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ORIGINAL ARTICLE

Monitoring ant assemblages of oak wood-pastures. A case study from Eastern Europe

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Abstract

Habitats associated with oak may host high insect diversity, especially saproxylic species, due to the availability of specific microhabitats such as dead wood, rotten logs and woody debris. Among the insects occurring in such habitats, ants play a major role in forest ecosystems as generalist predators. Ant assemblages were investigated using different types of traps. Sampling was carried out in 2015 and 2016, in a wood-pasture located in eastern Transylvania (Romania). Differences between species richness and composition in different types of oak habitats were tested: solitary oak in pasture, oak stand, and the ecotone separating them. Altogether a total of 17 ant species were identified. The three habitats shared an important proportion of the species, yet significant differences between the three habitats in terms of community composition were found. The highest diversity of ant species was recorded in the oak stands, followed by the solitary trees, though differences were not significant. Oak associated habitats can sustain high diversity of ant species and moreover thermophilic specialists thrive. Finally, the abundance of *Lasius brunneus* is highlighted, the species being proposed as a wood-pasture specialist.

Keywords Formicidae · Community ecology · Trapping · Thermophilic ants · *Lasius brunneus*

Introduction

Wood-pastures possess both cultural and ecological significance, can provide favourable micro-habitats for a highly diverse group of organisms (Moga et al. 2009; Hartel et al. 2013; Falk 2014; Öllerer 2014; Hartel and Plieninger 2014; Gallé et al. 2017). These silvopastoral systems occur in a wide range of physiognomies, varying from park-like stands, open grassland areas with scattered trees, to near-natural forests in which grazing has a determinant role (Bergmeier et al. 2010; Hartel and Plieninger 2014). A characteristic feature of these highly threatened ecosystems is represented by scattered, often veteran trees, particularly oaks, with high ecological and conservation value for a great variety of species (Manning et al. 2006; Horák and Rébl 2013; Horák et al. 2014; Sebek et al. 2016; Moga et al. 2016), including ants (Reyes-López et al. 2003; Dolek et al. 2009; Frizzo and Vasconcelos 2013).

Ants are considered ecosystem engineers, being key components of various habitats (Frouz and Jilková 2008; Del Toro et al. 2012). Ants are generalist predators being involved in belowground processes by altering their physical and chemical environment through their effects on plants, microorganisms and other soil organisms (Frouz and Jilková 2008). Moreover, they are important seed dispersers and can influence the carbon and nutrient cycles in soils (reviewed in Del Toro et al. 2012). Nonetheless, they make up as food resource for birds (Dorresteijn et al. 2013) and big carnivores such as bears (Roellig et al. 2014). Thus, ant assemblages play an important ecological role also in open woodlands (Reyes-López et al. 2003; Spitzer et al. 2008; Dolek et al. 2009; Sebek et al. 2016). This is due to the presence of solitary trees in wood-pastures, which have several characteristic features favouring ant, and insect assemblages in general, like

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large canopy, hollows and deadwood exposed to the sun, increasing the diversity of available micro-habitats (Dolek et al. 2009; Horák and Rébl 2013; Horák et al. 2014). High ant abundance occurs in grazed semi-natural grasslands (Dauber et al. 2006), whereas forests typically support lower ant abundances than open areas (Rolstad et al. 2000). Yet, the richness of arthropod assemblages may depend on both canopy and understory strata (Floren and Schmidl 2008; Ulyshen 2011; Sebek et al. 2016) and this may result in differences in community composition and richness.

The scope of the present study was thus to characterize the ant assemblages of oak wood-pasture in a historical region of Europe, known for its extensive wood-pastures threatened by changes in their management (Hartel et al. 2013; Öllerer 2014; Moga et al. 2016). The aim was to study the species composition of oaks located in different areas of the wood-pasture, and to assess the differences between ant assemblages of solitary oak, oak stand and the ecotone in between. Thus, we hypothesized that the presence of oaks in the pasture would enhance the abundance of forest ant species. Furthermore, possible indicator species of oak wood-pastures were searched for.

Material and methods

Study area

The study was carried out in an oak (*Quercus petraea*) wood-pasture in Harghita County (Transylvania, Romania, 46.130273 N, 25.415747 E, alt. 570 m a.s.l.). The ground vegetation in the pasture sites (5% canopy cover) is represented by mesophilous grassland. Besides scattered oaks, the woody vegetation is represented by pears *Pyrus* sp., hawthorn *Crataegus monogyna*. Dog rose *Rosa canina* and young poplar *Populus nigra var. italica*. The wood-pasture is currently used for cattle grazing (Fig. 1). The surrounding oak stands bear the character of Pannonic woods with oak *Q. petraea* and hornbeam *Carpinus betulus* as characteristic tree species, together with beech *Fagus sylvatica*, linden *Tilia* sp. and *Pyrus* sp.

Within the wood-pasture three types of habitats were investigated: solitary oak, oak stand and the ecotone habitat between them. In each type of habitat one oak was investigated (Fig. 1). The selected oak had to be a veteran tree and should present at least a cavity.

Ant sampling and identification

The ants were collected using three different types of traps: Moericke (yellow pan traps) for the canopy assemblages, Barrier traps for the ants dwelling on the trees, and pitfall traps for the ground dwelling assemblages. For each chosen oak, two Moericke traps were installed at a height of at least 2.5 m above the ground, two pitfall traps on the ground, at 2 m distance from its trunk, and one barrier trap (white colour plastic cylinder of 1.5 L volume), close to the trunk cavity. Sampling was carried out twice per month between May–October (10 field campaigns) for two years, 2015–2016, and emptied every two weeks.

The traps were filled with killing and preserving agent (glycol: water, 2: 1 solution). Only 220 from a total of 300 traps were retrieved (73.33% retrieval due to accidental destruction by cattle grazing in the study area). All individuals were identified to species level using the keys of Czechowski et al. (2012) and Seifert (2007). Specimens are kept in the private collection of Ioan Tăuşan. For statistical analyses, only worker ants were considered. Habitat preferences of each ant species were assigned according to Czechowski et al. (2012).

Data analyses

Data from both years of investigation were merged into one data set. Differences between habitats in terms of abundance, species richness, Shannon-Wiener diversity were tested using a factorial ANOVA (the type of trap was considered as random factor). Moreover, the same factorial ANOVA (the type of habitat was considered as random factor) was used to test whether the type of trap had any effect on the different parameters (abundance and diversity). Finally, NMDS ordination (ant species represented by at least five individuals were considered) was carried out for all of sites. Differences in species composition of habitats were tested using a perMANOVA (5000 permutations, Poisson distribution). All statistical analyses were done using R software (R Core Team 2013).

In the statistical analysis the trap where *Lasius brunneus* was found in a huge number of individuals (more than 1000 workers and several tens of queens) was not considered. Even though it is widely known that dominant ant species may affect the species richness and the distribution of other ants, because they can monopolize concentrated food sources, this is not the case for *L. brunneus,* which is rather a timid species, inhabiting tree trunks (especially dead wood) or can be found under the bark (Wilson 1955). These individuals were caught by Moericke trapping in the ecotone habitat, as a colony was close to the trap.

Results

Trapping yielded a total of 2544 individuals. Altogether 17 species were recorded belonging to three subfamilies (Formicinae – 8 species, Myrmicinae – 7 species and Dolichoderinae – 2 species). A detailed list of the species is given in Table 1. Most of the species are common for Romania. However, some arboricolous and thermophilic

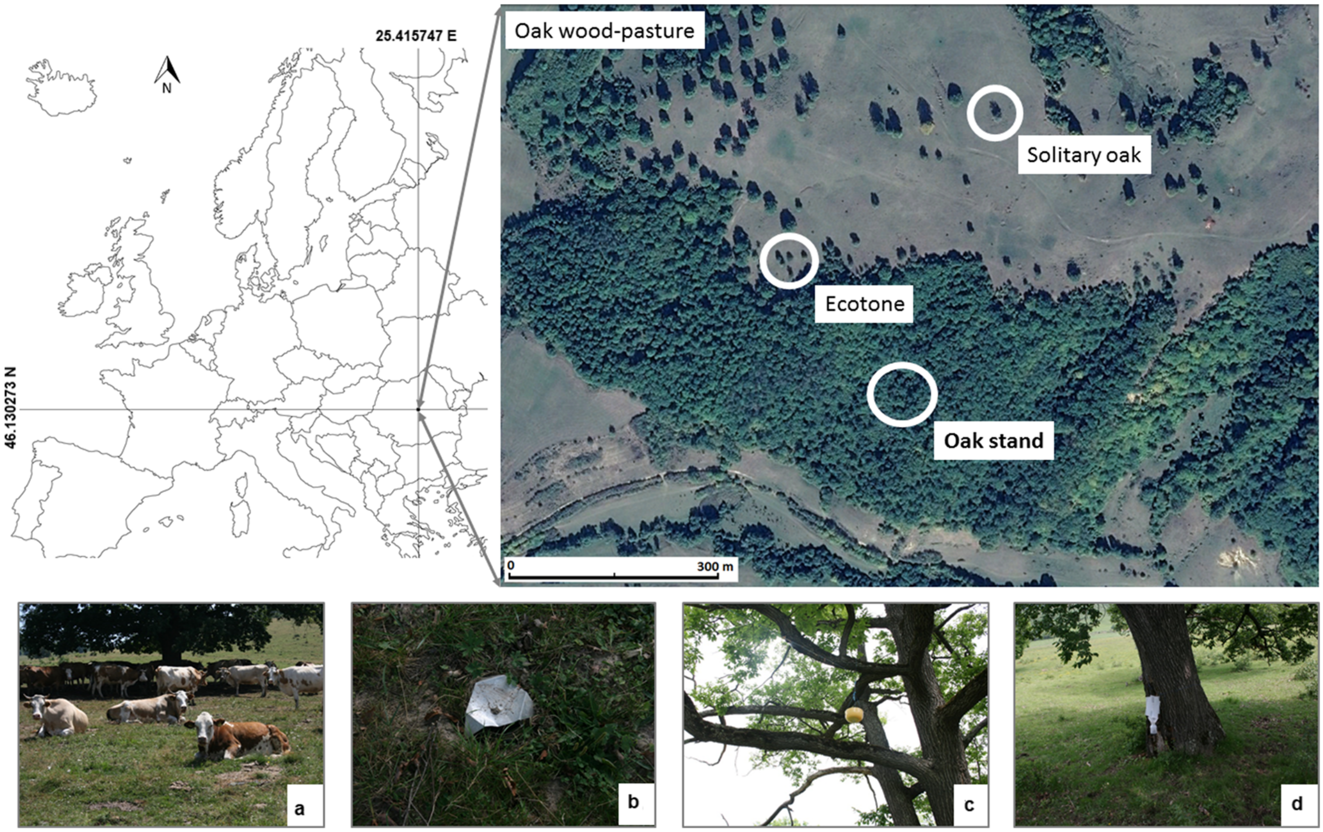

Fig. 1 General view of the oak wood-pasture with the investigated habitats and type of traps. **a** Cattle grazing, **b** pitfall trap, **c** Moericke yellow trap, **d** barrier trap

species were also found, like *Dolichoderus quadripunctatus, Temnothorax affinis* and *Temnothorax corticalis*.

In terms of species abundance, *Lasius brunneus* was found to be abundant in all three habitats. The high abundance of *L. brunneus* in the ecotone habitat is easily explained by the presence of the nest inside the studied oak and thus the Moericke trap collected a large part of the colony, being close to the nest entrance. These samples were however not considered in the analyses. Based on the ecology of *L. brunneus*, and particularly its timid behaviour (Wilson 1955), we can assume that, despite its abundance, the species richness and the distribution of other ant species was not influenced by its presence. The most abundant species in the solitary oak was *Myrmica scabrinodis,* followed by *L. brunneus* and *L. platythorax*. In the case of the oak stand, *Formica fusca* and *L. platythorax* were the most abundant. The most abundant ant species in the ecotone was by far *L. brunneus*, followed by *L. alienus* (Table 1). However, no significant differences (factorial ANOVA, $p = 0.634$, $F = 0.457$, type of trap was considered as random factor) were found in terms of abundance within the three habitats (Fig. 2).

Regarding diversity, there are no significant differences in terms of species number (factorial ANOVA, $p = 0.462$, $F = 0.777$, type of trap as random factor), within the three habitats (Fig. 3). However, the highest number was recorded in the oak stand, whereas the lowest number was recorded in the ecotone habitat. Looking at Shannon-Wiener diversity no significant differences between the habitats (factorial ANOVA, $p = 0.364$, $F = 1.017$, type of trap as random factor) were observed. Nonetheless, similarly as for species richness, the highest value was recorded in the oak stand, followed by the solitary oak habitat.

The only significant differences recorded were in the case of the type of trap. Thus, significant differences in terms of species richness (Fig. 4), Shannon-Wiener diversity and numerical abundance (Fig. 5) were found between the three trapping methods. In all cases, it seems that pitfall trapping yielded the highest values.

Analysing the overall community composition, the three habitats shared an important proportion of species, yet significant differences between the three habitats in terms of community composition (perMANOVA, $p < 0.05$, 5000 permutations) were recorded. Mainly, species such as *L. brunneus, L. alienus*, *Camponotus ligniperda* and *Tetramorium* cf. *caespitum* were abundant in the ecotone, whereas *L. platythorax, Formica cunicularia* and *D. quadripunctatus* were abundant in the solitary oak habitat. The oak stand was characterized by the presence of *Camponotus aethiops* and *F. fusca* (Fig. 6).

Table 1 List of captured ant species and habitat requirements according to Czechowski et al. (2012)

Habitat type	Solitary oak			Ecotone			Oak stand			Characteristic habitat
Type of traps	PB	B	M	PB	B	M	PB	B	M	
Species										
Camponotus aethiops (Latreille, 1798)	–	1	–	1	–	–	14	3	12	OPEN
Camponotus ligniperda (Latreille, 1802)	1	1	–	14	25	3	6	–	–	FOR
Dolichoderus quadripunctatus (Linnaeus, 1771)	2	4	5	–	–	1	1	2	2	FOR
Formica clara Forel, 1886	1	1	7	2	–	–	–	–	–	OPEN
Formica cunicularia Latreille, 1798	12	11	21	6	4	6	10	–	–	OPEN
Formica fusca Linnaeus, 1758	–	–	3	1	–	–	47	5	8	F/O
Lasius alienus (Förster, 1850)	2	–	–	83	8	5	29	1	–	F/O
Lasius brunneus (Latreille, 1798)	21	9	30	–	1668	1	33	–	1	FOR
Lasius platythoax Seifert, 1991	53	–	–	4	–	–	–	–	–	FOR
Myrmecina gramnicola Latreille, 1802	–	–	–	–	–	–	5	–	–	FOR
Myrmica ruginodis Nylander, 1846	3	–	–	–	–	–	18	–	–	FOR
Myrmica scabrinodis Nylander, 1846	96	1	–	99	1	2	60	–	–	F/O
Tapinoma erraticum (Latreille, 1798)	–	–	–	–	–	–	3	–	–	OPEN
Temnothorax affinis (Mayr, 1885)	–	–	–	–	2	–	–	–	–	FOR
Temnothorax corticalis (Schenck, 1852)	–	–	–	–	–	–	–	–	1	FOR
Temnothorax crassispinus (Karavaiev, 1926)	1	–	–	2	1	1	11	–	–	FOR
Tetramorium cf. *caespitum*	1	–	1	5	1	–	37	–	1	UBIQ
Total no. of individuals/type of trap	193	28	67	217	1710	19	274	11	25	
Total no. of individuals	288			1946			310			
Total no. of species/type of trap	11	7	6	10	8	7	13	4	6	
Total no. of species	13			12			15			

Codes: *FOR* forest species, *OPEN* open habitat species, *F/O* species occurring in both forest and open habitats, *UBIQ* ubiquitous species – which are found in any type of habitat including anthropogenic ones, *F/O* species occurring in both forest and open habitats, *M* Moericke traps, *PB* pitfall traps, *B* barrier traps

Discussion

Altogether 17 ant species occurred during the field sampling. Most of the species are common, except for *Temnothorax affinis* and *T. corticalis,* which are poorly known in Romania (Markó et al. 2006).

This number is in accordance with other studies conducted in similar habitats. Thus, Dolek et al. (2009) have obtained

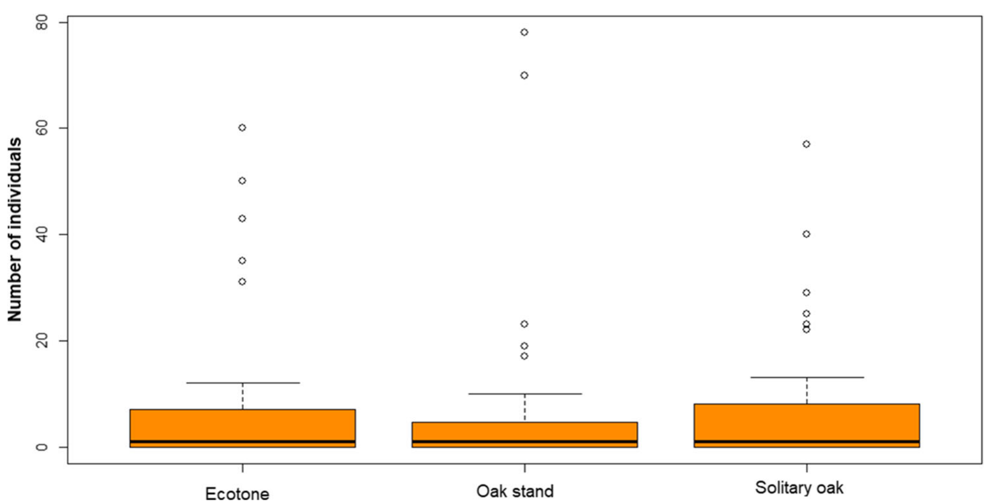

Fig. 2 Box-plot of ant abundance in the investigated habitats (factorial ANOVA, $p = 0.634$, $F = 0.457$)

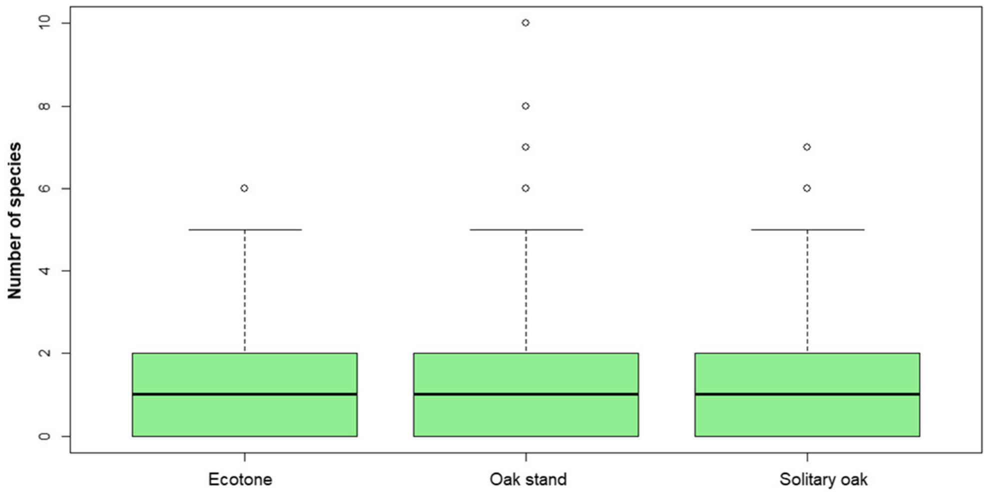

Fig. 3 Box-plot of ant species richness in the investigated habitats (factorial ANOVA, $p = 0.462$, $F = 0.777$)

similar results in Bavaria. They identified 17 ant species, more than half being common with those reported here. Species such as the strict arboricolous *Dolichoderus quadripunctatus, T. affinis* and *T. corticalis* were present in both studies. Sebek et al. (2016) identified 18 species in temperate open woodlands in South Moravia, Czech Republic. Thus, the overall species number is quite accurate and typical for such systems, even though the number of investigated oaks is low, highlighting once again the value of these silvopastoral systems. Despite the inclusion of the oak stand, typical forest species did not prevail. Moreover, species such as *Formica* cf. *rufa* or *Formica sanguinea* and probably some more *Myrmica* spp. and *Lasius* spp. (Buschinger 2004) were not present in the collected samples, underling that wood-pastures are not suitable for forest specialists, at least for ants, in a similar way as reported by Dolek et al. (2009) and Sebek et al. (2016).

Myrmica ruginodis which is the most common forest ant species in Romania (Markó et al. 2006; Tăuşan et al. 2017) was characterized by low abundance in the investigated wood-pasture. Yet in other studies this species was abundant (Müller and Schlumprecht 2004; Dolek et al. 2009). One possible explanation is that the studied oak stand (Fig. 1) is smaller than the forests considered in the other studies, being rather a particular area of the wood-pasture. Thus, the only abundant forest species are *Lasius brunneus* and *L. platythorax* which may occur in open habitats as well (Czechowski et al. 2012). Roellig et al. (2014) reported the typical grassland species *L. flavus, L. niger*, and *Tetramorium* cf. *caespitum* from Transylvanian wood-pastures. The two *Lasius* species were not collected; however, these were found in the pasture, but not close to the oaks, due to their preferences of direct sunlight (Czechowski et al. 2012, Tăuşan et al. 2017).

The results do not concur with findings of other studies, where the highest ant species diversity was recorded in solitary oaks (Dolek et al. 2009; Sebek et al. 2016), whereas the forest stands harboured to lowest number of species (Dolek et al. 2009; Sebek et al. 2016). These differences might be the outcome of the low number of sampled trees compared to the other studies, but also of the fact that in the present study species assemblages of veteran oaks were compared, which might provide similar essential microhabitat conditions notwithstanding their location in open pasture, ecotone habitat or

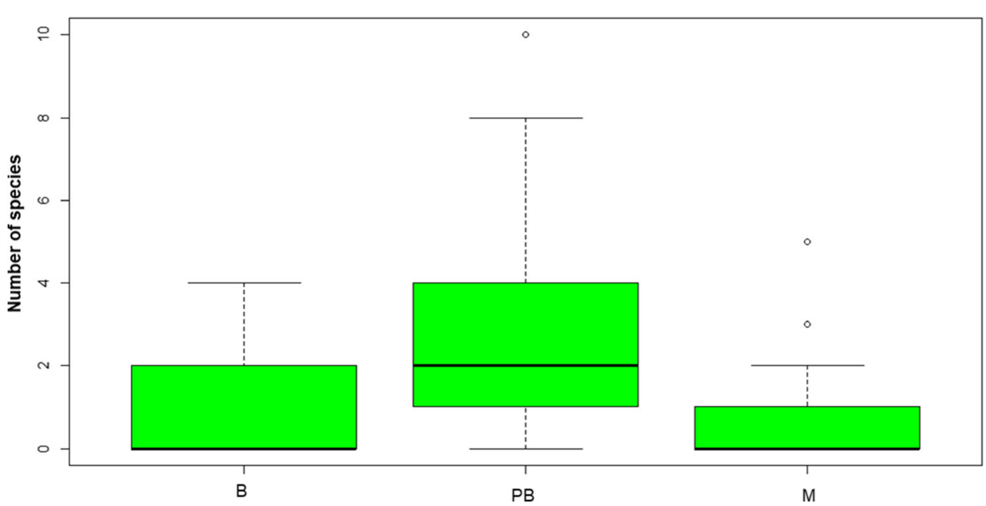

Fig. 4 Compared analysis of species richness based on the type of trap (factorial ANOVA, $p < 0.05$, $F = 23.36$, type of habitat as random factor). B – barrier traps, PB – pitfall traps, M – Moericke traps

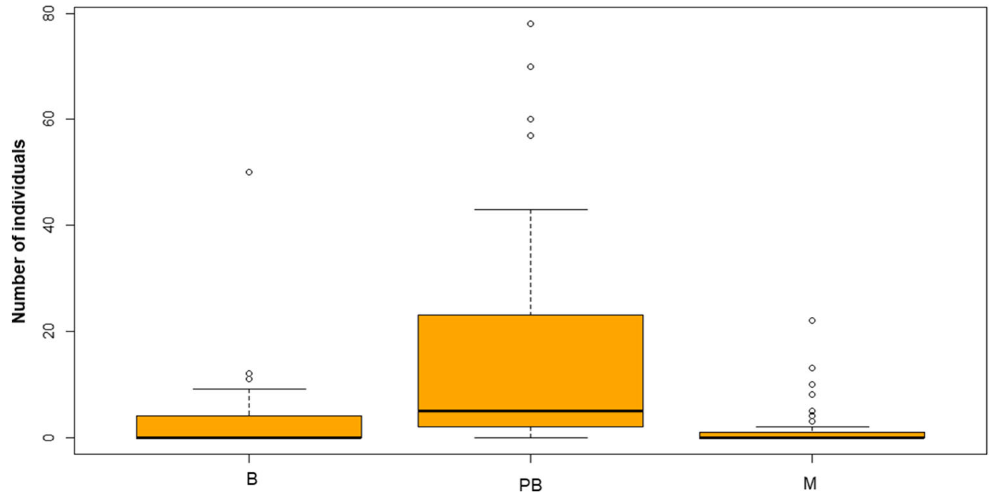

Fig. 5 Compared analysis of numerical abundance based on the type of trap (factoral ANOVA, $p < 0.05$, F = 10.06, type of habitat as random factor). B – barrier traps, PB – pitfall traps, M – Moericke traps

oak stand. This was expected, because most of the ants are ground-dwelling, few of them being tree specialists (*T. corticalis, T. affinis* and *D. quadripunctatus*) (Dolek et al. 2009). Yet, each type of trap contributed to the ant community composition of the wood-pasture.

Sebek et al. (2016) showed that species composition within solitary trees was more like that of trees in edge habitat types than to those of the forest interior. Results indicate clear separation of the habitats despite sharing a high proportion of the species spectrum. This could be due to the availability of microhabitats (e.g. dead-wood, debris) in all types of oak habitats.

The importance of *L. brunneus* is emphasized*,* the species which was the most abundant in the present study and was collected in all habitats and with all three types of traps. Despite being a rather forest species (Seifert 2007; Czechowski et al. 2012), the species prevailed in the open habitat represented by the solitary pasture oak, probably due to the availability of favourable microhabitats provided by the veteran characters of the studied oak. Moreover, the species prevailed in wooded areas even in urban greenery (Czechowski and Ślipiński 2008) and urban areas (Ślipiński et al. 2012), According to Seifert (2018) the species may prefer a wide range of habitats: "from dark forests to isolated trees in agricultural areas or human settlements". Therefore, this species may be considered as a good wood-pasture indicator.

The general pattern of ant communities from the studied oak pasture is that the composition is a combination of open habitat and forest species. There is no dominance from a typical forest specialist, yet species occurring in forest habitats are present. Thus, the above presented results suggest that the overall system is a reservoir of diversity with a mosaic of

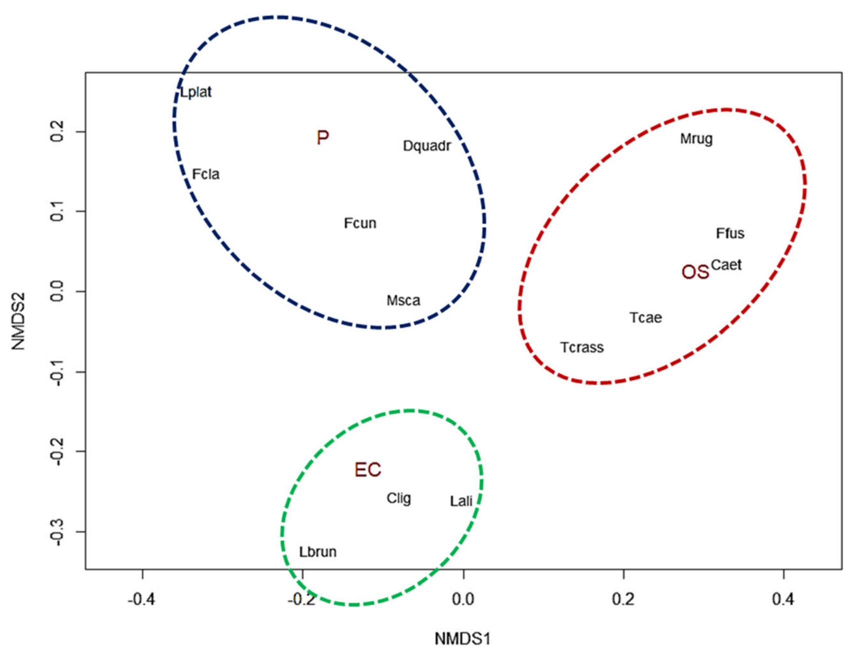

Fig. 6 Non-metric multidimensional ordination (Bray-Curtis index of similarity) of ant communities of oak wood-pasture. P – solitary oak in pasture, OS – oak stand, EC – ecotone; species represented by at least 5 individuals – Stress = 0. Codes: Caet – *Camponotus aethiops*; Clig – *Camponotus ligniperda*; Dquadr – *Dolichoderus quadripunctatus;* Fcla – *Formica clara*; Ffusc – *Formica fusca*; Fcun – *F. cunicularia*; Lali – *Lasius alienus;* Lplat – *Lasius platythorax*; Lbrun– *L. brunneus*; Msca –*M. scabrinodis*; Mrug – *Myrmica ruginodis*; Tcae – *Tetramorium* cf. *caespitum;* Tcrass – *Temnothorax crassispinus*

habitats that sustain species with various ecological demands. Our findings support furthermore the conservation importance of wood-pastures, and thus the need for adequate policies and conservation measures for their maintenance (Hartel and Plieninger 2014; Bobiec et al. 2019).

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval "All applicable international, national, and/or institutional guidelines for the care and use of animals were followed."

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