

# Effects of plot size on the ordination of vegetation samples

Otýpková, Zdenka<sup>1</sup> & Chytrý, Milan\*

*Institute of Botany and Zoology, Masaryk University, Kotlářská 2, CZ-611 37 Brno, Czech Republic;*  
<sup>1</sup>*E-mail zdenkao@sci.muni.cz; \*Corresponding author; Fax +420 532146213; E-mail chytry@sci.muni.cz*

## Abstract

**Questions:** Do ordination patterns differ when based on vegetation samples recorded in plots of different size? If so, how large is the effect of plot size relative to the effects of data set heterogeneity and of using presence/absence or cover-abundance data? Can we combine plots of different size in a single ordination?

**Methods:** Two homogeneous and two heterogeneous data sets were sampled in Czech forests and grasslands. Cover-abundances of plant species were recorded in series of five or six nested quadrats of increasing size (forest 49–961 m<sup>2</sup>; grassland 1–49 m<sup>2</sup>). Separate ordinations were computed for plots of each size for each data set, using either species presences/absences or cover-abundances recorded on an ordinal scale. Ordination patterns were compared with Procrustean analysis. Also, ordinations of data sets jointly containing plots of different size were calculated; effects of plot size were evaluated using a Monte Carlo test in constrained ordination.

**Results:** The results were consistent between forest and grassland data sets. In homogeneous data sets, the effect of presence/absence vs. cover-abundance was similar to, or larger than, the effect of plot size; for presence/absence data the differences between ordinations of differently sized plots were smaller than for cover-abundance data. In heterogeneous data sets, the effect of plot size was larger than the effect of presence-absence vs. cover-abundance. The plots of smaller size ( $\leq 100$  m<sup>2</sup> in forests,  $\leq 4$  m<sup>2</sup> in grasslands) yielded the most deviating ordination patterns. Joint ordinations of differently sized plots mostly did not yield patterns that would be artifacts of different plot size, except for plots from the homogeneous data sets that differed in size by a factor of four or higher.

**Conclusions:** Variation in plot size does influence ordination patterns. Smaller plots tend to produce less stable ordination patterns, especially in data sets with low  $\beta$ -diversity and species cover-abundances. Data sets containing samples from plots of different sizes can be used for ordination if they represent vegetation with large  $\beta$ -diversity. However, if data sets are homogeneous, i.e. with low  $\beta$ -diversity, the differences in plot sizes should not be very large, in order to avoid the danger of plot size differences distorting the real vegetation differentiation in ordination patterns.

**Keywords:**  $\beta$ -diversity; Cover-abundance data; Data set heterogeneity; Detrended Correspondence Analysis; Plant community; Presence/absence data; Principal Components Analysis.

## Introduction

Ordinations of vegetation samples are commonly used to reveal and visualize variation patterns in plant or animal communities (Jongman et al. 1995; Podani 2000; McCune et al. 2002). In plant communities, species composition is commonly sampled in plots, mostly of quadratic or rectangular shape (Whittaker 1973; Kent & Coker 1992). The appropriate size for plots has been frequently debated in vegetation science. Attempts to find the ‘correct’ plot size by determining the minimal area, i.e. the smallest area that contains the species of regular occurrence in the stand (Moravec 1973; Barkman 1989), did not bring satisfactory results. Today it is commonly accepted that vegetation can be studied in plots of any size, and the choice of a specific size determines the resolution (grain) of the study and affects its results (Wiens 1989; Levin 1992). The appropriate plot size should be thus chosen with respect to the aims of the study. Økland (1990) considered the selection of plot size as being a compromise between homogeneity, suggesting small plots, and representativity, suggesting large plots. In terms of homogeneity, the plot should be so small that it comprises little variation along the most finely grained gradient of interest, relative to the total variation along this gradient. In terms of representativity, it should be so large that it enables reliable prediction of site conditions from the species composition.

In studies of vegetation pattern at the landscape scale, plot sizes usually range from 1 m<sup>2</sup> to hundreds of m<sup>2</sup> (Chytrý & Otýpková 2003). Meta-analyses such as ordinations or classifications based on data sets compiled from different sources may be strongly affected by the large variation in plot size existing among, and often also within, individual studies. Effects of varying plot size on the results of such analyses are insufficiently known, while there is a growing need of understanding these effects now, when large databases, containing plots from heterogeneous sources and of different sizes, are being compiled and analysed (Hennekens & Schaminée 2001).

It has been demonstrated that data sets based on smaller plots may show weaker relations between variation in vegetation and environment (Reed et al. 1993),

different interspecific associations (Jonsson & Moen 1998), higher  $\beta$ -diversity estimated from floristic similarity between plots (Dietvorst et al. 1982), from the length of ordination axes (Økland et al. 1990; Jonsson & Moen 1998) or from Gaussian response curves (Palmer & Dixon 1990), higher evenness (Wilson et al. 1999), lower efficiency of parameter estimation (Kenkel & Podani 1991) or shifted boundaries of vegetation types (Fortin 1999). In the context of ordination, the effect of plot size can interact with the  $\beta$ -diversity of the data sets analysed, i.e. whether they originate from a homogeneous or heterogeneous environment (van Groenewoud 1992). It is also important to know the importance of the plot size effect relative to other options that are known to influence ordination patterns, e.g. the use of either presence/absence or cover-abundance data (Jensén 1970; van der Maarel 1979; Allen et al. 1984; Kovář & Lepš 1986; Vermeersch et al. 2003).

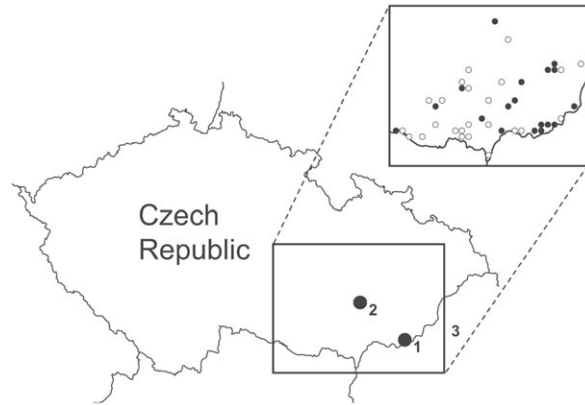
The aim of this paper is to analyse whether differences in plot size influence ordination results, and if so, whether the effect of plot size is (1) more pronounced in homogeneous or heterogeneous data sets; (2) stronger than the effect of using either presence/absence or cover-abundance data; (3) consistent between forest and grassland vegetation data sets. Using field data from forests and grasslands, we will also demonstrate the patterns yielded by ordinations jointly involving data from plots of different size.

## Methods

### Field sampling

The data sets for the analysis of the effect of plot size on ordination were obtained by field sampling of deciduous forests and semi-natural grasslands in southern Moravia (SE Czech Republic). Each of these two vegetation types was sampled separately on a local and a regional scale in order to obtain homogeneous and heterogeneous data sets, respectively (Fig. 1). Four data sets were prepared:

1. A homogeneous data set from forests sampled in the central part of the Bílé Karpaty Mts. (White Carpathians; 48°53' - 48°56' N, 17°31' - 17°48' E) in beech and oak-hornbeam forests of the phytosociological alliances *Fagion sylvaticae* and *Carpinion betuli*;
2. A homogeneous data set from grasslands sampled in nature reserves between the towns of Rousínov, Slavkov and Bučovice (east of Brno, 49°07' - 49°12' N, 16°55' - 16°58' E) in species-rich dry grasslands dominated by *Carex humilis* and *Brachypodium pinnatum*, belonging to the phytosociological alliances *Cirsio-Brachypodium pinnati* and *Festucion valesiacae*;



**Fig. 1.** Sampling sites in southern Moravia (Czech Republic). 1. Homogeneous data set from forests in the White Carpathians; 2. Homogeneous data set from grasslands between the towns of Rousínov, Slavkov and Bučovice; 3. Heterogeneous data sets from forests (full circles) and grasslands (empty circles).

3. A heterogeneous data set from forests sampled at different sites across southern Moravia including different vegetation types of broad-leaved deciduous forests belonging to the phytosociological classes *Querceto-Fageteta* and *Querceteta robori-petraeae*;
4. A heterogeneous data set from grasslands sampled across a wider geographical area and different habitats in southern Moravia, focusing on meadows belonging to phytosociological classes of meadows (*Molinio-Arrhenatheretea*) and dry grasslands (*Festuco-Brometea*).

Sampling sites at the local scale (homogeneous data sets) were selected in a preferential (subjective) way in floristically homogeneous vegetation that developed in similar habitats. Although preferential sampling is generally not recommended for studies of vegetation patterns, it does not cause any problems in the current study, because it is not aimed at description of vegetation pattern nor is it aimed at estimation of vegetation attributes in the study area.

Selection of sampling sites at the regional scale (heterogeneous data sets) was done using *a priori* sampling plans, which were based on the analysis of existing vegetation-plot data from the study area, available in the Czech National Phytosociological Database (Chytrý & Rafajová 2003). All vegetation plots of forests and grasslands from southern Moravia available in the database were classified using TWINSpan (Hill 1979), resulting in 47 clusters for forests and 64 for grasslands. Small clusters containing less than four samples and very heterogeneous clusters were omitted. The remaining clusters were assumed to represent all the major vegetation types of forests and grasslands in the study area. From each of these remaining clusters, one plot

was randomly selected, and the site of this plot was visited and sampled.

At each site a series of nested square plots of increasing size with a common corner was sampled. Nesting was used because the analyses required that the plots of different size were located at the same site, so the differences in species composition between plots resulted solely from the differences in plot size, and not from the habitat differences between sites. The range of plot sizes used was selected so as to include the sizes commonly used in European phytosociology for the sampling of forests and grasslands (Chytrý & Otýpková 2003). The plot sizes used for sampling of forests were 49, 100, 225, 400, 625 and 961 m<sup>2</sup> and the sizes used for grasslands were 1, 4, 16, 25 and 49 m<sup>2</sup>. In the plots of each size, all vascular plants were recorded and their cover was estimated using the nine-degree Braun-Blanquet scale of abundance and dominance (Westhoff & van der Maarel 1973).

The numbers of sampled sites and the total numbers of plots of all sizes, respectively, were as follows:

homogeneous data sets: forests 32 and 192, grasslands 33 and 165;  
heterogeneous sets: forests 23 and 138, grasslands 30 and 150.

To quantify the heterogeneity of the four data sets, we calculated gradient lengths in standard deviation units on the first axis of DCA, detrended correspondence analysis (CANOCO 4.5 program; ter Braak & Šmilauer 2002) for the set of plots of each size, using species cover-abundance in ordinal values. The ranges of these gradient lengths between the ordinations with smallest and largest plots were as follows:

homogeneous sets: forests 4.6 - 3.5, grasslands 3.2 - 2.4;  
heterogeneous sets: forests 8.4 - 4.8, grasslands 9.6 - 6.7.

The corresponding ranges of the mean number of vascular plant species per plot were:

homogeneous sets: forests 22.2 - 46.9, grasslands 28.3 - 62.8;  
heterogeneous sets: forests 27.6 - 55.6, grasslands 25.5 - 58.5.

### *Data analysis*

Data editing was done with the TURBOVEG 2.0 (Hennekens & Schaminée 2001) and JUICE 6.3 (Tichý 2002) programs. Species records from different vegetation layers in the same plot were merged for the analyses so that each species was included only once in the analysis. The effect of plot size on the ordination of vegetation samples was evaluated by comparing separate ordinations of the plots of the same size, performed for each of the four data sets. Two variants of each data set were used in the analyses, one with species presences/absences and the other with cover-

abundances expressed on an ordinal scale from 1 to 9, which replaced the original alpha-numeric values of the Braun-Blanquet nine-degree scale. This enabled comparison of the effect of plot size with the effect of data set heterogeneity and with the effect of using either presence/absence or cover-abundance data. It is commonly accepted in ecological ordination studies that homogeneous data sets should be preferably analysed with methods based on the model of linear response of species to environment, while heterogeneous data sets with methods based on the model of unimodal response (Jongman et al. 1995). Therefore the homogeneous data sets in this study were analysed with principal components analysis (PCA) on covariance matrix and heterogeneous data sets with detrended correspondence analysis (DCA). The CANOCO 4.5 program (ter Braak & Šmilauer 2002) was used for the PCA and DCA computations.

The resulting separate ordinations of plots of the same size were compared within each of the four data sets with Procrustean analysis, which attempts to match different ordination patterns through their rotation and dilatation. This analysis was calculated using the PROTEST program (Jackson 1995; Peres-Neto & Jackson 2000; [www.zoo.utoronto.ca/jackson/pro1.html](http://www.zoo.utoronto.ca/jackson/pro1.html)). Procrustean analysis provides a goodness-of-fit statistic  $m_{12}$ , describing the degree of concordance between two ordinations. The values of the  $m_{12}$  statistic, which increase with the dissimilarity of ordinations, were used to create a distance matrix between all pairs of ordinations within each of the four data sets. This matrix was used as the input for principal coordinates analysis (PCoA), computed using the CANOCO 4.5 program which visualized the pattern of dissimilarity between ordinations of plots of different size within each of the four data sets.

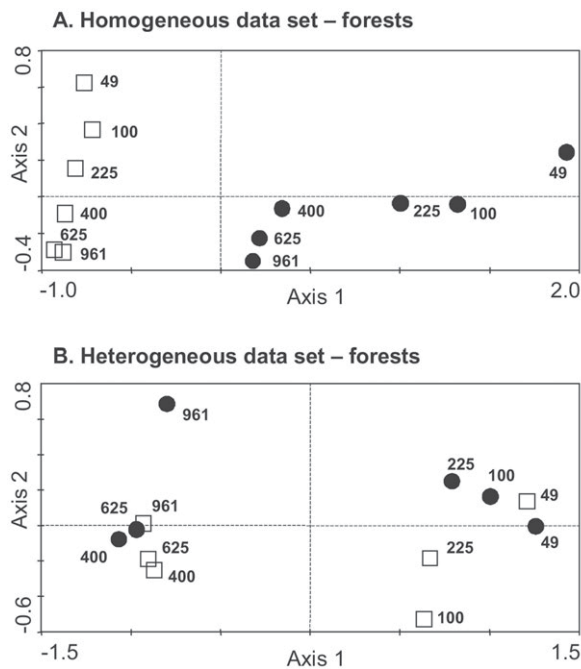
To test how ordinations can be influenced by combining samples from plots of different size in a single input data set, we performed PCA and DCA ordinations of data sets containing plots of two different sizes from each site. In addition, constrained ordinations of these combined data sets were performed with redundancy analysis (RDA) and canonical correspondence analysis (CCA) for the homogeneous and heterogeneous data sets, respectively. Plot size was used as a single explanatory variable in constrained ordinations and its effect on the ordination pattern was tested using the Monte Carlo test with 10 000 permutations. Constrained ordinations and Monte Carlo tests were computed using the CANOCO 4.5 program.

**Results**

*Comparison of separate ordinations of samples from plots of different size*

Differences among ordinations due to different plot sizes and due to using either presence/absence or cover-abundance data are summarized in Figs. 2 and 3. Each individual ordination is represented by a single point in the PCoA ordination diagrams: the more similar the ordinations, the closer the points are situated towards each other in these diagrams.

For the homogeneous data set from forests (Fig. 2A), cover transformations had a stronger effect on ordination results than plot size, as evident from the separated positions of ordinations based on either presence/absence or cover-abundance data. However, plot size also considerably affected ordination results: ordinations of larger plots (400 - 961 m<sup>2</sup>) yielded similar patterns if performed with the same cover transformation, but ordinations of smaller plots (49 m<sup>2</sup>, and less so for 100 and 225 m<sup>2</sup>) resulted in deviating patterns, especially in combination with cover-abundance data.

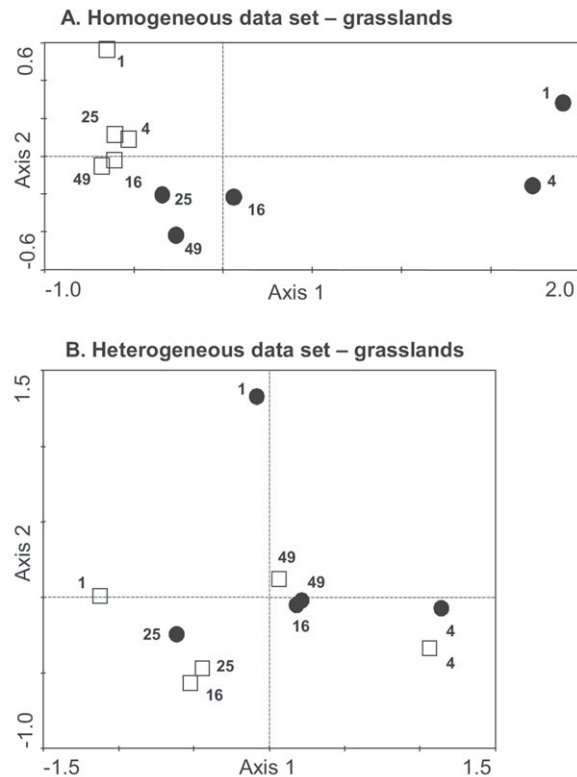


**Fig. 2.** PCoA diagrams showing the differences between separate ordinations of plots of the same size of forest vegetation (PCAs in homogeneous data set and DCAs in heterogeneous data set). Each symbol represents one separate ordination. □ = ordinations of presence/absence data; ● = ordinations of cover-abundance data. Numbers next to symbols indicate plot sizes in m<sup>2</sup>.

The differences due to plot size were smaller if the ordination was based on the presence/absence rather than cover-abundance data.

Similar trends were found for the homogeneous data set from grasslands (Fig. 3A), where the differences between ordinations based on either presence/absence or cover-abundance data were also important. However, for larger plots (16 - 49 m<sup>2</sup>) these differences were not as striking as in the forests. By contrast, ordinations based on small plots of 1 and 4 m<sup>2</sup> and cover-abundance data strongly differed from all the other ordinations. Ordinations of 1-m<sup>2</sup> plots with presence/absence data also produced deviating results. Similarly as for the homogeneous data set from forests, the effect of plot size was smaller with presence/absence than cover-abundance data.

Ordinations of the heterogeneous data sets were more affected by the plot sizes than the use of either presence/absence or cover-abundance data. For forest data sets (Fig. 2B), smaller plot sizes (49 - 225 m<sup>2</sup>) produced similar ordination patterns irrespective of the



**Fig. 3.** PCoA diagrams showing the differences between separate ordinations of plots of the same size of grassland vegetation (PCAs in homogeneous data set and DCAs in heterogeneous data set). Each symbol represents one separate ordination. □ = ordinations of presence/absence data; ● = ordinations of cover-abundance data. Numbers next to symbols indicate plot sizes in m<sup>2</sup>.

**Table 1.** Monte Carlo tests of the effects of difference in plot size on ordination patterns, performed for ordinations that jointly included plots of two sizes. Tests were carried out separately for homogeneous (with RDA) and heterogeneous (with CCA) data sets, forests and grasslands, and with presence/absence and cover-abundance data. The *F*-statistic of the Monte Carlo tests and significance levels (*P*) are shown; n.s. = non significant.

Plot size (m <sup>2</sup> )	Homogeneous data set				Heterogeneous data set			
	presence/absence		cover-abundance		presence/absence		cover-abundance	
	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>
<b>Forests</b>								
49 & 100	0.34	n.s.	0.24	n.s.	0.22	n.s.	0.17	n.s.
49 & 225	1.22	n.s.	0.65	n.s.	0.37	n.s.	0.28	n.s.
49 & 400	2.37	0.001	1.18	n.s.	0.45	n.s.	0.37	n.s.
49 & 625	3.51	<0.001	1.70	n.s.	0.52	n.s.	0.41	n.s.
49 & 961	4.74	<0.001	2.25	0.008	0.57	n.s.	0.46	n.s.
100 & 225	0.42	n.s.	0.22	n.s.	0.21	n.s.	0.18	n.s.
100 & 400	1.18	n.s.	0.54	n.s.	0.32	n.s.	0.27	n.s.
100 & 625	2.02	0.008	0.93	n.s.	0.40	n.s.	0.32	n.s.
100 & 961	3.03	<0.001	1.35	n.s.	0.45	n.s.	0.37	n.s.
225 & 400	0.35	n.s.	0.15	n.s.	0.13	n.s.	0.11	n.s.
225 & 625	0.84	n.s.	0.37	n.s.	0.22	n.s.	0.18	n.s.
225 & 961	1.54	0.045	0.67	n.s.	0.27	n.s.	0.23	n.s.
400 & 625	0.17	n.s.	0.10	n.s.	0.11	n.s.	0.10	n.s.
400 & 961	0.54	n.s.	0.25	n.s.	0.18	n.s.	0.15	n.s.
625 & 961	0.20	n.s.	0.10	n.s.	0.12	n.s.	0.08	n.s.
<b>Grasslands</b>								
1 & 4	1.08	n.s.	0.73	n.s.	0.25	n.s.	0.20	n.s.
1 & 16	3.04	<0.001	1.95	0.007	0.41	n.s.	0.68	n.s.
1 & 25	3.99	<0.001	2.50	<0.001	0.46	n.s.	0.41	n.s.
1 & 49	5.59	<0.001	3.62	<0.001	0.54	n.s.	0.48	n.s.
4 & 16	0.76	n.s.	0.45	n.s.	0.23	n.s.	0.23	n.s.
4 & 25	1.27	n.s.	0.72	n.s.	0.30	n.s.	0.27	n.s.
4 & 49	2.31	<0.001	1.42	n.s.	0.40	n.s.	0.36	n.s.
16 & 25	0.15	n.s.	0.09	n.s.	0.21	n.s.	0.10	n.s.
16 & 49	0.64	n.s.	0.42	n.s.	0.13	n.s.	0.18	n.s.
25 & 49	0.28	n.s.	0.23	n.s.	0.11	n.s.	0.12	n.s.

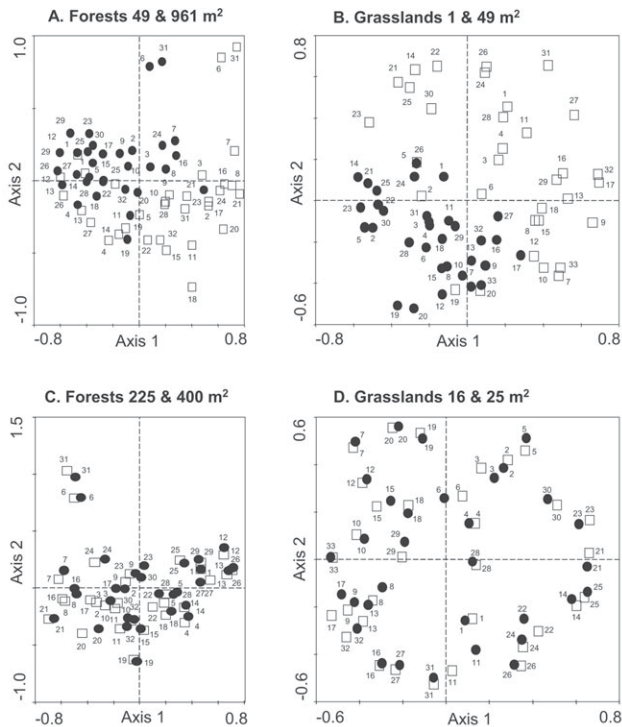
cover transformation, while another group of mutually similar ordinations resulted from the analyses based on larger plot sizes (400 - 961 m<sup>2</sup>). Similarly as in the homogeneous data sets, the ordinations based on large plots were generally more similar to each other than those based on small plots (except for the largest size of 961 m<sup>2</sup> in combination with cover-abundance data).

For the heterogeneous data set from grasslands (Fig. 3B), differences in ordination patterns were also mainly due to plot size and much less due to the use of either presence/absence or cover-abundance data. Ordinations of plots of the same size usually yielded very similar results for both presence/absence and cover-abundance data sets, with the exception of the smallest plots of 1 m<sup>2</sup>, and partly of the plots of 16 m<sup>2</sup>.

*Joint ordinations of samples from plots of different size*

Monte Carlo tests in RDA and CCA showed that plot size never affected ordination patterns when plots of two different sizes from heterogeneous data sets were jointly used in a single ordination analysis (Table 1). In contrast, combinations of plots differing in size often

significantly affected ordinations of homogeneous data sets. In homogeneous data sets from both forest and grassland, the most striking difference appeared when ordination was performed with presence/absence data. Naturally, the largest effect of plot size was revealed in ordinations that involved plots with largest difference in size (49 and 961 m<sup>2</sup> in forests; 1 and 49 m<sup>2</sup> in grasslands). In the homogeneous data set from forests with presence/absence data, the effect of plot size was significant in most cases in which the larger plots were more than four times larger than the smaller plots. When cover-abundances were used in the same data set, plot size only had a significant effect in the joint ordination of the smallest and the largest plots (49 and 961 m<sup>2</sup>). In the homogeneous data set from grasslands the effect of plot size was significant if the size of plots differed more than ten times (with presence/absence data) or more than fifteen times (with cover-abundance data). Joint ordinations of plots of neighbouring size were never affected by the differences in plot size. Some examples of ordination diagrams from PCA that involved plots with a large and small size difference from the homogeneous data sets and with presence/absence data are shown in Fig. 4.



**Fig. 4.** Four examples of PCA ordinations that combined samples from two remarkably different (**A, B**) and two similar plot sizes (**C, D**), each based on homogeneous data sets and presence/absence data. ● = smaller plots; □ = larger plots. Pairs of plots with the same numbers are from the same sites.

## Discussion

### *Comparison of separate ordinations of samples from plots of different size*

Our analysis suggests that ordination patterns based on different plot sizes are more similar to each other if the plot sizes are larger (Figs. 2 and 3). Ordinations based on the smallest plot sizes (49 m<sup>2</sup> in forests and 1 - 4 m<sup>2</sup> in grasslands) gave the most deviating results, especially in the homogeneous data sets, while ordinations of larger plots (400, 625 and 961 m<sup>2</sup> in forests and 16, 25 and 49 m<sup>2</sup> in grasslands) did not usually differ considerably from each other. This result reflects the well-known observation that floristic similarity between plots increases with increasing plot size, because larger plots contain more species and many of these species occur in several plots (Moravec 1973; Dietvorst et al. 1982). In the context of ordination, Økland et al. (1990) measured gradient lengths on DCA axes for data sets consisting of plots of different size. They demonstrated that data sets from smaller plots yielded longer gradients because of their higher  $\beta$ -diversity, and that interspecific relationships in such data sets are generally

weaker. In data sets of small plots, which are poorer in species than large plots, stochastic variation in occurrence of a certain number of species may result in larger between-plot floristic dissimilarity than in data sets of large (i.e. species-rich) plots. This dissimilarity based on stochastic processes may result in a lower stability of ordination patterns computed with data from small plots. Stochasticity of species composition in small plots can be especially high in vegetation stands that contain species with aggregated spatial pattern, provided that the plot size is smaller or similar to the mean size of species clumps (Kenkel & Podani 1991; Jonsson & Moen 1998).

The effect of this stochasticity on the data analyses seems to be important not only in small plots, but also in homogeneous vegetation, where stochastic variation in species occurrences may be the main source of dissimilarity between otherwise rather similar plots. By contrast, plots in heterogeneous data sets differ considerably in their species composition; therefore the stochastic variation in species occurrence accounts for a minor proportion of between-plot variation, and cannot influence the pair-wise resemblances and ordination patterns to such a large extent as in homogeneous data sets.

Our results clearly show that plot size does influence ordination patterns, but for practical applications it is necessary to assess magnitude of its effects relative to the effects of other choices commonly made in numerical analysis of vegetation data, e.g. the type of cover transformation (van der Maarel 1979). We used two common data types, species presence/absence and ordinal cover-abundance based on the Braun-Blanquet scale. The relative importance of the plot size on the one hand and of using presence/absence or cover-abundance data on the other hand differed between the homogeneous and heterogeneous data sets.

For the homogeneous data sets, both from forests and grasslands, the differences in ordination patterns due to using either presence/absence or cover-abundance data were of a similar or larger magnitude as the differences due to varying plot size (Figs. 2A and 3A). The effect of plot size was smaller when presence/absence data were used. Some authors (Austin & Greig-Smith 1968; Vermeersch et al. 2003) suggested that the dominance of species is of greater importance for data analyses in homogeneous data sets than in heterogeneous data sets, because the former contain many species distributed in most plots of the data set, so the main source of between-site variation is due to variation in cover-abundances of species. Thus the inherently higher stochasticity of species composition of smaller plots in homogeneous data sets can be further amplified by adding cover-abundances, which is another piece of rather stochastic information. This explains why the combination of small plots and

cover-abundance data resulted in the most deviating ordination patterns in homogeneous data sets (Figs. 2A and 3A). This observation provides a lesson for practical data analysis: If a data set of small plots, especially from homogeneous vegetation, is analysed with the aim of revealing more general patterns that are expected to be also partly valid with larger plots, it is better to do such an analysis with presence/absence data.

By contrast, for the heterogeneous data sets, the effect of using presence/absence or cover-abundance data was small relative to the effect of plot size (Figs. 2B and 3B). In this case the limited effect of cover-abundance reflects a more unique species composition of each plot within the heterogeneous data sets, which overrides the effect of variation in cover-abundance values. Interestingly, in the heterogeneous data set of forests, there were two groups of similar ordination patterns, one from plots of 49 - 225 m<sup>2</sup> and the other from plots of 400 - 961 m<sup>2</sup>. This may suggest an existence of two dissimilar patterns found at scales smaller and larger than 300 m<sup>2</sup>, respectively.

#### *Joint ordinations of samples from plots of different size*

Often it is necessary to carry out ordinations or other data analyses with samples from plots of different size, particularly if the data are taken from phytosociological databases originating from many different sources (Ewald 2001; Hennekens & Schaminée 2001; Wisser et al. 2001; Chytrý & Rafajová 2003; Gégout et al. 2005). With such data sets, there is a danger that the resulting ordination patterns will partly reflect the differences in plot size in addition to real vegetation differentiation. Our current analysis suggests that ordination patterns are not sensitive to different plot sizes contained in the input data set, when such a data set includes heterogeneous vegetation with a high  $\beta$ -diversity. In sufficiently heterogeneous data sets, even large differences in plot sizes, such as 49 and 961 m<sup>2</sup> for forests and 1 and 49 m<sup>2</sup> for grasslands do not matter in ordination (Table 1). In homogeneous data sets, small differences in plot size, such as 225 and 625 m<sup>2</sup> for forests or 16 and 49 m<sup>2</sup> for grasslands do not matter either. However, differences of an order of magnitude of one, or in some cases even smaller, may lead to ecologically misleading ordination patterns in which variation due to differences in plot size distorts or overrides the real vegetation differentiation (Table 1, Figs. 4A and 4B). Forbes & Sumina (1999) demonstrated that even in a homogeneous vegetation of arctic tundra, rather large differences in plot size (1 and 25 m<sup>2</sup>) did not have any important effect on the ordination pattern. This may have resulted from the fact that in species-poor tundra even such a large difference in plot size did not lead to a large difference in the number of

species. In contrast, our test with a homogeneous data set of species-rich grasslands showed that the same difference in plot sizes significantly distorted ordination pattern. Forbes & Sumina (1999), however, used five times more small plots than large plots and made their conclusion subjectively, based on a visual observation of ordination patterns.

Joint ordinations of samples from plots of different size seem to be less distorted if they use cover-abundance rather than presence/absence data. This probably reflects the fact that species with high cover-abundance in a large plot usually also have high cover-abundance in a small plot located at the same site and vice versa. By contrast, cover-abundances may be an important source of variation in the between-site comparisons. Thus, calculations with cover-abundance values are likely to provide a smaller contribution to the differences between plot sizes and a larger contribution to the differences between sites.

Our study shows, consistently for forest and grassland vegetation, that for the purposes of ordination, simultaneous analysis of samples from plots of different size is possible in many cases. If the data set is heterogeneous, there is a low risk that these differences will influence the resulting ordination pattern. In homogeneous data sets, however, plot size is much more likely to influence the ordination patterns, and joint analyses of data from plots of very different sizes should be avoided. If the plot size differences are not very large, e.g. the smaller size roughly equals a half or a third of the larger size, their joint ordination still seems to be possible even in homogeneous data sets.

**Acknowledgements.** We appreciated comments on the previous version of the manuscript by Norman Kenkel and two anonymous referees. This study was funded from the GAČR 206/05/0020 and MSM 0021622416 projects.

#### **References**

- Allen, T.F.H., Sadowsky, D.A. & Woodhead, N. 1984. Data transformation as a scaling operation in ordination of plankton. *Vegetatio* 56: 147-160.
- Austin, M.P. & Greig-Smith, P. 1968. The application of quantitative methods to vegetation survey: II. Some methodological problems of data from rain forest. *J. Ecol.* 56: 827-844.
- Barkman, J.J. 1989. A critical evaluation of minimum area concepts. *Vegetatio* 85: 89-104.
- Chytrý, M. & Otýpková, Z. 2003. Plot sizes used for phytosociological sampling of European vegetation. *J. Veg. Sci.* 14: 563-570.
- Chytrý, M. & Rafajová, M. 2003. Czech National Phytosociological Database: basic statistics of the available vegetation-plot data. *Preslia* 75: 1-15.

- Dietvorst, P., van der Maarel, E. & van der Putten, H. 1982. A new approach to the minimum area of plant communities. *Vegetatio* 50: 77-91.
- Ewald, J. 2001. Der Beitrag pflanzensoziologischer Datenbanken zur vegetationsökologischen Forschung. *Ber. R. Tüxen Ges.* 13: 53-69.
- Forbes, B.C. & Sumina, O.I. 1999. Comparative ordination of low arctic vegetation recovering from disturbance: reconciling two contrasting approaches for field data collection. *Arct. Antarct. Alp. Res.* 31: 389-399.
- Fortin, J.-M. 1999. Effects of quadrat size and data measurement on the detection of boundaries. *J. Veg. Sci.* 10: 43-50.
- Gégout, J.-C., Coudun, C., Bailly, G. & Jabiol, B. 2005. EcoPlant: A forest site database linking floristic data with soil and climate variables. *J. Veg. Sci.* 16: 257-260.
- Hennekens, S.M. & Schaminée, J.H.J. 2001. TURBOVEG, a comprehensive data base management system for vegetation data. *J. Veg. Sci.* 12: 589-591.
- Hill, M.O. 1979. TWINSpan – A FORTRAN program for arranging multivariate data in an ordered two-way table by classification of the individuals and attributes. Cornell University, Ithaca, NY.
- Jackson, D.A. 1995. PROTEST: A PROcrustean randomization TEST of community environment concordance. *EcoScience* 2: 297-303.
- Jensén, S. 1970. Influences of transformation of cover values on classification and ordination of lake vegetation. *Vegetatio* 37: 19-31.
- Jongman, R.H.G., ter Braak, C.J.F. & van Tongeren, O.F.R. 1995. *Data analysis in community and landscape ecology*. Cambridge University Press, Cambridge, UK.
- Jonsson, B.G. & Moen, J. 1998. Patterns in species associations in plant communities: the importance of scale. *J. Veg. Sci.* 9: 327-332.
- Kenkel, N.C. & Podani, J. 1991. Plot size and estimation efficiency in plant community studies. *J. Veg. Sci.* 2: 539-544.
- Kent, M. & Coker, P. 1992. *Vegetation description and analysis – a practical approach*. Wiley, New York, NY.
- Kovář, P. & Lepš, J. 1986. Ruderal communities of the railway station Česká Třebová (Eastern Bohemia, Czechoslovakia) – remarks on the application of classical and numerical methods of classification. *Preslia* 58: 141-163.
- Levin, S.A. 1992. The problem of pattern and scale in ecology. *Ecology* 73: 1943-1967.
- McCune, B., Grace, J.B. & Urban, D.L. 2002. *Analysis of ecological communities*. MjM Software Design, Gleneden Beach, OR, US.
- Moravec, J. 1973. The determination of the minimal area of phytocenoses. *Folia Geobot. Phytotax.* 8: 23-47.
- Økland, R.H. 1990. Vegetation ecology: theory, methods and applications with reference to Fennoscandia. *Sommerfeltia, Suppl.* 1: 1-233.
- Økland, R.H., Eilertsen, O. & Økland, T. 1990. On the relationship between sample plot size and beta diversity in boreal coniferous forest. *Vegetatio* 87: 187-192.
- Palmer, M.W. & Dixon, P.M. 1990. Small-scale environmental heterogeneity and the analysis of species distributions along gradients. *J. Veg. Sci.* 1: 57-65.
- Peres-Neto, P.R. & Jackson, D.A. 2000. How well do multivariate data sets match? The advantages of a Procrustean superimposition approach over the Mantel test. *Oecologia* 129: 169-178.
- Podani, J. 2000. *Introduction to the exploration of multivariate biological data*. Backhuys Publishers, Leiden, NL.
- Reed, R.A., Peet, R.K., Palmer, M.W. & White, P.S. 1993. Scale dependence of vegetation-environment correlations: A case study of a North Carolina piedmont woodland. *J. Veg. Sci.* 4: 329-340.
- ter Braak, C.J.F. & Šmilauer, P. 2002. *CANOCO reference manual and CanoDraw for Windows user's guide: software for canonical community ordination*. Version 4.5. Microcomputer Power, Ithaca, NY, US.
- Tichý, L. 2002. JUICE, software for vegetation classification. *J. Veg. Sci.* 13: 451-453.
- van der Maarel, E. 1979. Transformation of cover-abundance values in phytosociology and its effects on community similarity. *Vegetatio* 39: 97-114.
- van Groenewoud, H. 1992. The robustness of correspondence, detrended correspondence, and TWINSpan analysis. *J. Veg. Sci.* 3: 239-246.
- Vermeersch, S., De Genst, W., Vermoesen, F. & Triest, L. 2003. The influence of transformations of an ordinal scale of a floristic gradient, applied on a TWINSpan classification. *Flora* 198: 389-403.
- Westhoff, V. & van der Maarel, E. 1973. The Braun-Blanquet approach. In: Whittaker R.H. (ed.), *Ordination and classification of plant communities*, pp. 617-737. W. Junk, The Hague, NL.
- Whittaker, R.H. (ed.) 1973. *Ordination and classification of plant communities*. W. Junk, The Hague, NL.
- Wiens, J.A. 1989. Spatial scaling in ecology. *Funct. Ecol.* 3: 385-397.
- Wilson, J.B., Steel, J.B., King, W.McG. & Gitay, H. 1999. The effect of spatial scale on evenness. *J. Veg. Sci.* 10: 463-468.
- Wiser, S.K., Bellingham, P.J. & Burrows, L.E. 2001. Managing biodiversity information: development of New Zealand's National Vegetation Survey databank. *N. Z. J. Ecol.* 25: 1-17.

Received 30 September 2005;

Accepted 25 April 2006.

Co-ordinating Editor: N. Kenkel.