

Appendix S2. Case study

Data

The study site is located close to the summit of Mount Labbro (1193m), on the calcareous massif of the Uccellina-Monte Amiata ridge in Tuscany. The area was intensely grazed until the 1960s. More recently, due to the progressive abandonment of marginal areas, the decreased grazing pressure triggered a secondary succession with distinct stages ranging from semi-natural grasslands to increasingly dense shrublands with *Prunus spinosa*, *Rubus ulmifolius*, and *Cytisus scoparius* (Maccherini et al., 2007).

As part of a Life-Nature project (LIFE NAT/IT/99/6229), the study site was subjected to restoration activities, which included cutting shrubs in overgrown grasslands and sowing native species. Restoration actions took place from late summer 2000 to early spring 2001 (see Maccherini et al., 2018). In 2001, a randomized block design composed of four blocks was used to assess the effect of grazing and sowing on plant diversity. The sowing was carried out in October 2001. The ungrazed plots have been fenced off to exclude livestock. From the start of the experiment, the study site has been grazed mainly by donkeys and horses that were recently reintroduced into the study area.

Four experimental plots of 3m × 5m were established in each block. Each plot was randomly assigned to one of four treatments: no grazing or sowing; sowing without grazing; grazing without sowing; sowing and grazing. Sowing had a very small effect compared to grazing (Maccherini & Santi, 2012; Maccherini et al., 2018). Therefore, only the grazing factor was considered in the present paper. During the project, one ungrazed plot was excluded from the analysis.

In June 2019, we estimated the cover of all vascular plant species within each 1m × 2m subplot at the center of the experimental plots. We used a point quadrat method with a density of 100 pins/m² (Moore & Chapman, 1986). Species present in a plot but not touched by any pin were recorded with an arbitrary cover of 0.5 pins. For the most abundant species in each treatment (8 grazed plots and 7 ungrazed plots), we measured four functional traits (mean of three replicates for each species in each treatment) that are typically associated with the leaf economics spectrum (Wright et al., 2004). The traits include: specific leaf area (SLA, mm²/mg), leaf dry matter content (LDMC, mg/g), and nitrogen and carbon content (N% and C%). Together, the species sampled account for ~70% of the total plant cover in each treatment. Therefore, as suggested by Grime's mass ratio hypothesis, they are expected to make a substantial contribution to community structure and functioning (Grime, 1998).

The selected traits reflect a trade-off between fast-growing acquisitive species versus slow growing, more conservative species (Wright et al., 2004). Specifically, SLA and LDMC are considered soft morpho-anatomical traits correlated with relative growth rate, photosynthetic rate, and nutrient concentration. Higher SLA values are associated with lower leaf span and higher photosynthetic rate. LDMC is related to the density of leaves; it has been demonstrated to scale negatively with potential growth rate and positively with leaf lifespan (Cornelissen et al., 2003). Finally, N% and C% are considered as a proxy of photosynthetic rates and nutrient uptake from the soil. Data on species abundances and functional traits are available in Ricotta et al. (2022, Appendix A) and in the Dryad Digital Repository <https://doi.org/10.5061/dryad.7pvmcvdzc> (Ricotta et al., 2023).

Methods

All trait data were linearly scaled in the range [0,1] by their minimum and maximum values. From the scaled values, the functional Euclidean distances between all pairs of species in both treatments were calculated. The functional distances were then divided by their maximum value in both treatments. The resulting scaled distances were finally used, together with the relative abundances of the species in each plot, to calculate the corresponding Rao's quadratic diversity Q , functional redundancy R , and the Simpson

dominance index D . All calculations were performed with the functions 'speciesdiv' (for the calculation of the Simpson diversity) and 'QE' (for the calculation of Rao's Q) of the R package 'adiv' (Pavoine, 2020).

To generate the ternary diagram of functional diversity, we used the R package 'compositions' (van den Boogaart et al., 2018). We then tested for significant differences in the DRQ ternary composition between the plots of both treatments (grazed and ungrazed) with distance-based multivariate analysis of variance (db-MANOVA) of the R package 'PERMANOVA' (Vicente-Gonzalez & Vicente-Villardón, 2021). This is a multivariate generalization of classical ANOVA used for testing for differences between two or more groups of plots based on every possible dissimilarity measure (Anderson, 2001). The essence of db-MANOVA is to compare the within-group dissimilarities with the between-group dissimilarities. The larger the between-group dissimilarities compared to the within-group dissimilarities, the more likely it is that the plots in both groups differ in their DRQ ternary composition (Anderson, 2001). For this purpose, in order to account for the compositional data structure of the DRQ components whose total sum for each plot is equal to one, we calculated pairwise dissimilarities among plots with the Bray & Curtis (1957) dissimilarity. P-values were obtained by 10000 random permutations of individual plots between the treatments, thus generating a reference distribution under the null hypothesis of no differences in the DRQ ternary composition between the plots in both treatments. For the single diversity measures, D , R , and Q , we further tested for significant differences between both treatments with standard univariate ANOVA and 10000 random permutations of individual plots between the treatments.

Note that from a statistical viewpoint, like for all compositional data carrying relative rather than absolute information with a constant sum constraint, the main disadvantage of ternary diagrams is that the three additive components violate the basic assumption of independence. For example, if Q and R are known, the value of D is automatically obtained due to the constant sum of 1. To enable the multivariate analysis of closed data sets, Aitchison (1986) proposed the application of log-ratio transforms, which have become very popular in many fields of research. For details on compositional data analysis, see van de Boogaart & Tolosana-Delgado (2013) and Greenacre (2021).

Unfortunately, log-ratio transformed data are more difficult to interpret compared to the original diversity values. Since community ecologists are usually familiar with the application and interpretation of distance-based multivariate methods, in this paper, we tested for differences in the functional diversity structure of grazed and ungrazed plots with permutational distance-based multivariate ANOVA and Bray-Curtis dissimilarity. This dissimilarity coefficient is part of a large family of set-theoretical measures extensively used in community ecology to summarize differences in species composition between plots in terms of Venn diagrams (Cross & Sudkamp, 2010; Roberts, 2017; Ricotta et al., 2021). Therefore, at least for exploratory data analysis, we think it is adequate to deal with DRQ compositional data with a constant sum constraint such that the proposed approach may represent a good compromise between statistical soundness and ecological interpretability.

References

- Aitchison, J. (1986). *The Statistical Analysis of Compositional Data*. Chapman and Hall, London.
- Anderson, M.J. (2001). A new method for non-parametric multivariate analysis of variance. *Austral Ecology*, 26, 32–46.
- Bray, J.R., & Curtis, J.T. (1957). An ordination of the upland forest communities of southern Wisconsin. *Ecological Monographs*, 27, 325–349.
- Cornelissen, J.H.C., Lavorel, S., Garnier, E., Díaz, S., Buchmann, N., Gurvich, D.E., Reich, P.B., ter Steege, H., Morgan, H.D., van der Heijden, M.G.A., Pausas, J.G., & Poorter, H. (2003). A handbook of protocols for standardised and easy measurement of plant functional traits worldwide. *Australian Journal of Botany*, 51, 335–380.

- Cross, V.V., & Sudkamp, T.A. (2010). *Similarity And Compatibility In Fuzzy Set Theory*. Physica-Verlag, Heidelberg.
- Greenacre, M. (2021). Compositional Data Analysis. *Annual Review of Statistics and its Application*, 8, 271–299.
- Grime, J.P. (1998). Benefits of plant diversity to ecosystems: immediate, filter and founder effects. *Journal of Ecology*, 86, 902-910.
- Maccherini, S., Bacaro, G., & Marignani, M. (2018). Beneficial effects of restoration practices can be thwarted by climate extremes. *Science of the Total Environment*, 626, 851–859.
- Maccherini, S., Marignani, M., Castagnini, P., & van den Brink, P.J. (2007). Multivariate analysis of the response of overgrown semi-natural calcareous grasslands to restorative shrub cutting. *Basic and Applied Ecology*, 8, 332–342.
- Maccherini, S., & Santi, E. (2012). Long-term experimental restoration in a calcareous grassland: identifying the most effective restoration strategies. *Biological Conservation*, 146, 123–135.
- Moore P.D., & Chapman S.B. (1986). *Methods in Plant Ecology*. Blackwell Scientific Publications, Oxford.
- Pavoine, S. (2020). *adiv: an R package to analyse biodiversity in ecology*. *Methods in Ecology and Evolution*, 11, 1106-1112.
- Ricotta, C., Bacaro, G., Maccherini, S., & Pavoine, S. (2022). Functional imbalance not functional evenness is the third component of community structure. *Ecological Indicators*, 140, 109035.
- Ricotta, C., Podani, J., Schmera, D., Bacaro, G., Maccherini, S., & Pavoine, S. (2023). Data from: The ternary diagram of functional diversity. Dryad Digital Repository, <https://doi.org/10.5061/dryad.7pvmcvdzc>
- Ricotta, C., Szeidl, L., & Pavoine, S. (2021). Towards a unifying framework for diversity and dissimilarity coefficients. *Ecological Indicators*, 129, 107971.
- Roberts, D.W. (2017). Distance, dissimilarity, and mean-variance ratios in ordination. *Methods in Ecology and Evolution*, 8, 1398–1407.
- Van den Boogaart, K.G., & Tolosana-Delgado, R. (2013). *Analyzing compositional data with R*. Springer, Heidelberg.
- Van den Boogaart, K.G., Tolosana-Delgado, R., & Bren, M. (2018). *Compositions: Compositional Data Analysis*. R package version 2.0-4. <https://CRAN.R-project.org/package=compositions>
- Vicente-Gonzalez, L., & Vicente-Villardón, J.L. (2021). PERMANOVA: Multivariate Analysis of Variance Based on Distances and Permutations. R package version 0.2.0. <https://CRAN.R-project.org/package=PERMANOVA>
- Wright, I.J., Reich, P.B., Westoby, M., Ackerly, D.D., Baruch, Z., Bongers, F., Cavender-Bares, J., Chapin, T., Cornelissen, J.H.C., Diemer, M., Flexas, J., Garnier, E., Groom, P.K., Gulias, J., Hikosaka, K., Lamont, B.B., Lee, T., Lee, W., Lusk, C., Midgley, J.J., Navas, M.L., Niinemets, U., Oleksyn, J., Osada, N., Poorter, H., Poot, P., Prior, L., Pyankov, V.I., Roumet, C., Thomas, S.C., Tjoelker, M.G., Veneklaas, E.J., & Villar, R. (2004). The worldwide leaf economics spectrum. *Nature*, 428, 821–827.