COMPARISON OF WILD BEE COMMUNITIES OF THREE SEMI-NATURAL MEADOW HABITATS AT HARGHITA-COVASNA REGION, TRANSYLVANIA, ROMANIA

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In the temperate climate wild bees are the most important pollinator organisms. Pollination is essential for the communities of semi-natural habitats since this ecosystem service directly affects plant reproduction. The diversity of wild bees living in such areas is remarkably high, but they are susceptible to various anthropogenic influences.

In our study, the composition and structure of wild bee communities were examined in Romania (Transylvania) at three semi-natural areas near Filia, Merești and Vârghiș. The surveyed areas were used as extensive meadows under relatively low but slightly different anthropogenic influence levels. We collected bees in these areas at several places (9 sampling points/area) by individual netting four times during the season. In the studied areas, 129 bee species were found, which makes up about 18% of the approximately 726 wild bee species registered in Romania. In addition to the high number of species, we also observed high diversity values. Our results showed that, even at our sampling site closest to the human settlements, the extensive use of the surveyed areas as meadows allows the development of diverse, species-rich bee communities.

Keywords: bumble bee, wild bee communities, diversity, pollination, solitary bee, Transylvania, meadow.

INTRODUCTION

One of the main reasons for the biodiversity decline is the intensive agricultural land use, which reduces natural habitats (Newbold *et al.* 2015). The habitat fragmentation and land use change result is a homogeneous landscape structure (Tilman *et al.* 2001). Intensive agriculture results in a loss of biodiversity (de Heer *et al.* 2005), which also leads to a decline in ecosystem services, including pollination, in many regions of the world (Kremen *et al.* 2002, Kremen *et al.* 2007, Potts *et al.* 2016).

The role of bees in pollination is particularly significant as they are responsible for pollinating many nutrient-rich plants important for human nutrition (ELLIS et al. 2015). More than 70% of the essential food crops depend on bee pollination (Klein et al. 2007), a service worth €153 billion worldwide (Gallai et al. 2009). The presence of wild bees in crop production is relevant even when the presence of honey bees is strong, because wild bee communities often prove to be more efficient pollinators than honey bees and inter-species interactions can increase pollination efficiency (Brittain et al. 2013, Woodcock et al. 2013). Diverse bee communities provide a high and stable supply of pollination services (Hoehn et al. 2008, Eeraerts et al. 2020, MacInnis et al. 2020), but recently the intensification of agriculture has greatly reduced the diversity and abundance of bee communities (Goulson 2003, Biesmeijer et al. 2006, Goulson et al. 2008, Potts et al. 2010). In more recent decades new farming systems and techniques have been developed through organic farming and agri-environmental schemes (AES), to mitigate the impacts mentioned above (Samu et al. 2010, Andersson et al. 2012, Batáry et al. 2015).

Pollination by wild bees is vital not only in agricultural areas but also in semi-natural habitats as it has a direct effect on the reproduction of plants, which form the basis of all (semi)natural communities. The main drivers of wild bee decline are related to historical landscape change, loss of natural and semi-natural areas, loss of nesting and feeding grounds, and loss of the most important flower sources (Goulson *et al.* 2005, Potts *et al.* 2005, Senapathi *et al.* 2015, Baude *et al.* 2016, Sárospataki *et al.* 2016). The breaking up and the utilisation of semi-natural areas for intensive cultivation lead to habitat fragmentation, which can adversely affect the size and interconnectivity of remaining habitat patches (Hooke *et al.* 2012). It may reduce gene flow between pollinator populations in the short term and may impact population persistence in the long term (Darvill *et al.* 2010).

In Europe, the natural, especially grassland habitats without any anthropogenic effects are almost absent (Evans 2006). However, high nature value (HNV) grasslands are present in Europe, and it is essential to have more information on the condition of these habitats (Veen et al. 2009). The main reasons for the existence of the diverse semi-natural grasslands are local, regional (Myklestad & Sætersdal 2003), historical (Marini et al. 2009), traditional, long-term, small-scale and non-intensive land use (Poschlod et al. 2005, Babai & Molnár 2014, Dorresteijn et al. 2015). These systems in Europe have developed and sustained landscapes having a high natural, cultural, and aesthetic value (Dahlström et al. 2013). In the Romanian section of the Carpathians, there are large semi-natural, HNV pastures and hay meadows (Huband et al. 2010). However, the intensity of land use (e.g., the number of mowing per year and manuring) of these habitats is reciprocally proportional with the proximity of the closest human settlements (Babai et al. 2015, Babai & Molnár 2016).

In this study, the community structure of wild bees in three semi-natural grassland areas in Romania (Transylvania) were examined. There was no information on the species composition and richness, and diversity of the pollinator communities of this area. The aim of our study was to (1) assess the diversity and species distribution of the wild bee communities living in the studied semi-natural, HNV areas, (2) collect new data on the rare and faunistically interesting species, (3) compare the bee communities of the three areas to understand the effects of various human presences.

MATERIAL AND METHODS

Our study was carried out in 2018 in Transylvania, in Harghita and Covasna counties, in three semi-natural, HNV areas where extensive farming takes place. In all three research areas, the average altitude is 530–630 m. The studied areas are located relatively far from the closest villages, have preserved quite well the complex natural habitat consisting of grassland-woodland-scrub mosaics. The grasslands are mainly used as meadows. Mowing on mosaic grassland patches occurs at different times, thus providing a continuous food resource for pollinators. The studied meadows and grassland patches are part of valleys in all three cases. Although all three valleys are semi-natural, HNV meadows, there were little but noticeable differences in anthropogenic impacts between the sites.

One characteristic of traditional farming in this region is more intensive agriculture on lands closer to the villages. The majority of traditional treatments (indeed) increase landscape diversity, though not in all cases. Treatments that can have a negative impact on diversity are the extensive use of fertilisers and mowing more than twice a year. These treatments are carried out mostly on lands close to villages. Nevertheless, thanks to these practices, a larger amount of hay can be harvested from these neighbouring areas, which will suffice to feed the animals. Therefore, on remote areas one mowing a year is sufficient, and this allows more plants to produce seeds and thus increase the diversity of remote meadows (Dahlström *et al.* 2013, Babai *et al.* 2015, Kun *et al.* 2019).

Further reasons of the higher anthropogenic effect on the closer areas may be poor infrastructure (lands closer to the villages are easier to reach); protection of the areas (lands closer to villages can be protected easier against the damages caused by wildlife); traditional farming (currently this is less common, but up until recent years farmers were still using horse-drawn carts for harvesting).

The Filia (F) area (46.1731241°N, 25.6236372°E) is the closest to human settlements (the average geographic distance of this sampling site from the closest human settlements is about 2000 m), with meadows, forest patches and few arable lands. The other two areas are relatively far from human settlements, and therefore the lands are used less intensively. Vârghiş Gorge (Cheile Vârghişului, V) area (46.2034539°N, 25.5344264°E) is a nature reserve, located furthest from human settlements (the average geographic distance of this sampling site from the settlement is about 7500 m), characterised by meadows and forest patches. The Mereşti (M) area (46.2394164°N, 25.5322366°E) is located at a medium distance from human settlements (the average geographic distance of this sampling site from the settlement is about 5000 m), characterised by meadows, pastures and forest patches. The distances between the sampling areas were as follows: F–M 10000 m; F–V 7000 m; M–V 3700 m.

In all three valleys sampling was performed four times in 2018 (once in May, twice in June, once in July). The sampling area was a circle with a radius of 1200 m that contained 9

sub-sampling sites. We randomly chose the 9 sub-sampling sites by picking the flower-rich meadows. Two people performed collection rounds for 20 minutes separately at each site while each of them walked along a 200 m transect. The distance between the two transects was approximately 50 m, and there was no overlap between the transects. All observed wild bee individuals were captured using a butterfly net and preserved in 70% ethanol. Individuals were identified at species level by a taxonomic expert. The bees sampled at the same time by the two collectors were pooled.

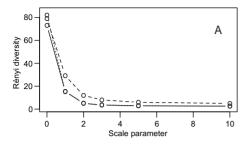
The sampled specimens were divided into two groups, bumble bees and other wild bees (later referred to as solitary bees), as these may show significant differences partly in social behaviour and partly in home-range size (Gathmann *et al.* 2002, Michener 2000). Diversity indices (Shannon, Simpson) and diversity profiles (Hill 1973) were calculated for both groups separately and the entire wild bee community. The endangered status of the species was determined based on the European Red List (Nieto *et al.* 2014). To characterise the differences of wild bee communities between areas, we compared the species composition and dominance relationships as well. Dominant and subdominant species were those with a dominance of more than 1% and 0.5%, respectively. To compare species composition of the bee communities, we calculated Jaccard similarity indices analysing the areas in pairs.

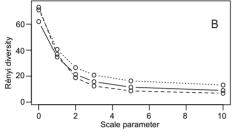
RESULTS

During the sampling period, a total of 1882 individuals of 129 wild bee species were observed in the three areas. The complete list of species and the total number of individuals per area is provided in the Appendix. The collected material included 12 bumble bee species (1049 individuals) and 117 solitary wild bee species (833 individuals). According to the IUCN European Red List, one species can be classified as EN (Endangered), 11 species as NT (Near Threatened), and 24 species as DD (Data Deficient); the other species belong to the LC (Least Concern) category (see Appendix).

Table 1. Community	parameters of the bee	assemblages at the	different sampling sites.

Site	Filia	Meresti	Vârghis	Total
			,	
No. of individuals	454	638	790	1882
No. of species	79	73	82	129
No. of unique species at the given site	22	13	25	
No. of unique species with more than 1 specimen	5	4	5	
Shannon diversity	3.37	2.74	2.72	
Simpson diversity	0.92	0.80	0.81	
Shannon for bumble bees	0.84	1.04	1.30	
Simpson for bumble bees	0.39	0.46	0.58	
Shannon for solitary bees	3.62	3.55	3.70	
Simpson for solitary bees	0.95	0.95	0.96	





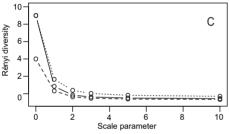


Fig. 1. Rényi diversity profiles of bee communities at the different sampling sites (Vârghiș: dotted line, Merești: solid line, Filia: dashed line). A: profiles for all species; B: profiles for solitary bee species; C: profiles for bumble bee species

Table 2. The number of common species (above the diagonal) and the Jaccard similarities (below the diagonal) of the given two sampling sites.

	Filia	Merești	Vârghiș
Filia		11	8
Merești	0.578		12
Vârghiș	0.489	0.563	

Species and individual numbers, and the number of unique species (species found only at the given area (F, M or V)) were highest in the area V (Table 1). Both the Shannon and Simpson indices showed very little difference between areas (Table 1). The diversity profiles of the areas also had a very similar course (Fig. 1). They intersect, so no significant difference in diversity could be assessed. The diversity profiles calculated for bumble bees is the only one where the curves did not intersect; nevertheless, they ran very close to each other. The number of unique species was highest at area V, followed by F and then the M (Table 1). The number of common species at areas M-F and M-V was almost the same, while significantly fewer common species were found at the areas F-V (Table 2). The values of the Jac-

card similarity indices also showed the smallest similarity between the areas F–V, while area M was almost equally similar to both areas V and F (Table 2).

DISCUSSION

Our study has yielded important and exciting faunistic results. In recent faunistic works on Romanian bumble bees (Ban-Calefariu & Sárospataki 2007, Tomozi & Toma 2011) faunistic data from this region (border of Harghita and Covasna counties) are lacking. Information on species on the European Red List is particularly important as new data in both endangered and data-

deficient species may be necessary from a conservation perspective (Nieto *et al.* 2014). Out of the 11 species classified in the NT category, 5 were found as a single specimen only, but the other 6 proved to be more common, and some species proved to be dominant (*Andrena ovatula* (Kribz, 1802), 65 individuals) or subdominant (*Andrena hattorfiana* (Fabricius, 1775), 14 individuals) in our study. Out of the 24 DD species, 17 were found in more than one specimen, and one of these was a subdominant species (*Andrena schencki* Morawitz, 1866, 11 individuals) of the community.

From a conservation biology perspective, it is fundamental to study the bee communities in semi-natural habitats, as the bees are the most important pollinators and have a direct impact on plant reproduction and thus on natural habitats (BAWA 1990, ASHMAN et al. 2004). MATACHE and BAN (2006) and BAN and Tomozei (2006) synthetised the data of publications and museum materials of Megachilidae, Andrenidae, Anthophoridae and Apidae from Dobrogea region. The data set was collected from more than 50 sampling sites, and it spanned several decades. They found 58 Megachilidae, 20 Andrenidae, 14 Anthophoridae, and 7 Apidae species. An intensive faunistical survey in Maramures (6 years, 41 sampling sites) reported 12 Megachilidae, 5 Anthophoridae and 17 Apidae species (BAN 2005). On the other hand, in very similar faunistical surveys in Hungary (Sárospataki & Fazekas 1995, Havas et al. 2008, Sárospataki et al. 2009), the number of collected bee species ranged 65-124. In our study, we collected 23 Megachilidae, 21 Andrenidae, 19 Anthophoridae and 12 Apidae species, and the total species number was 129 in a one-year field survey. Hence, we can argue that the species richness of our sampling areas was very high. About 18% of the approximately 726 wild bees and 30% of the 40 Bombus species registered in Romania (http1.) were collected in our study areas.

In comparison to the species occurring in the Maramureş (Romania) region (Ban 2005) and Southern Transylvania (Kovács-Hostyánszki *et al.* 2016), a significant difference could be observed regarding the proportion of the wild bee individuals belonging to the different families. In our study, the largest number of collected specimens belonged to the family Apidae (61.3%), and Halictidae (20.4%), while in the Maramureş region the proportion of individuals of the two dominant families was reverse (Halictidae: 65.7%; Apidae: 19%). Similar results were found in Southern Transylvania: Halictidae (62%), Apidae (17.2%).

In addition to the high number of species, we also found high diversity values. In the three sample areas, the diversity of the communities did not significantly differ from each other (Table 1, Fig. 1). The highest number of species was observed in the V area, although the differences between the areas were not significant (F: 79, M: 73, V: 82 species). The two topographically closest areas were M and V, and the similarity in their species composition was relatively high. However, area M showed fairly high similarity in species

composition to area F, which is topographically more distant from it, while area V and F, which are relatively close to each other, had the least similarity. Area V is a protected area (http2.) and presents the most natural habitats, while area F is the most anthropogenic and closest to the human settlements.

The responses of pollinators to human-induced habitat disturbances are predominantly negative (Winfree et al. 2011); however, the direction and strength of pollinator response are variable (WINFREE et al. 2009, 2011, QUIN-TERO et al. 2010, Doré et al. 2021). Furthermore, changes in pollinator richness and abundance may vary among disturbance types (Doré et al. 2021) and were significantly reduced by habitat change only in systems experiencing extreme habitat loss (Winfree et al. 2009). Other community parameters (e.g. species composition, the relative abundance of specialist and generalist species) are often more sensitive to the anthropogenic effects (Winfree et al. 2011, Quintero et al. 2010). Our study sites were semi-natural, HNV meadows, and there were little variances in anthropogenic effect between the sites derived only from the mowing intensity (see Materials and methods, Dahlström et al., 2013, Babai et al. 2015, Kun et al. 2019). This little variance in the anthropogenic influence between the areas was probably not enough to cause significant species number and diversity differences based on our results. However, although not strongly, differences in anthropogenic effects can be detected based on the similarities/differences in species composition. After all, the two areas with the most different anthropogenic effects (V–F), showed the least similarity in species composition, even though they were closest to each other.

Our study suggests that the extensive use of the surveyed areas as meadows allows for developing diverse, species-rich bee communities, even at our sampling site closest to the human settlements. The monitoring of the bee communities of these and similar meadows could present useful data for the conservation of these high-value grasslands.

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APPENDIX

The number of individuals, dominance and IUCN status of collected species. EN (Endangered), NT (Near Threatened), DD (Data Deficient), LC (Least Concern) species

Species	Filia	Merești	Vârghiș	Sum	Dominance	IUCN
Andrena bicolor		6		6	0.32	LC
Andrena combinata			2	2	0.11	DD
Andrena dorsata	4	5		9	0.48	DD
Andrena flavipes	56	12	25	93	4.94	LC
Andrena fulvicornis	4			4	0.21	DD
Andrena hattorfiana	3	6	5	14	0.74	NT
Andrena labialis	2	1	3	6	0.32	DD
Andrena labiata	1	2	1	4	0.21	DD
Andrena minutula		4		4	0.21	DD
Andrena minutuloides	5	1		6	0.32	DD
Andrena nitida	1	1	1	3	0.16	LC
Andrena nitidiuscula	2		1	3	0.16	LC
Andrena ovatula	22	21	22	65	3.45	NT
Andrena pandellei	1	3	3	7	0.37	LC
Andrena proxima		1		1	0.05	DD
Andrena schencki	6	2	3	11	0.59	DD
Andrena strohmella			1	1	0.05	LC
Andrena subopaca			1	1	0.05	LC
Andrena susterai	1			1	0.05	DD
Andrena taraxaci	4		2	6	0.32	DD
Andrena ungeri	1			1	0.05	LC
Anthidium manicatum			2	2	0.11	LC
Anthophora aestivalis	2	2	1	5	0.27	LC
Anthophora furcata	1	1		2	0.11	LC
Bombus argillaceus		4	4	8	0.43	LC
Bombus campestris		2	1	3	0.16	LC
Bombus hortorum	3	12	43	58	3.08	LC
Bombus humilis	15	49	75	139	7.39	LC
Bombus hypnorum		3	1	4	0.21	LC
Bombus lapidarius		2	7	9	0.48	LC
Bombus pascuorum	9	23	41	73	3.88	LC
Bombus pratorum			1	1	0.05	LC

Species	Filia	Merești	Vârghiș	Sum	Dominance	IUCN
Bombus ruderarius	3	3	12	18	0.96	LC
Bombus subterraneus		3		3	0.16	LC
Bombus sylvarum	2	4	16	22	1.17	LC
Bombus terrestris	108	274	329	711	37.8	LC
Camptopoeum friesei		1		1	0.05	LC
Ceratina cyanea	1			1	0.05	LC
Chelostoma campanularum		1		1	0.05	LC
Chelostoma florisomne	1		5	6	0.32	LC
Coelioxys conoidea			1	1	0.05	LC
Colletes similis	2		1	3	0.16	LC
Dasypoda hirtipes		1		1	0.05	LC
Epeoloides coecutiens			1	1	0.05	LC
Eucera interrupta	1			1	0.05	LC
Eucera longicornis	4	16	15	35	1.86	LC
Eucera nigrescens	14	7	3	24	1.28	LC
Eucera proxima		1		1	0.05	DD
Eucera taurica		1	1	2	0.11	DD
Halictus eurygnathus	6	3	10	19	1.01	LC
Halictus fulvipes	1			1	0.05	LC
Halictus langobardicus	12	15	10	37	1.97	LC
Halictus maculatus	7	2	5	14	0.74	LC
Halictus quadricinctus		1		1	0.05	NT
Halictus rubicundus	4	3	6	13	0.69	LC
Halictus scabiosae	2			2	0.11	LC
Halictus seladonius	2	1	1	4	0.21	LC
Halictus sexcinctus	1	2		3	0.16	LC
Halictus smaragdulus			1	1	0.05	LC
Halictus subauratus	8	3	2	13	0.69	LC
Halictus tumulorum	13	8	12	33	1.75	LC
Hoplitis leucomelana			1	1	0.05	LC
Hoplitis tridentata			1	1	0.05	LC
Hylaeus annularis	2			2	0.11	DD
Hylaeus brevicornis	2	1		3	0.16	LC
Hylaeus communis		1	1	2	0.11	LC

Species	Filia	Merești	Vârghiș	Sum	Dominance	IUCN
Hylaeus confusus	2	2	1	5	0.27	LC
Hylaeus cornutus	1			1	0.05	LC
Hylaeus duckei	1			1	0.05	DD
Hylaeus variegatus	2			2	0.11	LC
Lasioglossum albipes	3	3	1	7	0.37	LC
Lasioglossum calceatum	11	36	12	59	3.14	LC
Lasioglossum convexiusculum			1	1	0.05	NT
Lasioglossum corvinum	3	5		8	0.43	LC
Lasioglossum costulatum		1		1	0.05	NT
Lasioglossum discum	4	8	1	13	0.69	LC
Lasioglossum fulvicorne			1	1	0.05	LC
Lasioglossum glabriusculum	2	2	3	7	0.37	LC
Lasioglossum interruptum	1			1	0.05	LC
Lasioglossum laevigatum		1	1	2	0.11	NT
Lasioglossum laticeps	2	1	3	6	0.32	LC
Lasioglossum lativentre	16	10	7	33	1.75	LC
Lasioglossum leucozonium	2	1	4	7	0.37	LC
Lasioglossum lineare			1	1	0.05	DD
Lasioglossum majus		1	2	3	0.16	NT
Lasioglossum minutulum			1	1	0.05	NT
Lasioglossum morio		2	3	5	0.27	LC
Lasioglossum nitidiusculum	3			3	0.16	LC
Lasioglossum parvulum			1	1	0.05	LC
Lasioglossum pauxillum	9	11	5	25	1.33	LC
Lasioglossum politum	10	2	1	13	0.69	LC
Lasioglossum puncticolle	1	6		7	0.37	LC
Lasioglossum villosulum	3	7		10	0.53	LC
Lasioglossum zonulum	4	2		6	0.32	LC
Megachile centuncularis			1	1	0.05	LC
Megachile ericetorum		1		1	0.05	LC
Megachile flabellipes			5	5	0.27	DD
Megachile lagopoda			1	1	0.05	LC
Megachile maritima			2	2	0.11	DD
Megachile melanopyga			1	1	0.05	LC
Megachile octosignata			1	1	0.05	DD

Species	Filia	Merești	Vârghiș	Sum	Dominance	IUCN
Megachile pilicrus	1		4	5	0.27	DD
Megachile pilidens			3	3	0.16	LC
Megachile pyrenaea	1		1	2	0.11	DD
Megachile versicolor	1		1	2	0.11	DD
Megachile willughbiella	5	2	3	10	0.53	LC
Melitta haemorrhoidalis		2		2	0.11	LC
Melitta leporina	2	2	3	7	0.37	LC
Melitta nigricans	1			1	0.05	LC
Nomada armata		3	1	4	0.21	NT
Nomada bluethgeni	1			1	0.05	LC
Nomada femoralis			1	1	0.05	LC
Nomada marshamella	1			1	0.05	LC
Nomada rhenana	1			1	0.05	NT
Osmia aurulenta	1		1	2	0.11	LC
Osmia caerulescens		1		1	0.05	LC
Osmia leaiana			1	1	0.05	LC
Pseudoanthidium nanum	1			1	0.05	LC
Rophites quinquespinosus	8	5	5	18	0.96	NT
Sphecodes ephippius	1			1	0.05	LC
Sphecodes gibbus	1			1	0.05	LC
Sphecodes puncticeps	1			1	0.05	LC
Tetraloniella alticincta	1			1	0.05	LC
Tetraloniella dentata	3	1		4	0.21	LC
Tetraloniella nana	1			1	0.05	DD
Tetraloniella salicariae	3	1		4	0.21	DD
Trachusa byssina		2	11	13	0.69	LC
Trachusa interrupta			1	1	0.05	EN
Xylocopa valga		2	14	16	0.85	LC