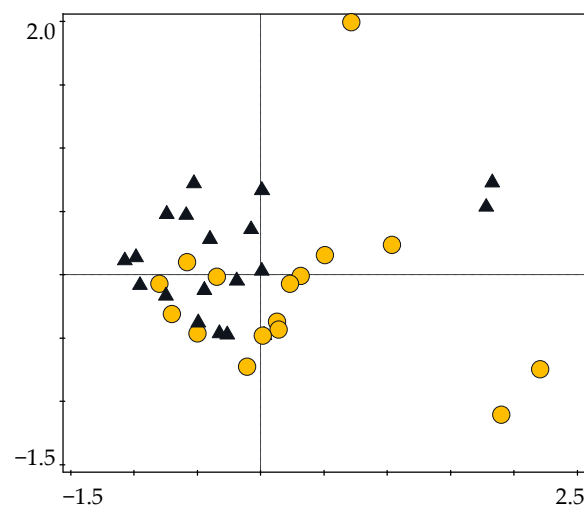
**Figure 6.** Sample-size-based rarefaction and extrapolation sampling curves within light condition sites showing Hill numbers  $q = 0$  (species richness) and  $q = 1$  (exponential of Shannon entropy index). Color-shaded areas are 95% confidence intervals. Solid symbols represent the total number of study samples.



**Figure 7.** Sample-size-based rarefaction and extrapolation sampling curves within growing location of tree sites showing Hill numbers  $q = 0$  (species richness) and  $q = 1$  (exponential of Shannon entropy index). Color-shaded areas are 95% confidence intervals. Solid symbols represent the total number of study samples.



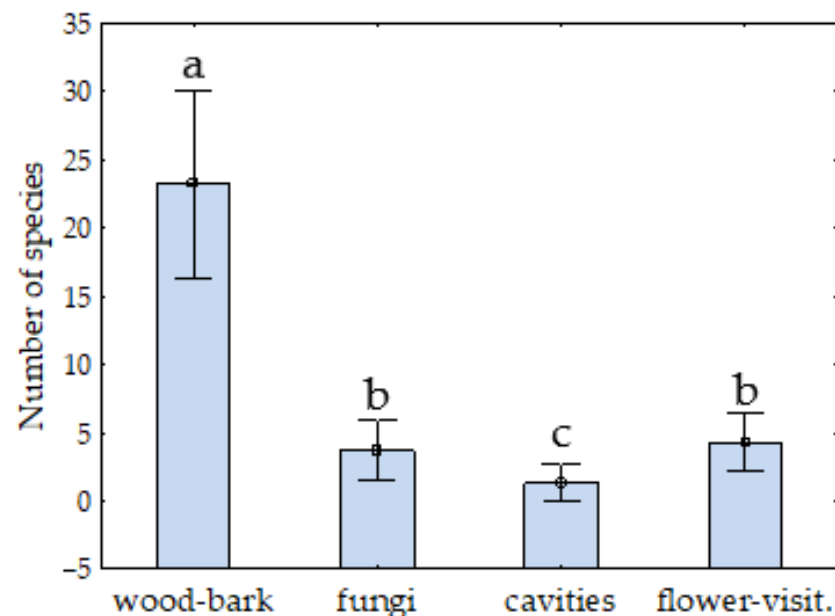
**Figure 8.** Both diagrams: relationship between species preferences and species richness by samples (the size of each circle indicates the species richness of the samples—absolute number of species in the trap) for heritage tree variables by RDA ( $pF = 1.4$ ;  $p = 0.040$ ). Explained by axes; axis 1—8.85%; axis 2—4.43%. Solid symbols are categorical variables and arrows of DBH and height are continuous variables.



**Figure 9.** PCoA summarized the beetle species similarity/dissimilarity by independent factor. Yellow circle—sunny trees and black triangle—shady trees.

### 3.2. Guilds' Composition

We found that our groups showed significant differences: KW-H (3; 140) = 99.81;  $p < 0.001$  (Figure 10). The most common group according to inhabited microhabitats was the wood-bark group, while the fewest species were linked with wood-decay fungi and cavities (Figure 10). In the search for variables of heritage trees, the relationship between the sun and fungal groups, and the negative relationship between the cavity-dwelling species and tree height was found (Table 2). The cavity-dwelling species were more dependent on increasing diameter, and the other groups were more dependent on the sunny factor, according to RDA (Figure 11). A total of 33 saproxylic species were recorded (16% of total saproxylic species), who as adults collect food from flowers.

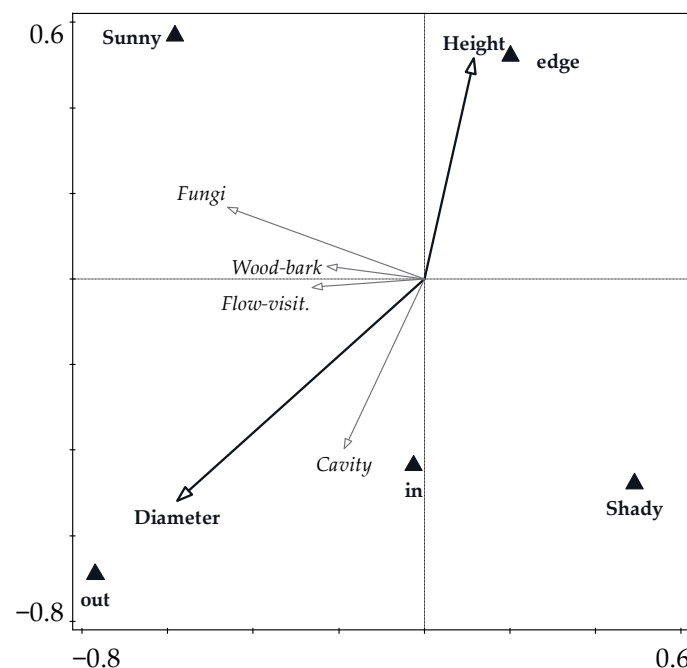


**Figure 10.** Distribution of species according to groups: wood-bark specialists; fungi specialists; cavity specialists; adult flower visitors. Error bars are standard deviation and solid point is mean (mean  $\pm$  SD). Letters above error bars indicate differences by Dunnett post-hoc test (MS = 14.679, df = 136.00).



**Table 2.** Species guild richness of saproxylic beetles in the study location (log  $\chi^2$  linear model, Poisson distribution). Value (+) corresponded to positive response of variable and (−) negative response. Statistically significant effects are marked in bold.

	Wood-Bark			Fungi			Cavity			Flower-Visitor		
	Value	$\chi^2$	<i>p</i> -Value	Value	$\chi^2$	<i>p</i> -Value	Value	$\chi^2$	<i>p</i> -Value	Value	$\chi^2$	<i>p</i> -Value
Diameter		0.77	0.380		1.21	0.271		1.34	0.246		1.01	0.315
Height		0.26	0.608		0.05	0.830	—	<b>5.16</b>	<b>0.023</b>		0.23	0.634
Location		3.06	0.217		3.23	0.198		3.24	0.199		1.93	0.381
Light condition (sunny)		2.61	0.106	+	<b>6.72</b>	<b>0.009</b>		0.00	0.997		1.35	0.244



**Figure 11.** Microhabitat guilds and their preferences for variables in heritage trees by RDA ( $pF = 1.2$ ;  $p = 0.282$ ). Explained by axes; axis 1—10.34%; axis 2—6.25%. Solid symbols are categorical variables and arrows of DBH and height are continuous variables.

#### 4. Discussion

Our study shows that massive heritage trees are indispensable elements in the landscape for saproxylic beetles. This result is consistent with a number of other studies [38,62]. Although free-growing trees were minimally represented in our study, they nevertheless showed the best values. We can agree with [58] that saproxylic beetles prefer free-growing large trees. These are equally important for other insects, plants, mosses, and also vertebrates, such as birds and bats [9,28]. Window flight interception traps are designed primarily for catching more active and flying species [63], and window traps are useful for comparing forest habitats [64]. Nevertheless, solitary trees that have no deadwood nearby reach the highest species richness of saproxylic beetles [9,58,63]. Further, based on the unattained asymptote of species richness (curves), we conclude that the species richness associated with heritage trees will be higher than that observed. Even so, we assume that the methodology implemented proved correct, as a large number of species with hidden life cycles were also captured, even though the trap allegedly failed to attract only the desired insect species. The life activity of these species is less dispersive, and they rarely leave their cavities [65,66]. For this reason, it is highly likely that these captured species are tied to the heritage tree itself, and the environmental variables identified are undoubtedly

critical. Next, we address how—and to what extent—individual variables affect the life of saproxylic beetle species.

#### 4.1. Species of Heritage Trees

The studied tree species are the most common deciduous trees in temperate forests, so these findings are applicable throughout vast regions of Europe. Oak (*Quercus* spp.) was the only tree species represented in our study; similarly, in many areas of Europe, it is the most frequent solitary and heritage tree species [5,18]. Oak is considered to be the most valuable tree species for saproxylic beetles [67,68]. Oak, as a long-lived tree species and a heritage tree, can often live over 1000 years [5] and thus can form trunks twice as thick as beech, for example [18]. Similarly, for non-saproxylic leaf-eater beetle species, oak is the most bountiful tree species [69], in comparison with the second most common solitary tree species, beech [18]. Beech trees begin to die at the age of 200 [70], and beech wood decomposes twice as fast as oak [71]. Some studies have found higher numbers of saproxylic species on beech than on oak [72]. From the point of view of the spatiotemporal endurance of optimal microhabitats, beech is less suitable than the enduring oak. However, only oak was included in our study, so we cannot confirm these aspects.

#### 4.2. Dimensions of Heritage Trees

As tree diameter increased, we observed a tendency for an increasing number of all saproxylic species. Meanwhile, endangered species were associated with statistical significance only with tree diameter. This is consistent with the findings of [25], who confirmed that bulky standing oaks are more valuable than thinner trees or even lying trunks. Some studies have also confirmed that the number of species increases with the dimensions of deadwood [73,74], and large specimens also host large species of beetles [75]. As the trunk diameter increases, the tree can offer a more diverse mosaic of microhabitats, such as large cavities or various stages of wood decomposition. This is mainly due to the different properties of massive heritage trees compared to trees within a closed canopy in managed stands, which are grown to produce quality raw material for the wood processing industry. Heritage and free-growing trees typically form massive dimensions [76], and consequently, large, often low-lying branches [77] and other microhabitats [7,78]. There is a causal connection between age and the diameter of the trunk. It is an asymptotic approximation to the maximum on the threshold of the tree species senescence. The natural dying of trees is important for species that need the gradual decay of the entire tree where habitat niches develop. Typically, recently dead trunks and branches host the families Buprestidae and Cerambycidae, medium-decayed deadwood hosts, e.g., the family Elateridae, and the advanced phase of wood decomposition—the rotted wood inside the cavities—hosts representatives of the families Scarabaeidae (typically *Lucanus*, *Gnorimus*, and *Protaetia* genera). Excessive safety or nature protection concerns (e.g., where dead heritage trees are non-native species) should not be the reason to destroy these moribund yet beneficial standing heritage trees wherever possible. It must be remembered that each species of saproxylic beetles has specific demands in relation to the dimensions of deadwood. The whole heritage tree offers numerous optimal dimensions, from branches to strong trunks affected by rot. One of the most essential microhabitats of massive trees is cavities, especially for specialist and Red List species [15,79,80]. Generally speaking, the rarest saproxylic species are bound to thick decaying trunks, typical representatives being old-growth relics listed by [81] for Central Europe. For example, we can mention: >60 cm for *Rhysodes sulcatus* (Fabricius, 1787) [82]; >100 cm for *Limoniscus violaceus* (P.W.J. Müller, 1821) [23]; >60 cm for *Cerambyx cerdo* (Linnaeus, 1758) [83].

#### 4.3. Sun Exposure

The effect of the sun exposure of a given heritage tree was found to be the most important predictor of the richness of saproxylic beetles. Beetles are ectothermic animals, and their temperature depends on the ambient temperature. The high biological value of

massive trees is strongly correlated with an open canopy and sufficient insolation, and has been confirmed by several studies [9,10,40,80,84]. At a higher temperature, they are more spatially active, and their ongoing development is accelerated. The importance of macroclimatic conditions for saproxylic beetles is in accordance with the findings of, e.g., [50,51,85,86]. Sun exposure also has a positive effect on epigeal beetle species [87,88]. Oaks (*Quercus* spp.) grow mainly in the warmest parts of Europe (lowlands); therefore, all species of biota have adapted to the same conditions. For this reason, oak saproxylic beetle species respond sensitively and positively to increases in temperature [89] and are therefore strongly affected by an open canopy, as detected in our study, which complies with numerous studies on oak stands and oak trees [19,40,62,80]. This confirms that the phylogenetic development of a tree affects the species associated with it [82,90].

#### 4.4. The Importance of Continuity

The loss of continuity is a significant problem for many animals. In the modern landscape, the continuity of habitats has all but disappeared. Heritage trees can be integral stones in the mosaic of active biodiversity promotion, together with, e.g., active deadwood enrichment [91]. A key bonus of heritage trees is that they can withstand many centuries and maintain suitable microhabitats on each site—even for about 1000 years, in the case of oaks [5]. When forest stands are enriched with deadwood, maintaining continuity is slightly more difficult in terms of wood decomposition [26]. Several authors have studied and compared areas of permanent and non-permanent forest environments, and they also confirm the importance of continuity for saproxylic beetle species [92–94], or at least demonstrate its positive additive effect [95].

### 5. Conclusions for Management

Our study has shown that heritage trees are significant in terms of cultural, heritage, and other notable reasons. They are also a crucial element in the landscape for the biodiversity of a model group of saproxylic beetles. In our study, we observed that sunny, large, solitary heritage trees show high values of species richness. This observed aspect is important for the management of this group of heritage trees. Often these trees are overgrown with dense vegetation, and sunlight is blocked. In this case, the surroundings of heritage trees must be cleared and released from the overgrowing vegetation, and all underplants need to be removed, as documented for example by [38,96]. In some areas, massive removal of unwanted undergrowth is carried out [32]. Another important factor was found to be the size of the tree for endangered species. It is documented that as trunk size increases, microhabitats become more frequent, especially valuable cavities [15,16]. In this case, it is important to monitor suitable trees that could be future heritage trees and provide them with appropriate protection. Perhaps a more important negative factor for many species, and not only for invertebrates, is the “active” protection of heritage trees. This activity consists of pouring concrete into cavities, sailing cavities, and coating cavities—as is often occurring [97,98]. This activity has immeasurable consequences for biodiversity, as species tied to these rare microhabitats will disappear; this should therefore be abandoned as far as possible. Although it was not the aim of the study, it is clear that the concreting of the cavity amounts to a complete destruction of the microhabitat. We recommend actively seeking out and protecting certain trees, without the “active management” of heritage trees, wherever possible, especially a particular group of large trees, regardless of the fact that they do not meet the parameters of a heritage tree, because their importance in the landscape is underestimated [99], and they can also be beneficial in terms of biological pest control [100].

**Supplementary Materials:** The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/f13071128/s1>. Supplementary Material (Caption: Result).

**Author Contributions:** Conceptualization, O.N. and V.Z.; Methodology, O.N. and V.Z.; Field work, O.N.; Investigation, V.Z. and O.N.; Analyses: V.Z.; Writing—original draft preparation, V.Z., O.N. and J.R.; Writing—review and editing, V.Z., O.N., J.R., M.M. and V.P. All authors have read and agreed to the published version of the manuscript.

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**Conflicts of Interest:** The authors declare no conflict of interest.

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