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Abstract	The role of fragment size, isolation and habitat diversity in the conservation of spider assemblages living fragmented landscape were studied in dry sandy grasslands (East Hungary, Nyírség). Spiders were collect using pitfall traps at eight dry grassland fragments from 2001 to 2009 from March to October fortnightly. tested the rules of island biogeography, which suggest that the species richness increases with the size at decreases with the isolation of fragments. The habitat diversity is an important factor for species richness since large areas usually have more habitats; therefore, the number of species may be higher in these are During the 9-year study period, altogether 10,544 individuals belonging to 106 species were collected. Contradicting the classical theory, we found a significant negative relationship between the total number spider species and the grassland size, while the ratio of sandy grassland specialist spider species increase with fragment size. The relationship between the ratio of generalist species and the fragment size was not significant. The overall species richness and the isolation of studied grasslands did not show a significant relationship. The ratio of sandy grassland specialist species decreased, while the ratio of generalist species richness. We concluded that to conserve the habitat diversity did not show any effect on spider species richness. We concluded that to conserve the habitat specialist species it is recommended to preserve the la and least isolated grassland fragments, furthermore to increase the size of small fragments with the restorat of the adjacent croplands.		
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Below is the link to the electronic supplementary material.

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#### ORIGINAL PAPER

- Large and least isolated fragments preserve habitat 2
- specialist spiders best in dry sandy grasslands in Hungary 3
- 4 Roland Horváth · Tibor Magura · Csaba Szinetár · János Eichardt ·
- 5 Béla Tóthmérész
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- 9
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#### Introduction

Reduction and fragmentation of natural and semi-natural habitats became a major threat to biodiversity and these processes cause a significant loss of biodiversity (Harrison and Bruna 1999; Henle et al. 2004). Habitat reduction and alteration negatively influence the distribution, abundance and diversity of characteristic plant and animal species of natural habitats (Quinn and Harrison 1988; Baur and Erhardt 1995; Gilbert et al. 1998) and have been regarded as the main threats to compositional and structural biodiversity (Noss 1991; Didham et al. 1996). Therefore, disturbance sensitive species may disappear from natural and semi-natural habitats.

During the last century there was a significant reduction in the size and continuity of diverse natural and semi-natural grasslands in whole Europe (Poschlod and WallisDeVries 2002; WallisDeVries et al. 2002). Grasslands are usually sensitive to habitat alteration and fragmentation (Poschlod and WallisDeVries 2002; Novák and Prach 2010). The reasons for alteration and fragmentation are the increase of large-scale farming and abandonment of the traditional management practices (Taboada et al. 2011). The efficiency of conservation actions depends on the management activities (livestock grazing, mowing and burning), which are important for the maintenance and preservation of grasslands (Poschlod and WallisDeVries 2002). These natural and semi-natural grasslands often harbor a high number of invertebrate species including several threatened, rare and endemic species (WallisDeVries et al. 2002; Kotze and O'Hara 2003; Woodcock et al. 2005).

During the last decades the increase of agricultural management has resulted in a significant decrease and degradation of the sandy grasslands. Therefore, this grassland type is the most threatened ecosystem in Central Europe (Eichberg et al. 2007). Agricultural intensification caused a decrease in biomass production and vegetation coverage, whereas it increased the areas covered by bare sand. Nowadays the development of sustainable agriculture can help the conservation of sandy grassland ecosystem (Xu et al. 2001).

Spiders are diverse and abundant predatory animal group of grasslands. They have been studied in various grassland types: sand dunes (Bonte et al. 2002), limestone grasslands (Rushton 1988) and moorlands (Cameron et al. 2004). However, spider assemblages of dry sandy grasslands were investigated rarely (but see Szinetár et al. 2005; Horváth et al. 2009; Buchholz 2010). Spider assemblages are important in natural and agricultural ecosystems by shaping the structure of arthropod communities due to the fact as that they are generalist predators and they provide biological pest control (Marc et al. 1999; Sunderland and Samu 2000). In general, they are useful taxa to understand the effects of land use changes, because they have short life cycles, high abundances, their ecology and systematics are well known and they are sensitive to environmental changes and fragmentation (Miyashita et al. 1998; Marc et al. 1999).

The classical theory of island biogeography explains patterns of both animal and plant species composition on real islands (MacArthur and Wilson 1967). This theory emphasizes the importance of the size and isolation as the two major factors determining the species richness of islands. This assumption predicts that there are more species on large islands than on small ones, because large islands have larger populations, therefore the extinction rates are lower on these islands. Isolated islands have fewer species than islands with close



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vicinity to other islands or mainland, because they have lower immigration rates; therefore the colonization of these islands is more difficult. Although this theory originally used to explain variation in diversity on real islands, it has later been applied to inland habitat islands. Recently, more authors refined the original hypothesis of the classical island biogeography theory as the surrounding habitats (edge and matrix) could have important effects on the species richness (Lövei et al. 2006; Devictor and Jiguet 2007; Magura and Ködöböcz 2007).

The habitat diversity is also an important factor for species richness especially at local scales (Davidowitz and Rosenzweig 1998). Large areas usually have more habitats; therefore in these areas there may be more species than in the smaller ones, which have less different habitat types (Davidowitz and Rosenzweig 1998; Ricklefs and Lovette 1999). Of the approximately same sized habitats, the more heterogeneous can maintain assemblages with higher species richness and a greater number of individuals than those which are more homogeneous (MacDonald and Johnson 1995). Nevertheless there are such smaller, spatially heterogeneous habitats, which also contain species rich assemblages (Martin et al. 2012).

Species with different habitat affinity (generalist and specialist) respond variously to the habitat size (Zabel and Tscharntke 1998; Bonte et al. 2002), isolation (Magura et al. 2001; Jonsson et al. 2009) and habitat diversity (Ricklefs and Lovette 1999; Kruess and Tscharntke 2002). Therefore, it is important to investigate the groups of species with different habitat affinity separately. Besides overall species richness we also analyzed the number of generalist and specialist species. In this study those species which occurred in high number in most grassland types were categorized as generalists, and those open habitat species which were associated with sandy soils were regarded as specialists (Buchar and Ruzicka 2002).

In our study, spider assemblages in dry sandy grassland fragments were investigated in Eastern Hungary (Nyírség) to study the relationship between fragment size, isolation and habitat diversity and the species richness of spider assemblages. Our hypotheses were as follows: (i) The fragment size and isolation should affect the opposite way the habitat specialist species richness and the generalist species richness. We expect that the increase of habitat size and decrease of isolation increases the habitat specialist species richness, while the generalist species richness should increase with the decrease of habitat size and increase of isolation. (ii) More isolated grassland fragments should increase the species richness, as the generalist species can enter from the surrounding habitats. (iii) Furthermore, increasing habitat diversity within habitat fragments should be more favorable for generalist species than sandy grassland specialist species, because generalist species can settle down permanently in more habitat types.

#### Materials and methods

110 Study area

- Eight dry sandy grassland fragments were selected for the investigation of ground-dwelling
- spider assemblages, located in the Nyírség region of the Great Hungarian Plain (Eastern
- 113 Hungary) (Table 1; Electronic Supplementary Material (ESM) Map 1). The sampling sites
- were selected in the way that every sampling site has the same vegetation type, the same
- intensity of management (moderate grazing) and these are embedded in the same matrix.
- 116 These criteria were assured for eight fragments. In the 19th century the natural habitat



**Table 1** Habitat characteristics of the eight studied dry sandy grassland fragments in Nyírség

Site name	Size (ha)	Inverse isolation index (ha)	Habitat diversity (Shannon index)
1. Bagamér	99.0	121.5	0.40589
2. Bátorliget	249.7	122.1	0.22711
3. Hajdúbagos	250.6	58.3	0.40917
4. Martinka	353.5	137.6	0.48714
5. Nyíregyháza	188.7	130.5	0.07685
6. Nyírtura	29.1	1.6	0.24182
7. Rohod	51.8	7.3	0.28886
8. Újtanya	2.3	137.3	0.16139

types in this region were dry habitats (sandy grasslands and sandy oak woods) and wetlands (marshes, fen meadows and mires). Due to increasing agricultural intensification since the mid 20th century these habitats have disappeared or have become highly fragmented. In present days, these fragmented dry sandy grassland fragments are encompassed by arable lands and non-native tree plantations. In all dry sandy grassland fragments, the typical grassland vegetation was *Cynodonti-Festucetum pseudovinae* (Török et al. 2008). These fragments have been lightly or moderately grazed by cattle and sheep. All of the studied fragments were surrounded by both arable lands (maize and corn) and non-native tree plantations (black locust and ennobled poplar species). Thus, the matrix habitats were similar to all the investigated fragments. The shortest distance between the studied grassland fragments was 2 km, while the distance of the two furthest fragments was 75 km.

## Sampling design

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130 (2001–2009). There were 10 randomly placed traps in all studied fragments. Each trap was 131 at least 50 m from the grassland edges, in order to avoid edge effects (Horváth et al. 2002). 132 Traps consisted of 100 mm diameter plastic cups and contained about 200 ml 70 % eth-133 ylene glycol as a killing-preserving solution. Pitfall traps were protected by fiberboard 134 from litter and rain. Spider species were collected every 2nd week from the end of March 135 to the end of October. Spiders were identified to species level using standard keys (Heimer 136 and Nentwig 1991; Roberts 1995). Nomenclature follows Platnick (2012).

Spiders were collected using pitfall traps during the 9-year investigating period

#### Data analyses

The size of the investigated dry sandy grasslands was measured using the ArcGIS program on a digitized 1:25000 map. Isolation of a habitat island (grassland fragment in our case) is often measured as the distance to the nearest fragment, although isolation also depends on the size of the nearest fragment. Isolation of the grassland fragments was measured by the inverse isolation index, defined as the total dry sandy grassland size within a radius of 1,000 m around the investigated grassland fragment. Its value decreases as the isolation of the grassland increases (Magura et al. 2001). The radius was selected 1,000 m, because spiders can cover this distance easily with their silk (Foelix 2011). Identification of different vegetation patches was based on aerial photographs (see Map 1 in ESM). Based on this map we expressed the habitat diversity with Shannon index of diversity.



The relationships between the habitat characteristics (size, isolation and habitat diversity) and the species richness of spider assemblages were examined by multiple linear regression analyses (Kutner et al. 2005). We analyzed the 9 years average value of each trap, weighted with the standard error (SE), depending on fragment size, isolation and habitat diversity. The total number of species, the ratio of generalist species and the ratio of sandy grassland specialist species were analyzed. Before the statistical analyses the given traps catches were pooled from the nine trapping years. The distribution of data used in the multiple linear regression analyses was normal (tested by the Kolmogorov–Smirnov test, Sokal and Rohlf 1995). Residuals are examined visually for normality; the distribution of residuals was normal (Kéry and Hatfield 2003).

## Results

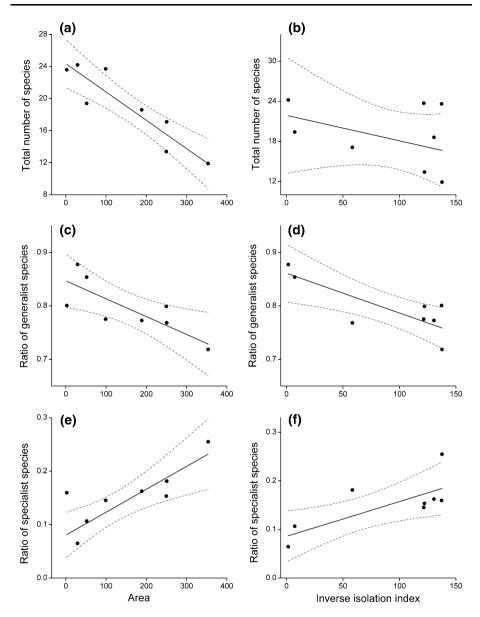
During the 9-year study period, 10,544 spider specimens belonging to 106 species were collected from the eight dry sandy grassland fragments (Electronic supplementary material (ESM) Table 1). Twenty-one species (about 20 % of the collected species) were identified as sandy grassland specialist species.

Multiple linear regression analysis pointed out that only the size of the grassland fragments had an influence on the overall species richness. The total number of spider species and the size of the dry sandy grassland showed a significant negative relationship (Fig. 1a; Table 2). There was no statistically significant relationship between the overall species richness and the inverse isolation index of grassland fragments (Fig. 1b; Table 2). The ratio of generalist species decreased with the increasing of fragment size, but this relationship was not significant (Fig. 1c, Table 2), while their ratio significantly increased with the decreasing of inverse isolation index (Fig. 1d; Table 2). A significant positive relationship was found between the ratio of open-habitat species associated with sandy soils to the total number of species and the size of grasslands (Fig. 1e; Table 2), showing the importance of habitat specialist spiders with increasing fragment size. The ratio of habitat specialist species and the inverse isolation index of the fragments also showed a significant positive relationship (Fig. 1f; Table 2). There was no significant importance of the habitat diversity on any of the dependent variables (Table 2).

#### Discussion

- 178 Effect of fragment size
- The results of several authors corroborated the theory of island biogeography as they pointed out a significant positive correlation between the overall species richness and the size of habitat islands (Toft and Schoener 1983; Webb and Hopkins 1984; Ricklefs and Lovette 1999; Tscharntke et al. 2002; Watson et al. 2005). In other studies the number of all animal species did not depend on the size of the habitat (Dauber et al. 2006; Kapoor 2008; Kappes et al. 2009; Muriel and Kattan 2009). Our results contrary to the classical theory of island biogeography showed that there was significant negative relationship between the size of the grassland fragments and the total number of spider species. Similarly to our results other investigations in which ground beetles (Webb and Hopkins 1984; Bauer 1989; Halme and Niemelä 1993; Magura et al. 2001; Jonsson et al. 2009) and spiders (Jonsson et al. 2009; Bailey et al. 2010) were studied showed that contrary to the





**Fig. 1** Relationship between the size of dry sandy grassland fragments and the total number of spider species (**a**), the ratio of generalist species (**c**), the ratio of sandy grassland specialist species (**e**) and relationship between inverse isolation index and the total number of spider species (**b**), ratio of generalist species (**d**), ratio of sandy grassland specialist species (**f**). *Dashed lines* represent the confidence bands (95 %)

classical theory of island biogeography there was significant negative relationship between the size of the fragments and the total number of species. In all of the papers that investigated only the habitat specialist species they found that this relationship was significantly positive with the size of the habitat similarly to our results (Zabel and Tscharntke 1998;



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Table 2 Relationship between the spider species and the studied habitat characteristics, determined by multiple linear regression

	Overall species richness $F_{(3,7)} = 10.332$ p = 0.024 r = 0.941	Ratio of generalist species $F_{(3,7)} = 18.329$ p = 0.008 r = 0.966	Ratio of sandy grassland specialist species $F_{(3,7)} = 11.818$ $p = 0.019$ $r = 0.948$
Size of the grassland fragments	_*	ns	+*
Inverse isolation index	ns	_*	+*
Habitat diversity calculated by Shannon index	ns	ns	ns

<sup>\*</sup> Significant positive or negative relationships are indicated: p < 0.05

Bonte et al. 2002; Sanchez and Parmenter 2002; Scott et al. 2006; Mohd-Azlan and Lawes 2011).

Real islands differ significantly from habitat islands. The surrounding habitats around real islands are usually rigorous to both plant and animal species; therefore species can penetrate with a low chance on real islands. Contrarily terrestrial habitat islands are surrounded by a wide range of matrix habitats which are less unfavorable for species. Since both the habitat specialist and generalist species are alike revealing for the inland habitat islands, the observed species-area relationship may be positive, negative or neutral depending on the ratio of habitat specialist and non-habitat specialist species. These results reveal the refinement necessity of the original hypothesis. When we would like to apply this prediction to mainland habitat islands, we have to separate specialist and non-habitat specialist species. The specialist species perceive the habitat fragments as islands (they can survive and reproduce only in these fragments), while the non-habitat specialist species occur in large numbers both in the habitat fragments and the adjacent habitats (Magura et al. 2001; Bonte et al. 2002; Cook et al. 2002; Mohd-Azlan and Lawes 2011).

## Effect of habitat isolation

The isolation of the habitat island may also be very important in the occurrence of the habitat specialist and the non-habitat specialist species (Magura et al. 2001; Jonsson et al. 2009). However, there were investigations when the species number did not depend on the habitat isolation (Webb and Hopkins 1984; Jonsson et al. 2009) or the relationship was negative (Toft and Schoener 1983; Usher et al. 1993; Miyashita et al. 1998; Bailey et al. 2010). We found positive correlation between the total number of species, the ratio of generalist species and the isolation of the habitat fragments, but it was not significant in case of total species pool. The reason for this is that the adjacent habitats around grassland fragments may be the source of the generalist species; therefore, these spiders can spread from the agricultural areas to the dry sandy grassland fragments (spill-over effect, see Tscharntke et al. 2012).

The sandy grassland specialist species richness showed a negative significant relationship with isolation in most studies (Magura et al. 2001; Watson et al. 2005; van Noordwijk et al. 2012), but neutral relationship was also reported (Sanchez and Parmenter 2002; Cardoso et al. 2010). In our study there was a significant positive relationship between the number of sandy grassland specialist species and the inverse isolation index. This result showed that due to habitat fragmentation, the increased isolation between the



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grassland fragments significantly decreased the number of these species. Therefore, the isolation of grassland fragments had more important effect on sandy grassland specialist species, than on generalist species, especially as the ratio of generalist species increased, while the ratio of sandy grassland specialist species decreased with the increase of isolation. Although spiders have a good spreading ability, it is likely that habitat specialist species could not persist in the neighboring arable lands and non-native tree plantations permanently, because these habitats were hostile for them. These unfavorable habitat types acted as barriers for sandy grassland specialist species, thus their proportion decreased in the more isolated fragments. Furthermore, if a sandy grassland specialist species disappears from a given grassland fragment, this species can not recolonize the grassland fragment because it is isolated.

### The impact of habitat diversity

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Usually larger habitat diversity increases the number of species in a given area (Ricklefs and Lovette 1999; Weibull et al. 2000; Kruess and Tscharntke 2002), but in some cases, the relationship between habitat diversity and the species richness is not significant (Ricklefs and Lovette 1999; Kruess and Tscharntke 2002). In contrast to our hypothesis the habitat diversity of the grassland fragments did not have any significant effect on the ratio of generalist species, either overall species richness, or the ratio of sandy grassland specialist species. Since there were various adjacent habitats around dry sandy grassland fragments (wetlands, wet grasslands, arable lands, tree plantations and other dry sandy grasslands), the generalist species found suitable circumstances and they could also settle down permanently in these habitats in large numbers. Therefore, the increasing habitat diversity within grassland patches did not influence the generalist species number. Our result suggest, that the size and the isolation of the dry sandy grassland fragments were more important for sandy grassland specialist spiders than the habitat diversity, because the species number was significantly influenced only the size and isolation of fragments, while the habitat heterogeneity did not affect the number of species of these spiders. Furthermore, the distribution of the habitat specialist species also depends strongly on the vegetation type, the structural heterogeneity of the vegetation and the special microclimate conditions in a given habitat (Foelix 2011).

## Habitat specialist and non-habitat specialist species

We pointed out that overall species richness and ratio of generalist species increased with the decreasing size of grassland fragments and the increasing of grassland isolation. Sandy grassland specialist species played a more important role in the assemblage when the size of the habitat fragments increased. We can explain this fact with the following two reasons: (i) generalist species can enter in higher numbers to the more isolated grassland fragments, because the proportion of their source habitats (especially arable lands) is higher around these fragments; (ii) sandy grassland specialist species need a minimum size of their habitat, therefore they occur in higher numbers only in bigger grassland fragments; furthermore they can not immigrate to more isolated fragments, because the environmental factors of the adjacent habitats are hostile for them.

Hermann et al. (2010) pointed out that habitat fragmentation and immigration of generalist species from adjacent habitats caused a significant loss of habitat specialist species richness, at the same time these spiders increased the total species richness. Furthermore, strongly isolated fragments were more favorable for the majority of



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generalist species because they occurred in greater number in these fragments than in the less isolated fragments. There are several publications in which spiders in different grassland types were studied and stressed that landscape features (habitat connectivity, structural heterogeneity, landscape structure) also played a very important role in species composition and abundance (Bonte et al. 2002; Finch et al. 2008). The habitat specialist species demand larger structural heterogeneity and special environmental conditions; therefore they need to have a minimum area size, where the necessary factors are at their disposal (Bonte et al. 2002; Scott et al. 2006) to have a viable population size (Bonte et al. 2002). Moreover, Bailey et al. (2010) showed that source-sink dynamic and trophic interactions may decrease the spider species richness, which correlates with increased area size. Sink populations are preserved by immigrant species, but the number of these species decrease with fragment size. Besides, the important spider predators (birds and wasps) may be present in smaller numbers in small fragments than their preys, therefore spiders could benefit from lower predation risk in smaller fragments (Bailey et al. 2010). The habitat size and the isolation also influences the extinction rate of spider species, because this rate is positively related to isolation and negatively to habitat size (Toft and Schoener 1983).

#### Conclusions

The once continuous grasslands became the most threatened habitat types in Europe by the increase of intensity of agricultural practices. Some of the grasslands disappeared, while the other parts of grasslands have become fragmented and reduced their size dramatically. These processes jeopardize the survival of most habitat specialist and rare animal and plant species. Therefore, nowadays there is a growing demand for the conservation of the different grassland types. The main goal is to preserve the larger continuous natural grasslands and to restore the remnant areas of natural and semi natural grasslands, because these remaining fragments can also maintain a diverse fauna. Our results showed that only small and highly isolated fragments can preserve a species rich spider assemblage in the Nyírség region due to the large number of generalist species. In contrast, the habitat specialist species characteristic to dry sandy grasslands occurred in higher number only in the large and less isolated fragments as they can not survive in the adjacent arable lands and they need to have a minimum size of the fragment. Based on our results for the conservation of spider species, we recommend the following measures during a nature conservation program. First as habitat specialist spider species are also present in the small grassland fragments, thus these fragments should be retained to serve as source habitats. Second, since the fragmentation and isolation decrease the number of habitat specialist species in the dry sandy grassland patches, therefore the further fragmentation has to be stopped. Third, there is a need to increase the size of these fragments with the restoration of grassland on the adjacent croplands that may contribute to the conservation of the sandy grassland specialist species.

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