Relation between plant species diversity and landscape variables in Central-European dry grassland fragments and their successional derivates

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Abstract – A systematic field survey of an area of 843 ha in the traditional Central-European agricultural landscape of Goričko Nature Park in Slovenia revealed 80 fragments of dry semi-natural grasslands. Vascular plant species diversity was studied in relation to landscape variables and to threat (Slovenian red-listed species). Our results show that fragment size does not affect plant species diversity. In addition, fragment shape index is not related to Alpha diversity. Higher Alpha diversity was observed for abandoned grassland fragments. The lowest Alpha diversity was perceived on more mesic fragments, where habitat specialists are much scarcer. It was confirmed that the highest diversity of specialists are in the driest fragments, both still mowed and abandoned. With an increase in the number of distinctly different bordering habitat types, the total number of species per fragment generally does not increase, except in the case of those fragments that are already in different succession stages. Abandoned and typical dry grasslands are associated with a higher number of bordering habitats. Typical dry grassland fragments and abandoned ones, which probably derived mostly from drier (less productive) grasslands, are found on lower altitude and have a lower shape index. Habitat specialists Sedum sexangulare, Polygala vulgaris and Spiranthes spiralis have higher frequency in fragments with a lower shape index. This means that these oligotrophic specialists occur in smaller fragments. But Orchis morio has higher frequencies of occurrence on polygons with a higher shape index, which confirms the observation that this species occurs in larger and more irregular fragments, as well as close to houses and fields and along the roads.

Key words: Goričko Nature Park, habitat specialists, NE Slovenia, semi-natural dry grasslands, traditional agricultural landscape

Introduction

Over the past century, grasslands and other semi-natural plant communities in Southern and temperate Europe have suffered a dramatic decline in the area they cover, owing to land use changes (Luoto et al. 2003). However, the primary driver of the current decrease in species densities (and their populations) and species loss is habitat fragmentation (Fattorini and Borges 2012). Habitat fragmentation not only causes habitat loss or reduction, but also promotes habitat isolation, diminishes habitat quality, and increases edge effects (Lomolino et al. 2010). The alteration of landscapes throughout the world has resulted in fragmentation of natural and semi-natural vegetation, imposing a great threat on

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numerous plant and animal species (Kiviniemi and Eriksson 2002). Many researchers have concluded that habitat fragmentation poses an important additional threat to biodiversity (Hanski 2005, Janišova et al. 2014, Zulka et al. 2014), but others claim that fragmentation *per se* is of secondary importance (Fahrig 2003). One reason why the effect of connectivity (or fragmentation itself) might not be significant is the slow response of populations to environmental change (Hanski 2005). One consequence of the intensification of agriculture in Europe over the last half-century has been the loss and fragmentation of traditionally managed habitats, such as semi-natural grasslands (Kiviniemi and Eriksson 2002, Cousins 2005). In fragmented landscapes, plant species persistence depends on functional

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connectivity in terms of pollen flow to maintain genetic diversity within populations and seed dispersal to re-colonize habitat patches following local extinction (Rico et al. 2011). Habitat fragmentation is predicted to lead to area-related reductions in population size and decreasing colonization rates as a result of isolation (Kiviniemi 2008).

According to island biogeography theory, the species richness of patches is determined by their size and spatial isolation, while in conservation practice, it is patch quality that determines protection and guides management (Zulka et al. 2014). Dry grassland species-area relationships can be explained by two ecological hypotheses: the first predicts higher species richness due to the higher habitat heterogeneity and the second claims that colonisation (extinction) dynamics causes increasing species numbers with increasing habitat area independent of habitat heterogeneity (Rosenzweig 1995, Krauss et al. 2004). The area effect per se cannot be easily disentangled from the habitat diversity effect and so it is difficult to quantify the interrelationship (Steinmann et al. 2011). Previous studies have already reported a positive correlation between habitat heterogeneity and species diversity (Rosenzweig 1995, Ricklefs and Lovette 1999). There are only a few studies dealing with habitat specialists in general while there are several studies of the frequency of particular grassland species in relation to fragment area, shape index and to habitat heterogeneity (Paušič and Kaligarič 2015).

The landscape structure in Goričko, i.e., the spatial distribution of ecotopes, is regarded as a mosaic of "frozen processes". In other words, landscape structure is assumed to mirror the processes that have been going on in a landscape (Forman 1995). Small-scale patchiness and its diversity reveal the "typical" traditional agricultural landscape, which was fragmented for socio-economic reasons at the end of the 19th and at the beginning of the 20th century. Goričko Nature Park was established in 2003 in order to protect this well-preserved, traditional Central-European agricultural landscape. The mosaic structure of landscape and the variety of secondary habitats in Goričko were caused by the simultaneous effect of natural and anthropogenic-geopolitical, economic and social factors (Kaligarič et al. 2008). In the traditional agricultural landscape of Goričko, there were basically four major land use (management) categories: arable fields, grasslands (including orchard grasslands), vineyards and gardens. During the past century, traditional land use changed drastically in Central Europe, and gradually also within the study area inside Goričko Nature Park (Škornik 2003). The area used for grasslands decreased mainly because of land abandonment and forest progression (Cousins et al. 2014).

The aim of this study was to investigate the relation between plant species diversity, fragment area and fragment shape index. The following methodological steps were undertaken: (1) analyse the differences in floristic composition of dry grassland fragments using TWINSPAN software, (2) compare the plant composition/frequency in the fragments with selected landscape variables, (3) study the differences between TWINSPAN groups and all three Shannon-Weaver diversity indices (Alpha diversity, Alpha diversity of habitat specialists and Alpha diversity of habitat generalists), (4) test the relations between the landscape variables and frequency of single habitat specialists, and (5) find differences between threatened (red-listed) plant species and TWINSPAN groups.

Material and methods

Study area and field methods

The study area lies in NE Slovenia within the borders of Goričko Nature Park (Fig.1), at approximately 46°50'– 46°52' N, 16°15'–16°52' E and covers 843 ha. The area has a Central-European climate with a relatively dry winter. Average annual rainfall is between 750 and 820 mm (ARSO 2014). The driest months are January and February, while the most rainfall occurs in July and August (ARSO 2014). Mean annual temperature is between 9 and 10 °C (ARSO 2014). Dry and semi-dry grasslands in Goričko are not as rich in species as similar grasslands elsewhere in Slovenia, owing to the low pH value of the soil (Škornik 2003).

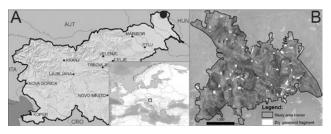


Fig. 1. Map of Slovenia with bordering countries (A) and study area (B) with 80 dry grassland fragments.

We analyzed 800 plots on 80 dry grasslands fragments (including some earlier or middle successional stages after abandonment) systematically mapped in the study area. They mostly belong to the association Hypochoerido-Festucetum rupicolae or drier stands of the association Ranunculo bulbosi-Arrhenatheretum elatioris. All dry grassland fragments (80) were sampled and analysed on randomly dispersed square meter plots (10 per each fragment). Species composition was recorded on 800 plots, where vascular plants were sampled using presence/absence data. For each plant species we calculate frequencies; 10 plot samples on 80 fragments. The vegetation composition data were collected in only one year (2013) by the first author. Plant species occurring only in one sample were removed (we exclude 11 plant species), in order to exclude casual occurrences from the analyses. Taxonomic nomenclature followed Martinčič et al. (2007). Plant species specialists and generalists were defined according to expert knowledge, by help of the syntaxonomic classification of the taxa. Plant species characteristic for the alliances Bromion erecti (and order Brometalia erecti) and Violion caninae represent species of drier, nutrient-poor neutral to acid soils of Central European provenance and climate. The rest of the species were considered generalists, although some species from e.g. ruderal or forest habitats are also considered to be in this group.

Species diversity

Shannon-Weaver diversity index (H) was calculated for all species (called Alpha diversity), and further indices were calculated only for species specialists (called Alpha diversity specialists) and for species generalists (called Alpha diversity generalists) (Shannon and Weaver 1963).

We also identified threatened plant species using the Red Lists of the threatened flora of Slovenia (Anonymous 2002). Of the 169 plant species included in the study, 7 were included in the Red List and from this we calculated threatened species frequency (Frk_red). Threatened species frequency was noted as the number of threatened plant species recorded in 10 plots on each dry grassland fragment.

Landscape variables

The spatial geometry (area and perimeter) of the grassland fragments was obtained using ArcGIS 9.3 spatial analyst tools (ESRI 2010) by digitalizing the plots – drawing polygons in vector format. The plots were first drawn on printed Orto-photo imagery in 2009 (pixel size=0.5 m) (GURS 2010). Later, the relation between fragment area and its perimeter – a straightforward "shape index", perimeter-area ratio for each grassland fragments was calculated (PARA index, referred to as "shape index" later on in the text).

All different bordering habitat types (polygons) (NofHT) for each grassland fragment were determined using an existing 2012 habitat type map owned by the national Institute for Nature Conservation (ZRSVN) which is based on the PHYSIS typology (Devillers and Devillers-Terschuren 1996), adopted and modified for Slovenian conditions (Jogan et al. 2004). The buffer analysis tool within ArcGIS 9.3 (ESRI 2010) was applied in order to quantify habitat types that share the border with the target grassland fragments. Additionally, the above-mentioned habitat type map was used to calculate the distance from the settlement/houses variable (DIS2H). In this regard, all polygons determined as buildings (houses) were selected and extracted from the initial habitat type map. The distance surfaces between all houses were calculated in the next step by using the Euclidean distance algorithm in the ArcGIS spatial analyst tools (ESRI 2010). The mean distance from the settlement/houses for each grassland fragment in the study area was determined in the end with the Zonal statistics tool in ArcGIS and was additionally log transformed for the purpose of statistical analysis. Accordingly, a digital elevation model (DEM) in a 5 m horizontal resolution (GURS 2010) served as the input data with a calculation of the mean height above sea level variable (HASL) for each grassland fragment and the application of the zonal statistics tool in ArcGIS 9.3 (ESRI 2010). All variables were z standardized for the purpose of landscape variables analysis.

Data analysis

To explain the relations between Alpha diversity of all 169 plant species and 46 habitat specialists the relations between fragment area and fragment shape index were tested separately with Pearson's correlation coefficient. The analysis was carried out in R (R Development Core Team 2009). To classify grassland fragments according to their similarity we used two-way indicator species analysis (TWINS-PAN) (Hill 1979). This analysis was run with frequencies data of 80 dry grassland fragments and 169 plant species.

Further, redundancy analysis (RDA) ordination was conducted with 80 grassland fragments, to relate variation in the whole of the species data set (169) to landscape variables and threatened species richness. TWINSPAN groups were added as supplementary variable without any effect on the analysis. Ordination method (RDA) and visualization of their results were carried out using the Canoco and CanoDraw programs (ter Braak and Šmilauer 2002).

Additionally, a comparison was performed between the TWINSPAN groups and all three Shannon-Weaver diversity indexes (Alpha diversity, Alpha diversity specialist and Alpha diversity generalist) using one-way ANOVA. One-way ANOVA was also performed for TWINSPAN groups and landscape variables. The analysis was carried out in R (R Development Core Team 2009).

We used Poisson or QuasiPoisson regression (depending on the data dispersal) to test the effect of landscape variables (HASL, DIS2H, NofHT and shape index) on specialist species. We implemented Poisson or QuasiPoisson regression within a generalized linear modelling (GLM) framework, using a Poisson link or QuasiPoisson link function. We tested only specialist species with frequencies higher than ten. The analysis was carried out in R (R Development Core Team 2009).

In order to present differences between threatened plant species frequency from the Red List of Slovenia we calculated sum of 10 plots on each dry grassland fragment. In addition we performed the Kruskal- Wallis test to find possible differences between threatened species frequency (Frk_red) and determined TWINSPAN groups.

Results

From 80 dry grassland fragments, we included in the dataset 169 vascular plant species (On-line Suppl. Tab. 1) with an average of 43 plant species per fragment. The total fragment area covers 25.56 ha. The area of the smallest fragment is 0.028 ha and the largest is 1.12 ha. From 169 vascular plant species, 46 were considered habitat specialists with an average of 13 plant specialists per fragment, mostly characteristic species of *Mesobromion* and *Violion* alliances. The average number of plant generalists per dry grassland fragment is 31 plant species.

As the first step, the relations between the Alpha diversity of all 169 plant species and 46 habitat specialists between fragment area and fragment shape index were tested separately. However, no significant relations between all mentioned parameters were found (results not shown).

At the second step, differences in floristic composition were analysed with TWINSPAN classification of matrix with 169 plant species and 80 dry grassland fragments (Fig. 2). Four groups were distinguished on the basic of this analysis. The first TWINSPAN group represents the most typical extensively used oligotrophic and mostly dry grasslands fragments (referred to as DRY), regularly mowed, with typ-

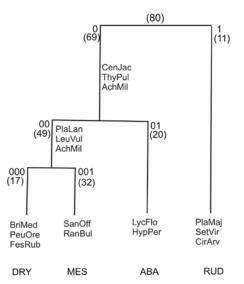


Fig. 2. Simplified TWINSPAN classification tree (dendrogram) of matrix with 169 plant species and 80 dry grassland fragments. For each division number of groups, number of dry grassland fragment (in brackets) in group and indicator plant species are shown. Grassland fragments were divided into four groups: DRY – extensively used oligotrophic and mostly dry grasslands fragments, MES – more mesic grassland fragments, ABA – abandoned grassland fragments mostly overgrown with young woody perennials, RUD – ruderalised grassland fragments. Abbreviation of plant species are explained in On-line Suppl. Tab. 1.

ical and abundant species such as Briza media, Peucedanum oreoselinum and Festuca rupicola. The second TWIN-SPAN group represents also extensive and still dry, but relatively more mesic, grassland fragments (referred as MES) with typical and abundant species as Sanguisorba officinalis, Ranunculus bulbosus and Centaurea jacea. The third TWINSPAN group with the most typical and abundant species Quercus petraea, Prunus domestica (remnant of orchards), Tanacetum vulgare, Lychnis flos-cuculi, Veronica camaedrys and Hypericum peforatum represents abandoned grassland fragments (referred as ABA), mostly already overgrown with young woody perennials. The fourth TWINSPAN group is constituted by grassland fragments, which are ruderalised (disturbed) fragments (referred as RUD), also containing weeds or ruderals as typical and abundant species: Plantago major, Cirsium arvense, Setaria viridis or Erigeron annuus.

In the third step, we compared the plant composition/ frequency in the fragments with landscape variables. In the RDA ordination of two matrixes: first one with 169 plant species and 80 dry grassland fragments and second one with 4 landscape variables (NofHT, DIS2H, HASL and shape index) plus threatened species richness was performed in order to differentiate the analysed dry grasslands fragment on the basic of selected landscape variables and their endangerment (threat) (Fig. 3). The first two axes of RDA analysis explained 33.1% of variation. The first axes were related positively to the shape of the fragments (shape index), altitude (HASL) and threatened species richness (Frk_red). However, the second axes were related positively to the number of different bordering habitat types (NofHT), threatened species richness (Frk_red) and nega-

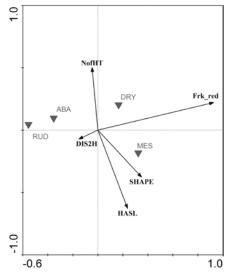


Fig. 3. Redundancy analysis (RDA) ordination diagram of two matrixes: first with 169 plant species and 80 dry grassland fragments and second matrix with 4 landscape variables, frequency of the species from the Red List and 80 dry grassland fragments. Eigenvalues: axis 1=0.158, axis 2=0.097, 25.5 % of variance in species explained by both axes. Shown species have the highest weight. Grassland fragments were divided into four groups with TWINSPAN analysis: DRY group, MES group, ABA group, RUD group; explained in Fig. 2 in detail. HASL – height above sea level; SHAPE – shape index; NofHT – number of bordering habitat types, DIS2H – distance from the settlements/houses, Frk_red – threatened species richness.

tively to shape index, distance from the settlements/houses (DIS2H) and altitude (HASL). Furthermore, the DRY and MES groups had more species from the Red List than ABA and RUD groups and the DRY and the ABA group have highest frequencies of bordering habitat types than the MES and RUD groups, in contrast to landscape variables shape index and altitude.

Additionally, as the fourth step, we were looking for the differences between TWINSPAN groups and all three Shannon-Weaver diversity indexes (Alpha diversity, Alpha diversity specialist and Alpha diversity generalist). We found significant differences for Alpha diversity (all 169 species considered) in the first three TWINSPAN groups -DRY, MES and ABA (p<0.001); only the RUD group it is not significantly different from the DRY and the MES group (Fig 4a). Very similar results were found when Alpha diversity was calculated for generalists only. We found significant differences between MES and ABA group (p< (0.001) and between the DRY and the ABA group (p<0.001). However, the Alpha diversity, calculated for generalists does not differentiate DRY from MES and ABA groups. We found also no significant differences between the ABA and RUD group, when calculating the Alpha diversity for generalists (Fig. 4b). What is more, Alpha diversity for specialists does not differentiate between the DRY and the ABA group or between the MES and RUD group, but shows significant differences between MES and RUD groups together and the ABA group (Fig. 4c). We found also significant differences in landscape variable "distance from the settlements/houses" (DIS2H), but we have not found differences

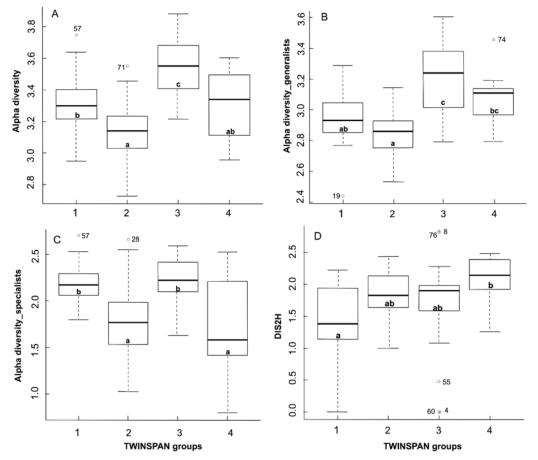


Fig. 4. Differences between TWINSPAN groups and all three Shannon-Weaver diversity indexes: Alpha diversity (A), Alpha diversity generalists (B) and Alpha diversity specialists (C) and for TWINSPAN groups and distance from the settlements/houses (DIS2H) (D). TWINSPAN groups: 1 - DRY group, 2 - MES group, 3 - ABA group, 4 - RUD group; explained in Fig. 2 in detail. Letter in box-plot are from ANOVA, Duncan post- hoc test.

in TWINSPAN groups between any other landscape variables (Fig 4d). Significant differences in log landscape variable "distance from the settlements/houses" were found between the DRY and the RUD group (p<0.05).

When testing the relations between the landscape variables and frequency of single habitat specialists, we found only some statistically significant correlations (results not shown). Higher number of neighbouring habitats is correlated with higher frequency of *Trifolium montanum*, *Spiranthes spiralis* and *Centaurium erythrea*. More distant from the settlements/houses are polygons with *Potentila argentea, Carex caryophyllea* and *Thymus pulegioides*. Relation with shape index gave opposite results: *Sedum sexangulare, Polygala vulgaris* and *Spiranthes spirales* have higher frequency on plots with lower shape index; while *Orchis morio* have higher frequencies of occurrence on polygons with higher shape index (On line-Suppl. Fig. 1). Finally, we present differences between Red listed (threatened) plant species frequency with highest frequency of species *Anacamptis morio* and *Fragaria viridis* (Fig. 5). We

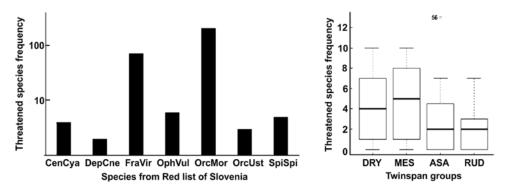


Fig. 5. Threatened species frequency of seven plant species from Red List of Slovenia (left) and frequency of threatened species in each TWINSPAN group (right). The abbreviations of species names: *Centaurea cyanus, Daphne cneorum, Fragaria viridis, Ophioglossum vulgatum, Orchis morio, Orchis ustulata, Spiranthes spiralis.* Abbreviations of TWINSPAN groups are explained in Fig. 2.

were looking for differences between the TWINSPAN groups and found the highest values for DRY and MES TWINSPAN group, but there were no significant differences among the four TWINSPAN groups.

Discussion

A large proportion of European biodiversity today depends on habitat provided by low-intensity farming practices, yet this resource is declining as European agriculture intensifies (Sutcliffe et al. 2014). One consequence of the rationalization of agriculture in Europe over the last half century has been the loss and fragmentation of traditionally managed habitats such as semi-natural grasslands (Cousins 2001). In many agricultural landscapes, as also in the Goričko area, only fragments have remained. The crucial question in the situation of the declining number and area of grasslands in the landscape is whether fragment size affects plant species diversity. This problem has received different answers in studies. However, our results show that the fragment size (large vs. small) does not affect species diversity of plants. This has been confirmed clearly already by Helm et al. (2006) – no effect of current grassland size on plant species diversity in Estonian alvar grasslands was confirmed. Our results indicate that there is no relation between species diversity (here measured as Alpha diversity of all species and habitat specialists only) and grassland area, which could be explained by the assumptions proposed by Helm et al. (2006). Long leaved perennial plants may also persist for relatively long time after the habitat has been degraded or converted to a later successional stage. In other words – since grassland fragments were assembled a long time ago and their structure is homogenous, these present remnants are still enough large to harbour more or less the entire species diversity of the previous (larger) area. The authors (e.g. Cousins and Eriksson 2002, Eriksson et al. 2002, Lindborg 2007, Kuussaari et al. 2009, Reitalu et al. 2010) summarized that it is not current but past grassland areas (prior to the drastic decline of grasslands) and the connectivity among them that affect the current species diversity in fragmented grasslands across Europe. Our study revealed that both fragment size and fragment shape index are not correlated to Alpha diversity: the shapes of the remnant grasslands are different, in many cases elongated or irregular, but this does not influence Alpha diversity.

In semi-natural grasslands, the land use treatment (management) is one of the major ecological factors, known to affect plant reproductive success and vegetation patterns (Lopez-Marino et al. 2000, Eriksson et al. 2002, Lindborg et al. 2005, Wissman 2006, Gustavsson et al. 2007). Sites with long land use continuity had higher plant species diversities than sites with shorter continuity (Gustavsson et al. 2007). Thus, we questioned if the fragments of dry grasslands are floristically homogenous. The TWINSPAN classification revealed four TWINSPAN groups, which show the influence of moisture/nutrient level (dryer – DRY; more mesic – MES), abandonment (ABA) and disturbance (ruderal – RUD). In the next step we correlated the species ables. The influence of landscape context in habitat fragmentation studies is often ignored (Wiens 1997, Hanski 1999). High habitat diversity in the surroundings might enhance the species diversity of a local site. Landscape context might affect gene flow via pollen and seed dispersal, the pressure of herbivory and the functional connectivity between habitat patches (Ricketts 2001). The complex effect of landscape context on local plant species diversity revealed the importance of the bordering habitats in many studies (Kollman and Schneider 1999, Metzger 2000, Söderström et al. 2001). High landscape diversity in the surrounding matrix provides more distinct habitat types for generalists or species with other habitat preferences, supporting the prediction that landscape diversity enhances the number of generalists, especially at the edges, but hardly of the specialists (Jonsen and Fahrig 1997). In our study area, the fragments (thought to be bigger and more connected polygons in the past) are currently surrounded by numerous habitat types: arable fields, forest edges, mesic meadow orchards, intensive meadows, vineyards, gardens, roads and even settlements. The RDA ordination diagram shows (Fig. 3) that the ABA and DRY groups are associated with a higher number of different bordering habitats types (NofHT). It is also worth mentioning that the most typical, dry fragments (DRY) and abandoned ones (ABA), which probably also derived mostly from drier (less productive) places, are found at lower altitudes and have lower shape indices.

composition in the fragments with some landscape vari-

It has been established by Tscharntke et al. (2002) that not all species depend equally on habitat area, isolation and landscape context. Habitat specialists proved to be more affected by habitat loss than generalists (Warren et al. 2001). The comparison of Alpha diversity considering all species, habitat specialists only and generalists only between the RUD groups revealed interesting results. Higher Alpha diversity was calculated for ABA, because abandonment results in colonization of species from the later colonization stages, while grassland species persist. The lowest Alpha diversity was perceived on more mesic fragments (MES), where habitat specialists are much scarcer. It was emphasized many times that early successional stages after abandonment represent the most species-rich assemblages (Denslow 1980, Balmer and Erhardt 2000). A similar pattern was revealed, when only generalists were considered. Interesting results were obtained when only habitat specialists were considered. It was confirmed again that the highest diversity of specialists are in the driest fragments, both still mowed (DRY) and abandoned (ABA), which means that among habitat specialists are mostly long-lived perennials which persist also during the earlier successional stages. Long-lived plants decline slowly and may survive for decades after environmental change (Eriksson 1996, Dahlstrom et al. 2006, Helm et al. 2006, Lindborg 2007, Hylander and Ehrlén 2013). It could be concluded that, with a reduction in grassland fragment area, the majority of species still conform to previous distribution patterns, when grassland areas were larger. There are no habitat maps from past decades to support this with exact figures. However,

another study in the NW part of the Goričko Park revealed that at the beginning of the 20th century the area was dominated by open agricultural land, with a forest cover around 39%; a landscape composition today shows forests covering around 57% of the landscape (Cousins et al. 2014). Furthermore, 65% of the forests in NW Goričko are between only 15 and 30 years old. Young forests are primarily developed on former arable fields or grasslands (Cousins et al. 2014). We claim that this is a strong proof that the grasslands in Goričko area were much larger in the past, although exact spatial data on their distribution are lacking.

High habitat diversity in the surroundings might enhance the species diversity of a local site. Furthermore, the surrounding matrix provides more distinct habitat types for generalists or species with other habitat preferences, supporting the prediction that landscape diversity enhances the number of generalists, especially at the edges, but hardly of the specialists (Jonsen and Fahrig 1997). Interesting results were obtained when the TWINSPAN groups were confirmed on the basis of the distance of polygons from settlements/houses. It seems peculiar at first sight, but the shorter distance to the settlements/houses of the most dry and regularly mowed meadows in Goričko can be explained by the fact that still mowed "best preserved" dry (oligotrophic!) fragments are close to the houses, due to the fact that houses are built on less productive soils in order to save better land for more productive (fertile) meadows and fields.

When testing the relation between frequencies of single habitat specialists we found interesting results: *Sedum sexangulare, Polygala vulgaris* and *Spiranthes spirales* have higher frequency on plots with a lower shape index. This means very small remnants of formerly larger polygons.

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But *Orchis morio* has higher frequencies of occurrence on polygons with higher shape index, which confirm the observation that this species occurs on fragments of different complex shapes, also close to houses and fields and along the roads.

Semi-dry grasslands, the remnants of traditional agricultural landscape, are today threatened mainly due to quick overgrowing, but also due to intensification or conversion to arable land. Disturbance in the form of ruderalisation (occasional fertilization or mulching) and the early stages of secondary succession emerged as much stronger drivers of community change than decreases in patch size per se. The selection of red-listed species on national level is not really supporting the conservation efforts for grasslands in Goričko area; only a few such species are present in the area. It can only be concluded that in still mowed (DRY and MES) fragments the number of red-listed species tends to be higher, although not significant and despite the relatively low number of such species. The dilemma whether biodiversity conservation should focus on a single or several large habitat fragments, or whether the protection of many small fragments (covering the same habitat area in a landscape) is of equal or even greater importance is still open (Tscharntke et al. 2012). The dilemma is commonly referred as SLOSS (single large or several small) and has been widely discussed by many authors (e.g. Ovaskainen 2002, Tjørve 2010, Tscharntke et al. 2012). In the case of Goričko Park it is of a crucial importance to preserve the regularly managed medium- and small- sized fragments, which still represent important centres for plant species diversity within the agricultural matrix of Goričko Nature Park. They are important as refugia and for future restoration activities within the park and in the wider region.

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