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Performance of indicators and the effect of grain size in the discrimination of plant communities for restoration purposes

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Abstract: There is strong pressure to embrace indicators for practical goals such as nature conservation and management and to evaluate the restoration success, but the selection of appropriate indicators is not straightforward. In addition, the grain and the type of data collected and data transformation adopted can influence restoration monitoring results. In this paper, we assessed the effect of changing indicator, grain size (i.e., plot dimension) and data transformation in discriminating different mapped plant communities, relying on vascular plant composition data. We considered flora entities at different taxonomic scales of resolution as indicators and used biological forms such as life forms, growth forms and a combination of the two types, i.e., life and growth forms, as rough plant traits. We also analysed the contribution of species as indicators of the different land cover classes by performing an Indicator Species Analysis. We evaluated the effect of changing indicator (taxonomic resolution, life and growth forms and indicator species), grain size and data transformation using permutational multivariate analysis of variance on cover data expressed in percentages and as simple presence/absence. Our results demonstrated that indicators such as taxonomic resolution and biological forms have partial success in discriminating between plant communities, only for the analysis performed on presence/absence data, and that the effects of changing indicator varied depending on the data transformation used. On the contrary, indicator species are coherently effective and changing the grain size has a moderate influence on their ability to discriminate among the habitat types investigated. Hence, indicator species emerged as a promising tool in restoration monitoring. Although indicators are not supposed to substitute comprehensive surveys of vegetation, their use can help redirect considerable time, resources and expertise to more replication and better sampling design.

Abbreviation: PERMANOVA – Permutational Multivariate Analysis of Variance.

Nomenclature: Pignatti (1982).

Introduction

One of the central research issues in community ecology is to understand the drivers of change in species diversity and composition (Holyoak et al. 2005, Vellend et al. 2008). For this purpose, species surveys are increasing in number and importance; however, total or near-total species richness surveys consume large amounts of time and money and complete measurement is seldom, if ever, used in practice (Nordén et al. 2007). To overcome this shortcoming, the identification of biodiversity indicators is increasingly used to achieve practical goals such as nature conservation, natural resource management, and restoration.

Landres et al. (1988) defined "an indicator species [as] an organism whose characteristics are used as an index of attributes too difficult, inconvenient or expensive to measure for other species or environmental conditions of interest". In the literature, indicator species have a long tradition of use in the assessment and monitoring of ecosystems (Noss 1990), to monitor ecological integrity (Carignan and Villard 2002), in forest monitoring (Lindenmayer et al 2000, Nordén et al. 2007, Zerbe et al. 2007), in conservation biology to verify the use of surrogates of species (Caro et al 1999), and to test the use of subsets of species in biodiversity surveys (Vellend et al. 2008).

In restoration ecology, the above topics lead to a more complex question, which needs to be dealt with in order to develop monitoring programs (Herrick et al. 2006): do the diversity and composition of species at sites A (i.e., reference sites) diverge from sites B and C (i.e., control and restored sites)? In the planning phase of restoration monitoring (Chapman and Underwood 2000, Lake 2001), it is important to map plant communities and test the efficiency of remote information in separating vegetation communities (Acosta et al. 2005, Marignani et al. 2008) in order to correctly define reference sites (areas which have the desired end conditions), control sites (degraded areas that are not being restored) and impact sites (restored sites), and to provide data for replication (Chapman and Underwood 2000). In fact, in a restoration experiment (e.g., a Before-After-Reference-Control-Impact (BARCI) design, Lake 2001), data from the restored sites and reference sites should be compared to control locations (Chapman and Underwood 2000) to unambiguously assess the success of a restoration program.

In this case, we are not interested in knowing the exact number and identity of every species at a given site, but rather how the communities compared can be distinguished from one another, starting from the period before restoration actions. Community response is perhaps the most comprehensive way to understand the effects of restoration (Pueyo et al. 2006, Maccherini et al. 2007) but unfortunately, mostly due to time and economic constraints, there is strong pressure to embrace indicators in the evaluation of restoration success. Hence, in order to establish the use of indicators, we should decide what to monitor and bear in mind that the choice of "management indicator species" (Milledge et al. 1991) or "restoration assemblages" (Lambeck 1997), is not straightforward. We must remember in this selection process that the indicators must be relatively easy and inexpensive to measure; they must have no taxonomic difficulties or measuring uncertainties and need to be sensitive to restoration measures (Lake 2001). Having said this, the type of data collected and data transformation can also have an influence; in fact, transformations are known to affect analyses of multivariate patterns (Legendre and Gallagher 2001) and, despite a loss of information, the reduction of abundance data down to presence/absence may lead to an increase in efficiency (Moore 1974, Anderson et al. 2005).

In addition, despite the fact that sample size in species surveys is generally determined by time constraints (Kenkel and Podani 1991), several papers have confirmed the potential benefits of investing resources to increase the sample size of a study, or another aspect of the study that increases statistical power (Vellend et al. 2008), such as selecting a suitable plot dimension (Block et al. 2001, Marignani et al. 2007).

The relationship between grain size (i.e., plot dimension) and the indicators of differences in plant communities (i.e., reference, control, and impact sites in restoration ecology) has not been investigated in-depth. In this paper, we assessed the effect of changing indicator, plot dimension (grain size) and data transformation in discriminating between different communities. We relied on vascular plant composition data in a heterogeneous environment in Tuscany (central Italy).

Study area

The Lucciola Bella Nature Reserve is located in Tuscany, central Italy (N 43° 02' 00'', E 11° 44' 50'', Datum WGS84) in the Upper Orcia River Valley, which is a graben filled with Pliocene marine sediments. Study area covers 745 ha; Marignani et al. (2007, 2008) provide a detailed description of the nature reserve, its management and trends in grassland overgrowth, while Chiarucci et al. (1995) describe the vegetation of badlands. Shrubs are overgrowing grasslands in the reserve, following the natural vegetation dynamics, and are threatening the conservation of the cultural landscape and plant community, which are included in the Habitat Directive (European Commission, 1992). Therefore, managers of the reserve have launched a study to design a restoration plan to control shrub overgrowth (Marignani et al. 2008).

Material and methods

For the study area, we recognized and mapped four land cover classes on a land cover map produced using an object-oriented technique (Marignani et al. 2008): 1. bare ground with little or no vegetation; 2. sparse and discontinuous herbaceous cover; 3. grassland; and 4. grassland with shrubs.

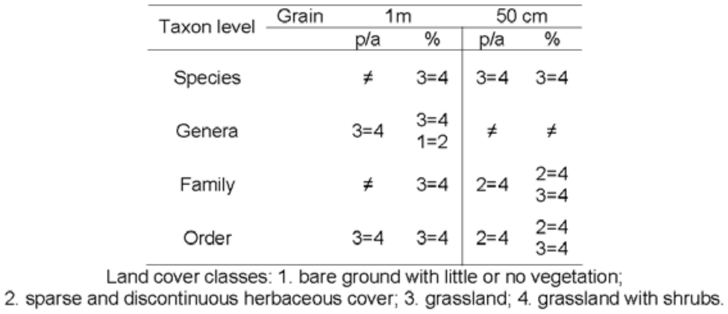
For restoration purposes, we defined as target communities bare ground with little or no vegetation and sparse and discontinuous herbaceous cover, focusing on the land cover classes that constitute the peculiarity of the nature reserve landscape (see Marignani et al. 2008), and undesired communities grassland and grassland with shrubs.

We then conducted balanced stratified random sampling of 64 plots (4 plots × 4 zones × 4 land cover classes), estimating vascular plant cover using a point-quadrat method, with a density of 100 pins/m^2. To assess the effect of grain size, the plots we surveyed were nested squares with side lengths of 0.50 m and 1 m; for herbaceous species we recorded the first species touched by the pins and for shrubs we recorded the first layer. Species data are taken from Marignani et al. (2007).

To assess the effect of changing indicator in discriminating between plant communities, we used two groups of variables. First, we considered the contribution of flora entities at different taxonomic scales of resolution (orders, families, genera and species, Pignatti 1982). Then we used biological forms, such as life forms (i.e., phanerophytes, P, and chamaephytes, Ch, Raunkiaer 1934), growth forms (i.e., scapose, suffruticose or caespitose, Pignatti 1982), and a combination of the two types life and growth forms (i.e., P+caespitose or Ch+suffruticose) as rough plant traits. Using these indicators, we also assessed the effect of using raw cover data expressed in percentages vs simple presence/absence.

In addition to these variables, we also analysed the contribution of species as indicators of the dominating physiognomy characterizing the different land cover classes, by performing an Indicator Species Analysis (Dufrene and Legendre 1997, McCune and Mefford 1999). In this analysis, the results are tested for statistical significance using a Monte Carlo technique: the null hypothesis is that the indicator value observed is no higher than that expected by chance (i.e., that the species has no indicator value, since its presence in the different land cover classes is just as expected by chance). Hence, having obtained the indicator species characteristic of the land cover classes investigated, we tested their efficiency in detecting differences between the four plant communities. We evaluated the effect of changing indicator (taxonomic resolution, life and growth forms and indicator species), grain size and data transformation using per-

Table 1. Permutational multivariate analysis of variance among land cover classes for plant species with presence/absence and cover percentage data, using Bray-Curtis measures at different levels of taxonomic resolution. Probability found for all the general tests was p=0.002. The pairwise tests have been corrected for multiple comparisons (corrected α for the multiple comparisons, α^1=0.008): symbol ≠ indicates that all possible pairwise comparisons between land cover classes are different (p <0.008); symbol = indicates that pairwise comparisons are not different significantly (p >0.008).



Taxon level / Grain	1m		50 cm	
	p/a	%	p/a	%
Species	≠	3=4	3=4	3=4
Genera	3=4	3=4 1=2	≠	≠
Family	≠	3=4	2=4	2=4 3=4
Order	3=4	3=4	2=4	2=4 3=4

Land cover classes: 1. bare ground with little or no vegetation; 2. sparse and discontinuous herbaceous cover; 3. grassland; 4. grassland with shrubs.

mutational multivariate analysis of variance (PERMANOVA, Anderson 2005), by testing the simultaneous response of the variables that describe the composition of the plant communities and comparing the four land cover classes. We used Bray–Curtis measures on cover data expressed in percentages and as simple presence/absence for the analysis.

Since we based all tests and conclusions on the Bray-Curtis measures, to provide for methodological confirmation, we also performed the same analysis using Euclidean distance.

The statistical significance of the multivariate variance components was tested with 499 unrestricted permutations of raw data using correct permutable units (Anderson 2001, 2005, Anderson and ter Braak 2003, Manly 1997, Pillar and Orlóci 1996).

Preliminary analysis found that “zone” did not contribute significantly to variation among land cover classes, therefore we pooled all data within land cover classes to increase our replication ability for detection of the effect of the “land cover class” factor (i.e., 16 plots for each land cover class). We performed separate analyses on data sets constructed for each level of taxonomic resolution (orders, families, genera and species), for the three levels of biological forms (life forms, growth forms, life and growth forms) and for the multivariate response of the plant communities, using a reduced set of species, i.e., only indicator species.

Results

We collected 126 vascular plant species in 64 plots of 1 m^2, the nested squares of 0.25 m^2 accounted for 86 species.

Grain size and taxonomic resolution

For the quadrats of 1 m in length, the 126 species represent 90 genera, 28 families and 23 orders, according to Pignatti (1982). The analyses performed produced similar results (Table 1): using presence/absence data and data in percentages at species, family, genus and order level, grassland and grassland with shrubs could not be consistently distinguished at every taxonomic resolution. At 50 cm length, we identified 87 species, 63 genera, 21 families and 18 orders: at genus level communities appeared as significantly different, while a contrasting characterization emerged at the species and order level on taxonomic investigation (Table 1). At this plot dimension, grassland and grassland with shrubs, and even sparse and discontinuous herbaceous cover and grassland, were impossible to differentiate at family and order level.

At both quadrat sizes, analyses using presence/absence data only emphasized changes in composition and showed an enhanced ability to discriminate between plant communities with decreasing taxonomic resolution.

The analysis performed with Euclidean distances showed different results compared to the one obtained with Bray-Curtis and data in percentages: in particular, using Euclidean distance, the test at order and family level showed that only land cover type 1 (bare ground with little or no vegetation) resulted separated from the other land cover types species' composition at 1 m and 50 cm grain size. On the contrary, using presence/absence data the results are comparable, showing that even though the two metrics are different, using presence/absence data consistency of results has been achieved.

Grain size and life/growth forms

For the quadrats of 1 m in length, the 126 species represent 7 life forms, 10 growth forms and 15 combinations of these categories (Pignatti 1982), while the number of forms decreased slightly for the 87 species collected in the 50 cm quadrats (Table 2). Considering presence/absence data, all communities appeared different for the two quadrat sizes analysed using the biological forms except for sparse and discontinuous herbaceous cover and grassland. Differences emerged only in the analyses performed with data in percentages: analogously to the results obtained with the taxonomic indicators, grassland and grassland with shrubs appeared to be indistinguishable.

The analysis performed with Euclidean distances produced different results compared to the one obtained with

Table 2. Permutational multivariate analysis of variance among land cover classes for plant species with presence/absence and cover percentage data, using Bray-Curtis measures for life and growth forms. Probability found for all the general tests was p = 0.002. The pairwise tests have been corrected for multiple comparisons (α^1=0.008): symbol ≠ indicates that all possible pairwise comparisons between land cover classes are different (p <0.008); symbol = indicates that pairwise comparisons are not different (p >0.008).

Grain	1m			50 cm		
Life and growth forms	Number of forms	p/a	%	Number of forms	p/a	%
Life forms	7	≠	3=4	6	≠	3=4
Growth forms	10	≠	3=4	10	≠	3=4
Life and growth forms	15	≠	3=4	14	2=4	3=4

Land cover classes: 1. bare ground with little or no vegetation; 2. sparse and discontinuous herbaceous cover; 3. grassland; 4. grassland with shrubs.

Table 3. Indicator species for the three land cover classes analyzed, evaluated at different grain sizes using data in percentages.

Life form	Growth form	Families	species	land cover class	50cm	1m
Ch	Suffr	Compositae	*Artemisia cretacea*	2	x	x
T	Scap	Leguminosae	*Melilotus sulcata*	2		x
T	Scap	Poaceae	*Parapholis strigosa*	2	x	x
H	Scap	Compositae	*Podospermum canum*	2	x	x
H	Caesp	Poaceae	*Bromus erectus*	3	x	x
H	Caesp	Poaceae	*Festuca trachyphylla*	3		x
H	Caesp	Poaceae	*Brachypodium rupestre*	4	x	x
T	Scap	Geraniaceae	*Geranium purpureum.*	4		x
NP		Oleaceae	*Ligustrum vulgare.*	4		x
P	Caesp	Rosaceae	*Prunus spinosa.*	4	x	
P	Caesp	Leguminosae	*Spartium junceum.*	4	x	x

Land cover classes: 2. sparse and discontinuous herbaceous cover; 3. grassland; 4. grassland with shrubs.

Bray-Curtis only for the 50 cm dimension, using presence/absence data, at life form and growth form level defining as different from the other land cover classes only class 1 (bare ground with little or no vegetation).

Grain size and indicator species

Due to the fact that the land cover class "bare ground with little or no vegetation" is characterized by a very low species richness (mean 2.13 ±SE 0.786), we considered it useless to perform the indicator species analysis on this class and consequently excluded it from this analysis. Results regarding indicator species for the three land cover classes analyzed showed a high and statistically significant Indicator Value (IV > 25, $p < 0.05$, Table 3), indicating seven species for the 50 cm grain size and 10 species for the 1 m grain size. Increasing the grain size from 50 cm to 1 m quadrat sides, three species were added to the indicator list (*Melilotus sulcata*, *Geranium purpureum* and *Ligustrum vulgare*) and one was excluded (*Prunus spinosa*).

The permutational multivariate analysis of variance on a selected subset of species only (i.e., indicator species), which was performed for each grain size, established the efficiency of indicator species in discriminating between the four land cover types analyzed. At 0.25 m^2 for data in percentages p = 0.002 (with pairwise comparison p = 0.002), except for grassland vs grassland with shrubs, p = 0.02. For presence/absence data, p = 0.002 (with pairwise comparison p = 0.002), except for grassland vs grassland with shrubs, p = 0.02.

At 1 m^2, for data in percentage**s,** p = 0.002, except grassland vs grassland with shrubs, p = 0.02. For presence/absence data, p = 0.002 (with pairwise comparison p = 0.002), except for grassland vs grassland with shrubs p = 0.04.

Discussion

Our results demonstrated that taxonomic resolution and biological forms are partially successful in discriminating between plant communities, but only for the analysis performed on presence/absence data, and that, in any case, the effects of changing indicator varied, depending on the data transformation used. On the contrary, indicator species are effective and changing the grain size moderately influences the ability to discriminate among the habitat types investigated.

Using both taxonomic resolution and biological forms as indicators, the results are habitat dependent: for example, the similarity recognized between grassland and grassland with shrubs for the smaller grain size (0.25 m^2), and even between sparse and discontinuous herbaceous cover and grassland, is mainly linked to grain size and coherent with the physiognomy of plant communities, where scattered patches of hemicryptophytes (e.g., *Bromus erectus* and *Brachypodium* *rupestre, Poaceae*) or phanerophytes (e.g., *Prunus spinosa* and *Spartium junceum*) can locally dominate and create similar local conditions in the three land cover types at a smaller scale.

Selecting indicators *a priori* for restoration is a very controversial issue: in the same heterogeneous environments we observed that, even using the entire community composition, we obtained different results when changing grain and sample size (Marignani et al. 2007). However, analyses performed at different taxonomic resolution and on life and growth forms confirmed the potentiality of rough plant traits (i.e., biological forms) in discriminating between communities.

In establishing indicators, progress in a restoration project may be detected either by an increase in desirable biota or properties or by a decrease in undesirable biota (Lake 2001). In this case study, the undesired communities are grassland and grassland with shrubs, dominated by *Poaceae* and caespitose phanerophytes, while the desirable ones, that represent the peculiarity of the Nature Reserve landscape, are communities of pioneer annual vegetation (i.e., sparse and discontinuous herbaceous cover), mostly composed of miohalophytes and therophytes, closely related to high salt content and shallow soil conditions, which are unfavourable for perennial vegetation such as *Bromus erectus* grasslands (Maccherini et al. 2000, Marignani et al. 2008). In this context, higher taxonomic level analyses were useful because we quickly identified species with distinct morphologies in the communities studied, but we spent a significant amount of time identifying particularly difficult grasses. If we had had the chance of surveying the species at genus level, we could have sampled more sites to detect differences between communities, indirectly influencing our statistical power (i.e., the relevant currency in a research enterprise, Vellend et al. 2008) in a positive way. In fact, since the bulk of species may be identified rapidly in a typical plant community survey, the relatively small number of species that are quite difficult to classify absorb a disproportionate amount of a researcher's time (Colwell and Coddington 1994). More to the point, recent studies affirm that, in many cases, statistical power will not be maximized by attempting to include every last species in a survey (Vellend et al. 2008) and using higher taxa, rather than species, can make it possible to relocate considerable resources and expertise to more replication and better sampling design (Anderson et al. 2005).

In the same way, the selection of a set of indicator species performed *ad hoc* for the study area, based on a preliminary analysis of dominant species, appeared as a positive alternative to complete species lists or to a predefined list such as rare species or red listed species, which are of little interest as monitoring tools (Noss 1999, Cousins and Lindborg 2004), mainly because they might not represent the increase/decrease in desirable characteristics of the reference plant community, and also because they are normally too time-consuming to find. The use of indicator species emerged as a promising tool in restoration monitoring and their use should be tested on both long and short-term time series (Kreyer and Zerbe 2006, Dzwonko and Loster 2007, Zerbe et al. 2007).

Regarding data types, when dealing with composition variability, abundance vs. presence/absence data can lead to completely different results (Cushman and McGarigal 2004, Anderson et al 2005). In our case, using different variables and grain size, presence/absence gave more definite results, suggesting that we could assess the differences between the four plant communities using a high taxonomic level alone (i.e., family level at 1 m^2), while the use of data in percentages consistently confused grassland and grassland with shrubs. Consequently, we could gather only presence/absence data to simply detect differences between communities, but it would reduce the capability of monitoring population fluctuation and the temporal pattern of change in the plant community (Maccherini et al. 2007).

Following a six-step scheme to develop restoration programs (Herrick et al. 2006), we sustain that, to assess the state of plant communities and develop a restoration strategy, it is necessary to start with a complete species survey, without using any indicators. Hence, the results of the preliminary survey must be used to select monitoring indicators, the number of monitoring plots and measurement frequency. For example, in our case study we could collect data on indicator species yearly, focusing on only eleven species, or only investigate the genera/family taxonomic level, or the changes in dominant biological forms in every community, defining a longer interval for the completion of species inventories (e.g., every five years) and assessing the species' responses over time (Dzwonko and Loster 2007).

The approach proposed is not supposed to substitute comprehensive vegetation surveys; nevertheless, applying simple classification of species in the field can be of help in conducting a monitoring survey. In fact, such assessments can be carried out by non-professional botanists or amateurs, who can be trained to learn the indicators selected, permitting continuous data collection that can be integrated and completed by extensive surveys accomplished by specialists in botany (Zerbe et al. 2007).

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