

## Comparison of two methods for sampling orthopterans in grassland: differences in species representation and sex ratios

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**Abstract:** Orthopterans are convenient indicators for threatened grassland ecosystems. Many sampling methods are known; among them, sweep netting is the most common. This study compares sweep netting with less common pan trapping and quantifies differences in species representation and sex ratio between the two sampling techniques. Sampling took place in the submontane grassland in the northeastern part of the Czech Republic (Central Europe) during July, August, and September 2010. Both sweep netting and pan trapping were used concurrently in 11 meadows. Sampled orthopteran adults were determined to the species level and their sex was noted. Both methods recorded the same pool of 14 species. A chi-squared test showed significant differences in representation of 7 out of 8 analyzed species in sweep-net and pan-trap samples. Sex ratios also noticeably differed. Possible causes of the differences are discussed. This study showed that pan trapping is a solid alternative to sweep netting.

**Key words:** Sweep netting, pan trapping, Orthoptera, grasshopper, relative abundance, species composition, sex ratio, incidence, Czech Republic

### 1. Introduction

Grassland ecosystems constitute one of the most prevalent types of the world's landscape and host a wide range of species (Stoate et al., 2009; Hoste-Danyłow et al., 2010), showing their extensive value for maintaining biodiversity and ecosystem services (Tscharntke et al., 2005; Balvanera et al., 2006). Sadly, a conspicuous decline in grassland biodiversity is observed (Benton et al., 2003; Stoate et al., 2009; Čížek et al., 2012). Therefore, grasslands deserve appropriate monitoring schemes to record, comprehend, and mitigate this trend. The insect order Orthoptera is considered a convenient indicator group for such monitoring (Báldi and Kisbenedek, 1997; Andersen et al., 2001; Kruess and Tscharntke, 2002; Fartmann et al., 2012). Orthopterans are predominantly associated with grasslands (Sergeev, 1998; Marini et al., 2009), comprising there more than half of the total arthropod biomass (Ryszkowski et al., 1993) and representing a substantial component of the food chain, as both consumers (Köhler et al., 1987; Blumer and Diemer, 1996) and prey (Belovski and Slade, 1993; Gardner and Thompson, 1998). Moreover, they are sensitive to changes in agricultural management and to other grassland disturbances (e.g., Andersen et al., 2001; Kruess and Tscharntke, 2002; Čížek et al., 2012).

Plenty of methods for Orthoptera sampling are known (Ingrisch and Köhler, 1998; Gardiner et al., 2005); among

them sweep netting, transect counts, box quadrats or open quadrats (Gardiner and Hill, 2006; Badenhausser et al., 2009), suction sampling (Doxon et al., 2011), acoustic monitoring (Fischer et al., 1997; Lehmann et al., 2014), pitfall traps (Schirmel et al., 2010), or pan traps (Evans and Bailey, 1993) can be mentioned. Sweep netting is a dominant method for sampling Orthoptera (Gardiner et al., 2005). This method is not time and equipment demanding with good efficiency for assessment of relative abundance (Gardiner et al., 2005; Nagy et al., 2007). On the other hand, there can be problems with varying performance under different surveyors or conditions (O'Neill et al., 2002; Gardiner et al., 2005; Whipple et al., 2010). Pan traps (also known as dish traps) are used primarily for the sampling of small flying insects (Moericke, 1951; Duelli et al., 1999). However, they were used also in studies handling with Orthoptera (Köhler and Weipert, 1991; Evans and Bailey, 1993; Nagy et al., 2007). This method is similar to pitfall trapping, which is used more frequently (Gardiner et al., 2005; Schirmel et al., 2010; Schirmel and Buchholz, 2010).

The present study focuses on the comparison of the two sampling techniques: sweep netting (as a dominant method for sampling Orthoptera) and pan trapping (as a less common method). Following an assumption that

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various Orthoptera species have various behaviors, they should respond differently to distinct sampling methods, resulting in a different representation of species in a sample. The same assumption stands for the sex ratio of caught animals because of the distinct behavior of sexes. The aim of this work was therefore to assess and to quantify the differences in species composition, species representation, and sex ratio between the two sampling techniques: sweep netting and pan trapping. Such comparison is useful for the methodology of future field surveys.

## 2. Materials and methods

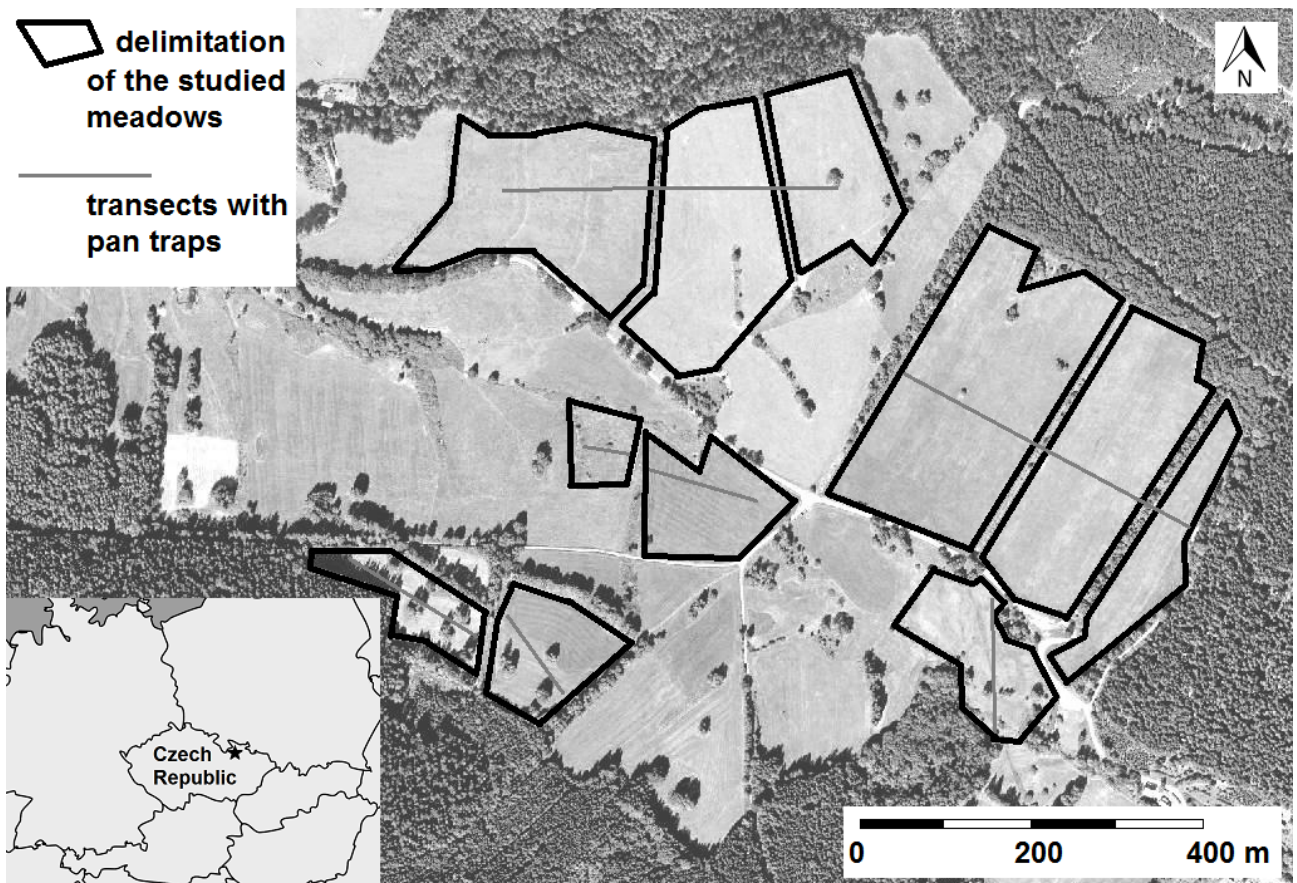
### 2.1. Study area

The survey was performed in the submontane grassland of the Hrubý Jeseník Mts., in the northeastern part of the Czech Republic. The selected grassland area of approximately 180 ha is almost completely surrounded by forest (GPS: 50°6'37.91"N, 17°3'17.48"E; Figure). The altitude is around 780 m above sea level. The mean annual temperature is 6.5 °C and long-term annual average rainfall is 900 mm (Tolasz, 2007). Among this grassland, 11 meadows (average size 3 ha) delimited by natural boundaries (such as

belts of trees, forest edges, or baulks) were selected. Several vegetation types are developed on these seminatural meadows, with the domination of grasses (*Arrhenatherum elatius* (L.) P.Beauv. ex J.Presl & C.Presl., *Festuca rubra* L., *Cynosurus cristatus* L., *Trisetum flavescens* (L.) P.Beauv., *Dactylis glomerata* L.) and with the common occurrence of flowering forbs. There were distinguishable differences in plant species composition between particular meadows but the vegetation of individual meadows was homogeneous. The meadows are managed by a single local farmer, who mows all of them once a year. In the season 2010, when this study took place, the area was mown gradually in six steps between 29 June and 25 August. Moreover, some of the meadows were extensively and temporarily grazed by cattle.

### 2.2. Study design

The sampling took place during July, August, and September 2010 (when the majority of Orthoptera species occur as adults) on 11 meadows within the study area. Both sweep netting and pan trapping were used concurrently on each meadow. Sweep netting was performed at a sufficient distance (at least 8 m) from disposed pan traps in an effort to avoid any cross-method influence.



**Figure.** Location of the study area in the Czech Republic near Nové Losiny village (marked by star), delimitation of the studied meadows and placement of pan-traps transects within them.

Sweep netting was carried out on sampling spots evenly distributed across each meadow. A number of sampling spots for each meadow were derived from an area of the particular meadow (1 spot for 0.5 ha). There were together 66 sampling spots within the 11 sampled meadows. One sampling on a sampling spot comprised a series of 10 sweeps. In the event of low and very low orthopteran numbers obtained, the number of sweeps was increased to 20 or 30, and then such outcomes were divided by 2 or 3, respectively. We used this technique in order to better encompass all species. The diameter of the sweep net was 35 cm. The sweep netting was conducted during three visits (23 July, 15 August, and 19 September 2010), always between 1000 and 1700 (Central European Summer Time). All three visits were carried out in suitable weather conditions (no to mild wind, no rain, temperature 17 °C or higher).

Pan traps were disposed across all meadows in the form of transects (1 transect for each meadow; Figure), totaling 77 traps. The distance between neighboring pan traps was approximately 20 m. The pan traps were yellow plastic bowls 15 cm in diameter and 8 cm deep, half filled with preserving liquid (water solution of sodium chloride enriched with commercial detergent). The traps were placed on the ground and if necessary the immediate surrounding was adjusted to avoid shading from vegetation. Contents of the traps were collected at approximately 10-day intervals (from 25 July to 21 September 2010; 7 collections in total); preserving liquid was refilled at the same time.

Samples obtained from pan trapping were stored in ethanol and consequently determined in the laboratory. Sweep netted orthopterans were either determined directly in the field or stored in ethanol and determined in the laboratory. Only adult orthopterans were included in the analysis because the determination of some nymphs to the

species level was not possible. The nomenclature follows Kočárek et al. (2005). The sex of each adult individual was noted.

### 2.3. Statistical analysis

For the analysis abundant species were selected with incidence higher than 1% (8 species; Table 1). For the evaluation of differences between the ratio of individual species in sweep-net samples and pan-trap samples a chi-squared test was used. Input values were an abundance of particular species in both methods and pooled abundance of the selected 8 species in samples from both methods. Sex ratio was calculated for each species (Table 1) and consequently Wilcoxon's signed rank test was used for pairwise comparisons to analyze an effect of sampling method on the sex ratio. All statistical tests were performed in software R 3.2.3 (R Core Team, 2015).

### 3. Results

Both sampling methods yielded the same pool of 14 Orthoptera species. See Rada et al. (2014) for the complete species list. Here are presented only 8 abundant species usable for the statistical comparison of the two methods (Table 1). The most numerous species were *Omocestus viridulus* and *Gomphocerippus rufus*.

Comparison of species representation within sweep-net and pan-trap samples by chi-squared test showed significant differences in abundance of all species with the exception of only *Chrysochraon dispar* (Table 2). The difference was especially profound in *O. viridulus* (Tables 1 and 2). A distinct divergence of the sex ratio of individual species can be seen in Table 1. There is an evident trend of a higher ratio in pan-trap samples. This trend was proved to be statistically significant (Wilcoxon's signed rank test,  $V = 3$ ,  $P = 0.039$ ).

**Table 1.** The species selected for analysis of differences between sweep netting (SN) and pan trapping (PT). Abundances (with the percentage of total abundance in brackets) and sex ratios (males/females) are given for each species and method.

	SN abun.	PT abun.	SN sex ratio	PT sex ratio
<i>Chorthippus apricarius</i> (Linné, 1758)	7 (2.2%)	36 (4.9%)	0.69	5
<i>Chorthippus biguttulus</i> (Linné, 1758)	64 (19.0%)	75 (10.3%)	0.76	1.42
<i>Chorthippus paralellus</i> (Zetterstedt, 1821)	60 (17.8%)	58 (8.0%)	0.81	2.41
<i>Chrysochraon dispar</i> (Germar, 1834)	32 (9.5%)	86 (11.8%)	0.67	3.3
<i>Euthystira brachyptera</i> (Ocskay 1826)	21 (6.4%)	87 (11.9%)	0.94	5.69
<i>Gomphocerippus rufus</i> (Linné, 1758)	86 (25.6%)	128 (17.6%)	1.16	1.67
<i>Metrioptera roeselii</i> (Hagenbach, 1822)	23 (6.8%)	14 (1.9%)	1.19	0.56
<i>Omocestus viridulus</i> (Linné, 1758)	43 (12.8%)	245 (33.6%)	0.61	1.08

**Table 2.** Comparison of species representation within sweep-net and pan-trap samples by chi-squared test (in all cases d.f. = 1).

	$\chi^2$	P value	
<i>Chorthippus apricarius</i> (Linné, 1758)	4.80	0.028	*
<i>Chorthippus biguttulus</i> (Linné, 1758)	15.71	<0.001	***
<i>Chorthippus paralellus</i> (Zetterstedt, 1821)	23.07	<0.001	***
<i>Chrysochraon dispar</i> (Germar, 1834)	1.17	0.279	n.s.
<i>Euthystira brachyptera</i> (Ocskay 1826)	8.08	0.004	**
<i>Gomphocerippus rufus</i> (Linné, 1758)	9.40	0.002	**
<i>Metrioptera roeselii</i> (Hagenbach, 1822)	16.72	<0.001	***
<i>Omocestus viridulus</i> (Linné, 1758)	50.17	<0.001	***

#### 4. Discussion

The basic requirement for successful sampling is to provide a complete species list. Both methods compared in this study recorded the same species pool. In this respect the unconventional method of pan trapping can be considered as effective as traditional sweep netting. An additional requirement is to provide relative abundances of all species as close as possible to the actual representation of species in the assemblage. Differences in relative abundances can be caused by different mechanics of the two methods, when some species are more susceptible to one of them.

This susceptibility is probably driven by certain aspects of grasshoppers' morphology and behavior. Evans and Bailey (1993) offered an explanation by wing morphology, when flightless grasshopper species should be more prone to be caught in a pan trap because of their inability to avoid landing in the trap. My results do not confirm this explanation. Flightless species *E. brachyptera* and *C. dispar* had a higher incidence in pan-trap samples, but that was also true for macropterous species *O. viridulus* and *C. apricarius*. Moreover, flightless *C. paralellus* had a higher incidence in sweep-net samples. Therefore, susceptibility to being caught by different methods is presumably shaped by an unknown mixture of behavioral and morphological aspects.

Another important factor is the one-shot nature of sweep netting versus permanent pan trapping. While sweeping was done always between 1000 and 1700 in suitable weather, pan traps worked continuously. Variation in activity between individual species during the day (Whipple et al., 2010) or in different weather conditions could result in different variation in obtained relative abundances. For example, the markedly higher incidence of *O. viridulus* in pan traps could be influenced by the capability of

this cold-tolerant species to activate in lower temperatures than other species (see Willott, 1997).

The results indicate a higher sex ratio in pan-trap samples (by some species very markedly). The higher number of males in pan traps could be caused by their higher vagility. By sweep netting, mobile males are more likely to escape. By pan trapping, their vagility increases the probability of falling into the trap. The only bush cricket (suborder Ensifera) within the analyzed species, *M. roeselii*, showed an inverse trend compared to the rest of the species (all of them grasshoppers sensu stricto – suborder Cealifera). Evans and Bailey (1993) presented different results: they obtained more males in sweep-net samples for one grasshopper species and balanced sex ratio for the rest of the species.

This study showed that pan trapping is a solid alternative to sweep netting. Both methods yielded the same species pool of Orthoptera, but with different relative abundances and sex ratios. The results and hypothesized causes of differences are in partial disagreement with a previous comparison (Evans and Bailey, 1993), which was done in North America. It may be a consequence of higher pans used and of diverse species features in Central Europe.

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