RESEARCH ARTICLE

Physical and land-cover variables influence ant functional groups and species diversity along elevational gradients

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Abstract Of particular importance in shaping species assemblages is the spatial heterogeneity of the environment. The aim of our study was to investigate the influence of spatial heterogeneity and environmental complexity on the distribution of ant functional groups and species diversity along altitudinal gradients in a temperate ecosystem (Pyrenees Mountains). During three summers, we sampled 20 sites distributed across two Pyrenean valleys ranging in altitude from 1,009 to 2,339 m by using pitfall traps and hand collection. The environment around each sampling points was characterized by using both physical and land-cover variables. We then used a self-organizing map algorithm (SOM, neural network) to detect and characterize the relationship between the spatial distribution of ant functional groups, species diversity, and the variables measured. The use of SOM allowed us to reduce the apparent complexity of the environment to five clusters that highlighted two main gradients: an altitudinal gradient and a gradient of environmental closure. The composition of ant functional groups and species diversity changed along both of these gradients and was differently affected by environmental variables. The SOM also allowed us to validate the contours of most ant functional groups by highlighting the response of these groups to the environmental and land-cover variables.

Keywords Ants · Community ecology · Elevation gradient · Landscape heterogeneity · Neural networks · Pyrenees

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Introduction

One of the main concerns in community ecology is to identify the environmental factors (either biotic or abiotic) that shape species assemblages (Rosenzweig 1995). Of particular importance in this respect is the heterogeneity created by the variation of these factors. According to the hypothesis of habitat heterogeneity suggested by MacArthur and MacArthur (1961), species richness should increase with increasing structural complexity of the environment. This relationship has indeed been found in many taxa, e.g. arthropods, birds, mammals, amphibians or reptiles (see Tews et al. 2004 for a review). Environmental heterogeneity can significantly influence not only species richness but also their relative distribution. The distribution of ants for example is significantly affected by the spatial heterogeneity generated by fire (Parr and Andersen 2008), anthropogenic disturbances (Kalif et al. 2001), habitat fragmentation (Vasconcelos et al. 2006), or grazing (Bestelmeyer and Wiens 1996). Natural gradients (e.g. altitude, latitude) are also a major source of spatial heterogeneity that can influence the structure of species assemblages. Mountainous areas in particular are characterized by rapid changes in climate, soil, or vegetation, over relatively short distances (Körner 2007). They thus offer considerable landscape heterogeneity on a condensed area and are ideal for exploring the ecological mechanisms underlying spatial patterns in species richness and distribution.

In this study, we investigated the influence of spatial heterogeneity and environmental complexity along altitudinal gradients on the distribution of ant functional groups and species diversity across two Pyrenean valleys: one located in Andorra, on the Southern side of the Pyrenees (the Madriu-Perafita-Claror valley), and another located in France, on the Northern side of the Pyrenees (the Pique valley). The categorization of organisms into functional groups has been widely used in the study of animal communities (birds: Cody 1985; reptiles: Pianka 1986). Species classification by functional groups reduces the apparent complexity of animal communities (Andersen 1997a) and thus facilitates the understanding of the general principles that govern the functioning of ecosystems. Although the classification in functional groups has been used to study the ant fauna of Australia (Andersen 1995; Hoffmann and

Andersen 2003), South (Bestelmeyer and Wiens 1996) and North America (Andersen 1997a; Stephens and Wagner 2006), and Asia (Pfeiffer et al. 2003), this method has been rarely used to study the ant fauna of Europe (but see Gómez et al. 2003).

Environmental heterogeneity may act at multiple scales on animals, both spatially and temporally (Wiens 1989; Levin 1992). All of these scales however may not be relevant to understand how an animal interacts with its environment and the choice of the spatial scale at which to study environmental heterogeneity should be consistent with its perception of the environment. This requires the selection of appropriate descriptive variables (Turner et al. 2001). Physical, chemical and biological data, however, are often difficult to analyze in an integrated way because they are complex, noisy, and vary and covary in a non-linear way (Lek and Guégan 2000). One solution is to use modeling techniques, such as artificial neural networks, that are able to take into account the complex structure of multi-dimensional datasets (Chon 2011). For example, the Self-Organizing Map algorithm (SOM, unsupervised neural network, Kohonen 2001) is a powerful and well-suited tool to detect patterns in animal communities in relation to environmental variables (Lek and Guégan 2000). SOMs have been used in ecology to study mostly aquatic insect or fish communities (e.g. Compin and Céréghino 2007; ants: Groc et al. 2007; Delabie et al. 2009; Céréghino et al. 2010). In this study we used SOM to fulfill two main objectives: (1) to describe landscape spatial patterns along altitudinal gradients and to explore whether the apparent complexity of mountain environments can be reduced to a few simple elements and (2) to address the question of how ant functional groups and pattern of species diversity respond to the changes in land-cover and physical variables along these gradients.

Methods

Study area and sampling sites

Our study area was located in the Pyrenees, a mountain range located in south-west Europe and that is shared between Spain, France and the Principality of Andorra. Because of their orientation and geographic location, these mountains present considerable

climatic contrasts. The northern and western sides of the Pyrenees have an oceanic climate, with rainfall throughout the year, mild winters and cool summers. The southern side in contrast has a more continental climate, characterized by high solar radiation, torrential rains at equinoxes, large temperature variations, and very cold winters and dry summers.

Two valleys were sampled in this study: the Madriu-Perafita-Claror, in Andorra, and the Pique valley, in France. The Madriu-Perafita-Claror valley is a glacial valley located in the southeast part of Andorra that covers an area of 4,247 ha. The valley is oriented along an east–west axis and extends along an altitudinal gradient ranging from 1,055 to 2,905 m. The valley is well preserved: the production of timber has ceased in the 1950s' and since the 1980s' there has been almost no human intervention. Because of its state of preservation, the Madriu-Perafita-Claror valley has been registered in 2004 as World Heritage by UNESCO for its cultural landscape (www.unesco.org, see Madriu-Perafita-Claror valley). The Pique valley is a glacial valley predominantly oriented along a north–south axis, extending along an altitudinal gradient ranging from 650 to 3,116 m. It is dominated by peaks over 3,000 m in altitude that lie on the border between France and Spain. This valley is part of the Natura 2,000 sites (www.natura2000.fr/); it covers an area of 8,251 ha divided into two main valleys (the Pique valley and the Lys valley).

We sampled ants at 20 sites (9 sites in the Madriu valley and 11 sites in the Pique valley) in July–August 2005 to 2007. To select the sampling sites, three main factors were considered: elevation, exposure, and type of vegetation cover. We sampled along an altitudinal gradient ranging from 1,300 to 2,300 m for the Madriu valley, and from 1,000 to 2,300 m for the Pique valley. Sampling could not be achieved over a larger altitudinal gradient, because of high anthropogenic pressures below 1,300 m in the Madriu valley, and below 1,000 m in the Pique valley. Locations higher than 2,300 m were not sampled because ant species richness beyond this altitude is known to be very low (Glaser 2006). The two valleys were thus sampled on 62.5 and 61.4 % of their altitudinal range, for the Madriu and Pique valleys respectively. The different categories of vegetation covers considered for the selection of the sampling sites were: forest, meadow, scree and bushes. Table S1 gives the main characteristics of the sampling sites for the two valleys.

Sampling methods and species identification

At each of the 20 sites, we used a variation of the ALL protocol (Agosti et al. 2000) to sample the ants. A 190 m long line transect was traced and sampling points were placed on this line every 10 m (making a total of 20 sampling points per site, yielding a total 400 sampling points for the two valleys). The position of the sampling points were recorded by means of a GPS (Garmin® eTrex®) and subsequently loaded into DIVA-GIS, a free geographic information system (www.diva-gis.org).

Two collection methods were used to sample the ants at each sampling point: pitfall traps and hand collection. The pitfall traps consisted of plastic cups (diameter: 35 mm, height: 70 mm), filled to one-third of their height with ethylene glycol. The cups were buried so that their upper lip was flushed with the surface of the substrate. The pitfall traps were therefore set in action immediately and were left in place for 5–8 days (Table S1). The pitfalls could not be operated for the same length of time because access to some of the transects was difficult and was sometimes prevented by adverse meteorological conditions. Pitfall trapping was supplemented by hand collecting around each sampling point at the moment the pitfalls were removed. Hand collecting consisted of one person (the same person for all transects) picking up all visible ants within a 2 m radius around each trap during a maximum of 3 min. Ants were searched on the ground and in the vegetation; potential nesting sites were also inspected (dead wood, underneath stones/bark). The combination of pitfall and hand collecting sampling techniques is known to perform well in temperate regions (Groc et al. 2007). Winkler extractors were not used because the leaf litter is generally shallow (because of heavy rainfall, the presence of rocks, and high slope inclination) or relatively poor (particularly in coniferous forests) in mountainous environments. All ants collected at each sampling point were placed in plastic vials filled with 90 % ethanol. Once in the laboratory, ants were identified to the species level using available keys (Seifert 2007).

Because ants are social insects, a single sample may contain a high abundance of a rare species. Our analyses are therefore based on the species occurrence in the samples rather than on the number of individuals. A sampling point thus corresponds to the presence/absence of various species collected at a

sampling site by a pitfall trap or by hand collection around the pitfall or by both sampling methods. Consequently, the theoretical maximum of a species occurrence in a transect is 20.

Environmental variables and habitat characterization

The 20 sites sampled and the micro-environment around each pitfall were characterized by using four physical and eight land-cover variables. These 12 variables were chosen because they have been shown to be consistently correlated with ant species richness in previous studies (for physical variables see for example: Kaspari et al. 2004; Sanders et al. 2007; Dunn et al. 2009a). The physical variables considered were: annual mean temperature (in °C), annual precipitation (in mm), elevation a.s.l (in m) and slope. Annual mean temperature and annual precipitation were obtained from two GIS data layers (30 arc-seconds) of the WorldClim 1.4 database (Hijmans et al. 2005), whereas elevation was recorded directly in the field by a GPS. WorldClim computes temperature as a function of elevation, which means that all points of a transect where characterized by the same temperature value in our study. The slope was characterized locally around each pitfall by the same person throughout the whole study using the following scale: 0 (null to gentle slope), 1 (moderate slope), 2 (strong slope). To describe the area surrounding the pitfalls, digital photographs centered on each pitfall were taken. Then, on each photograph, we considered an area of 1 m^2 centered on the pitfall and used an image analysis software to delineate the outline of the following land-cover variables within this area: shrub, bare rock/pebbles, dead wood/stump, litter, grass and bare soil. The percentage of area covered by each of these elements was then determined. In addition, we also noted the presence/absence of either a hardwood or coniferous canopy above each pitfall.

Ant functional groups

All ant species collected in the Madriu and Pique valleys were classified into five functional groups (see Table S2) according to the categorization proposed by Roig and Espadaler (2010). This latter is an adaptation for the Iberian Peninsula and Balearic Islands of the classification used by Andersen (1995, 1997a, 2000) for

Australian and North American ants and on that used by Bestelmeyer and Wiens (1996) for South American ants. Given that some genera (e.g. *Formica* and *Lasius* in this study) are heterogeneous in terms of their ecology and behaviour, different functional groups sometimes share species of the same genus. The five following functional groups were distinguished:

- *Opportunists (O)* these are, in general, unspecialized species, whose distributions are strongly influenced by competition with other ants. According to Andersen (2000), these species often span a large diversity of habitats. They predominate in areas where stress or disturbance limit ant diversity and biomass and thus in which behavioural dominance is low. In our study, this group is represented by species of the genus *Formica* and by species like *Tapinoma erraticum* and *Tetramorium impurum*. *T. erraticum* was not classified as a *Dominant Dolichoderinae* because its societies are small (Seifert 2007) with a much reduced worker number compared, e.g., to the polydomous species *T. nigerrimum*.

- *Social Parasites (SP)*: this group gathers species that are either temporary (e.g. *Lasius mixtus*) or permanent (e.g. *Strongylognatus testaceus*) social parasites.

- *Coarse Woody Debris Specialist (CWDS)* this group is represented by two species: *Camponotus herculeanus* and *C. ligniperda*. These species nest in stumps or tree trunks.

- *Cold Climate Specialists/Shadow Habitats (CCS/SW)*: these species have their distributions centered on cold climate areas (Andersen 2000). They are generally characteristic of habitats where the abundance of dominant dolichoderines is low (Andersen 2000). This group is mainly represented in the Madriu and Pique valleys by the genera *Formica*, *Lasius* and *Myrmica*. The ants of the genus *Myrmica* were classified as *CCS/SW* rather than *Opportunists* because this genus is mainly present within mountainous, humid and grassy environments (Radchenko and Elmes 2010).

- *Cryptics (C)* these species are small to tiny species. This group is predominantly represented by myrmicines and ponerines that nest and forage within soil, litter and dead branches (Andersen 2000). These ants are mainly present in forested habitats. In our study, this group is represented by

the two genera *Leptothorax* and *Temnothorax* and by one species of the *Lasius* genus: *L. flavus*. *Leptothorax* and *Temnothorax* species were included in the *Cryptics* functional group because they have cryptic behaviour in the sense that they forage singly, move slowly and "have little interaction with other epigaeic ants" (Andersen 1995).

Data analysis

To estimate total ant species richness at the valley and transect levels and to evaluate the completeness of our samples, Chao2, a non parametric richness estimator, was calculated with the program EstimateS 7.5.2 (100 replicates) (Colwell 2005).

We used the SOM Toolbox (version 2) for Matlab® developed by the Laboratory of Information and Computer Science at the Helsinki University of Technology (http://www.cis.hut.fi/projects/somtoolbox/, see Vesanto et al. (1999) for practical instructions). The SOM is an unsupervised learning procedure which transforms a set of multidimensional data into a two dimensional map subject to a topological constraint (see Kohonen 2001 for details). The data are projected onto a rectangular grid composed of hexagonal cells, forming a map (Giraudel and Lek 2001). The SOM plots the similarities of the data by grouping similar data items together as follows:

(i)   Virtual samples (visualized here as hexagonal cells) are initialized with random samples taken from the input data set.
(ii) The virtual samples are updated in an iterative way: (1) a sample unit is randomly chosen as an input unit, (2) the Euclidean distance between this sample unit and every virtual sample is computed, (3) the virtual sample closest to the input unit is selected and called 'best matching unit' (BMU), and (4) the BMU and its neighbours are moved a bit towards the input unit.
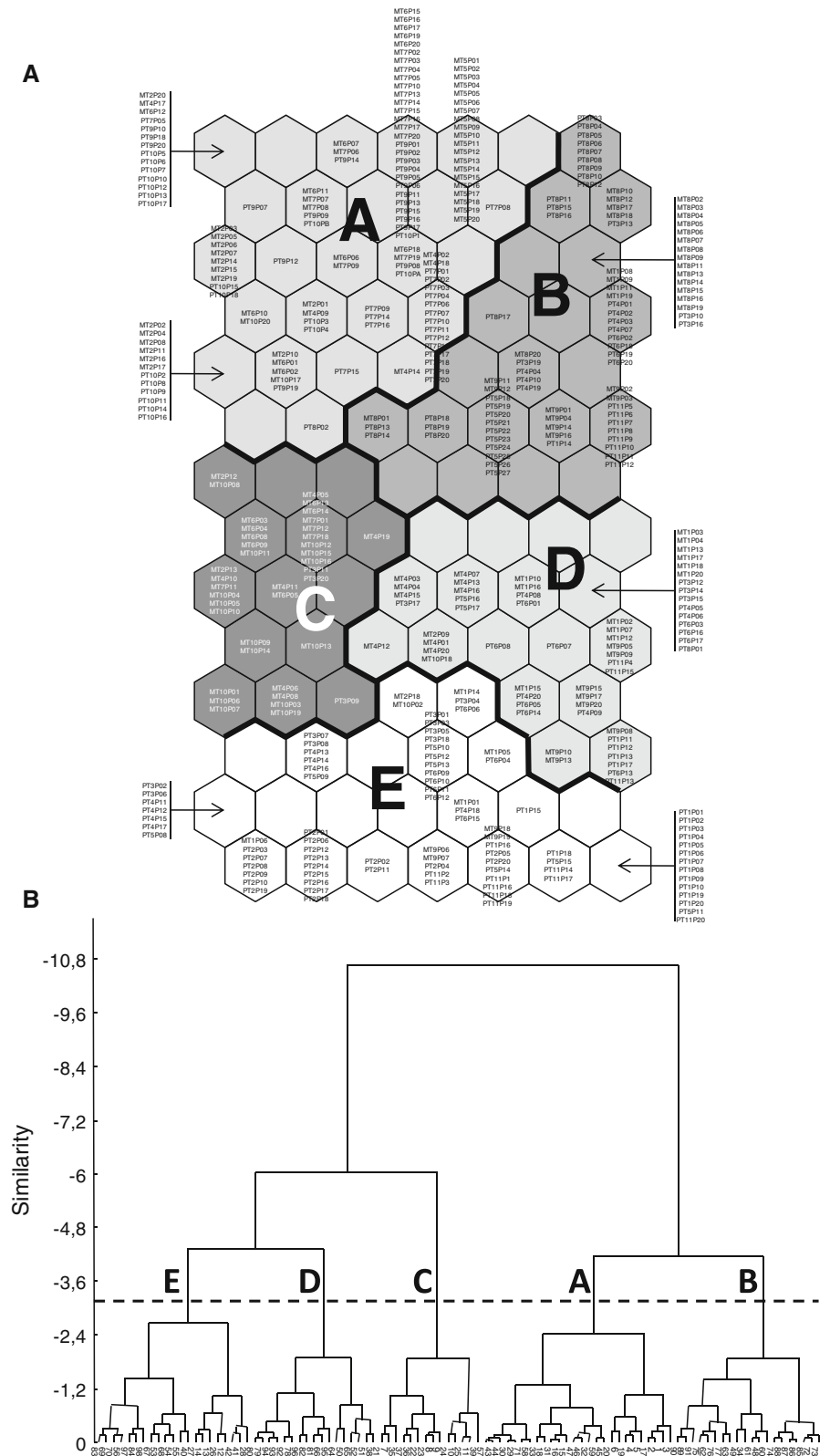
The training is separated into two parts:

(i)   Ordering phase (the 3,000 first steps): when this phase takes place, the samples are highly modified in the wide neighbourhood of the BMU.

(ii) Tuning phase (7,000 steps): during this phase, only the virtual samples adjacent to the BMU are lightly modified. At the end of the training, the BMU is determined for each sample, and each sample is set in the corresponding hexagon of the SOM map. Neighbouring samples on the grid are expected to represent adjacent clusters of samples. Consequently, sampling points appearing distant in the modelling space (according to physical and land-cover variables) represent expected differences among sampling points in real environmental characteristics.

The self-organizing map for this study consists of two layers of neurons connected by weights: an input layer and an output layer. The input layer was composed of 12 neurons (one per variable) connected to the 400 sampling points. The output layer was composed of 98 neurons (see below) visualized as hexagonal cells organized on an array of 14 rows by 7 columns (Fig. 1a). The number of output neurons (map size) is important to detect the deviation of the data. If the map size is too small, it might not explain some important differences that should be detected (Compin and Céréghino 2007). Conversely, if the map size is too big, the differences are too small. We followed the procedure described in Park et al. (2003) and Céréghino and Park (2009): the network was trained with different map sizes (4–200 neurons) and we chose the optimum map size based on local minimum values for quantization and topographic errors. Quantization error is the average distance between each data vector and its BMU and, thus, measures map resolution. Topographic error represents the proportion of all data vectors for which 1st and 2nd BMUs are not adjacent, and is used for the measurement of topology preservation The number of 98 output neurons retained for this study fitted well the heuristic rule suggested by Vesanto et al. (2000) who reported that the optimal number of map units is close to $5\sqrt{n}$, where n is the number of samples. For each sampling point, we made a list of the different species collected and determined the values of the environmental variables characterizing the sampling point. To highlight the relationships between the different ant functional groups and the environmental variables, the number of species occurrences of each functional group was introduced into the SOM previously trained

Fig. 1 Distribution of the sampling points on the self-organizing map (SOM). **a** The sampling points are distributed according to four physical variables (altitude, slope, mean annual temperature and mean precipitation) and eight land-cover variables variables (presence of a hardwood or coniferous canopy, percent area covered by shrub, bare rock/pebbles, dead wood/stump, litter, grass and bare soil in a 1 m^2 area around each pitfall). Altitude, mean annual temperature and mean precipitation were obtained by using a GIS software while slope and the eight environmental variables were estimated directly in the field or by the analysis of digital photographs centered on each sampling points. The codes used to designate the sampling points on the SOM refer to the valley (M for Madriu, P for Pique), the transect number within the valley, and the location of the sampling points within the transect (e.g.: MT2P20, Madriu valley, Transect 2, sampling points 20). Neighboring sampling points on the self-organizing map share similar environmental characteristics. **b** The SOM units were classified into five clusters (*A*, *B*, *C*, *D* and *E*). The boundaries of the five clusters (*A*, *B*, *C*, *D* and *E*) were obtained by applying Ward's algorithm to the weights of the variables in the SOM hexagons. The smallest branches with numbers in the dendrogram correspond to the SOM neurons

with the four physical and the eight land cover variables that characterize each sampling point. During the training of the map, we used a mask to give a null weight to the five functional groups, whereas physical and land-cover variables were given a weight of 1. Therefore, the search for the BMU was based on the 4 physical and 8 land-cover variables only. Setting the mask value to zero for a given component (here for each of the five functional groups) removes the effect of that component on the map organization (Vesanto et al. 2000). The values and distributions of the functional groups were then visualized on the SOM previously trained with physical and land-cover variables only and formed by the 98 hexagonal cells.

In a last step, Ward's algorithm was used to identify the boundaries between each cluster on the Kohonen map (Fig. 1b). The distributions of the number of species occurrences in each ant functional groups in the different clusters were compared using the χ^2 test for independent samples (Siegel and Castellan 1988). To further analyze the distribution of functional groups within each cluster, an analysis of residuals was performed (Siegel and Castellan 1988). This analysis tests the contribution of each functional group to each cluster. Moreover, it reveals whether a functional group is positively or negatively associated to a given cluster.

We used a generalized linear mixed model (GLMM) with a Poisson error distribution to examine the variation in ant species richness per sampling point among the SOM clusters. To account for spatial autocorrelation among sampling points located in the same transects and in the same valley, the variable transect was nested within the variable valley and was entered as a random variable in the model. To assess the overall effect of the SOM clusters on species diversity we fitted a first model in which the SOM cluster variable was entered as a fixed effect categorical factor and the transect variable (nested within valley) was entered as a random effect categorical factor. We then fitted a second model with no fixed effects and compared the two models with a likelihood ratio test (Zuur et al. 2009). The different SOM clusters were then regrouped by removing non-significant factor levels in a stepwise a posteriori procedure (Crawley 2007). The models were fitted with the statistical software R 2.11.0 (R Development Core Team 2011) and the R-package lme4 (linear mixed-effects models using S4 classes, Bates et al. 2011) using the function glmer.

Results

Classification of sampling sites

After training the Kohonen map with the four physical variables and the eight land-cover variables, five clusters of sampling sites obtained from the SOM output were identified (Fig. 1a, b).

The SOM allowed us to identify two main gradients (Fig. 2 and Fig. S1): a first gradient extending from the lower right to the upper left corner of the map (clusters D and E vs. cluster A, B and C), which corresponds to an altitudinal gradient ranging from low to high altitudes, and a second gradient, extending from the bottom to the top of the map (clusters C, D and E vs. cluster A and B), which corresponds to a gradient of environmental closure, ranging from closed to open areas.

Cluster B corresponds to sampling sites of medium elevations located in open areas, e.g. grassland areas (Fig. 2). Cluster A is equivalent to cluster B but for high elevation. It includes sampling sites typical of mountain environments, e.g. screes of high altitudes located on steep slopes. A large proportion of the sampling sites of cluster A is dominated by bare rocks and shrubs (Fig. 2). Clusters C and E correspond to sampling sites in forest areas: cluster E to low altitude forests dominated by hardwood, with a high abundance of litter and dead wood, and cluster C to high altitude forests in which conifers are predominant. Note that for cluster E the sampling sites with high slopes are also characterized by bare soil. Cluster D corresponds to a transition area between hardwood forests and grasslands (Fig. 2).

Distribution of ant functional groups and species diversity

In total, 42 ant species were found in the two valleys. The number of species collected at each transect varied between 25 at 1,351 m and 2 at 2,339 m, and between 14 at 1,009 m and 4 at 2,299 m, for the Madriu and Pique valley respectively. The Chao2 estimator indicated that between 66 and 100 % (mean ± SD = 93.48 ± 10.89) of the expected maximum number of species were collected for the 9 transects in the Madriu valley, and between 63 and 100 % (mean ± SD = 90.74 ± 12.29) for the 11 transects in the Pique valley using pitfall traps and
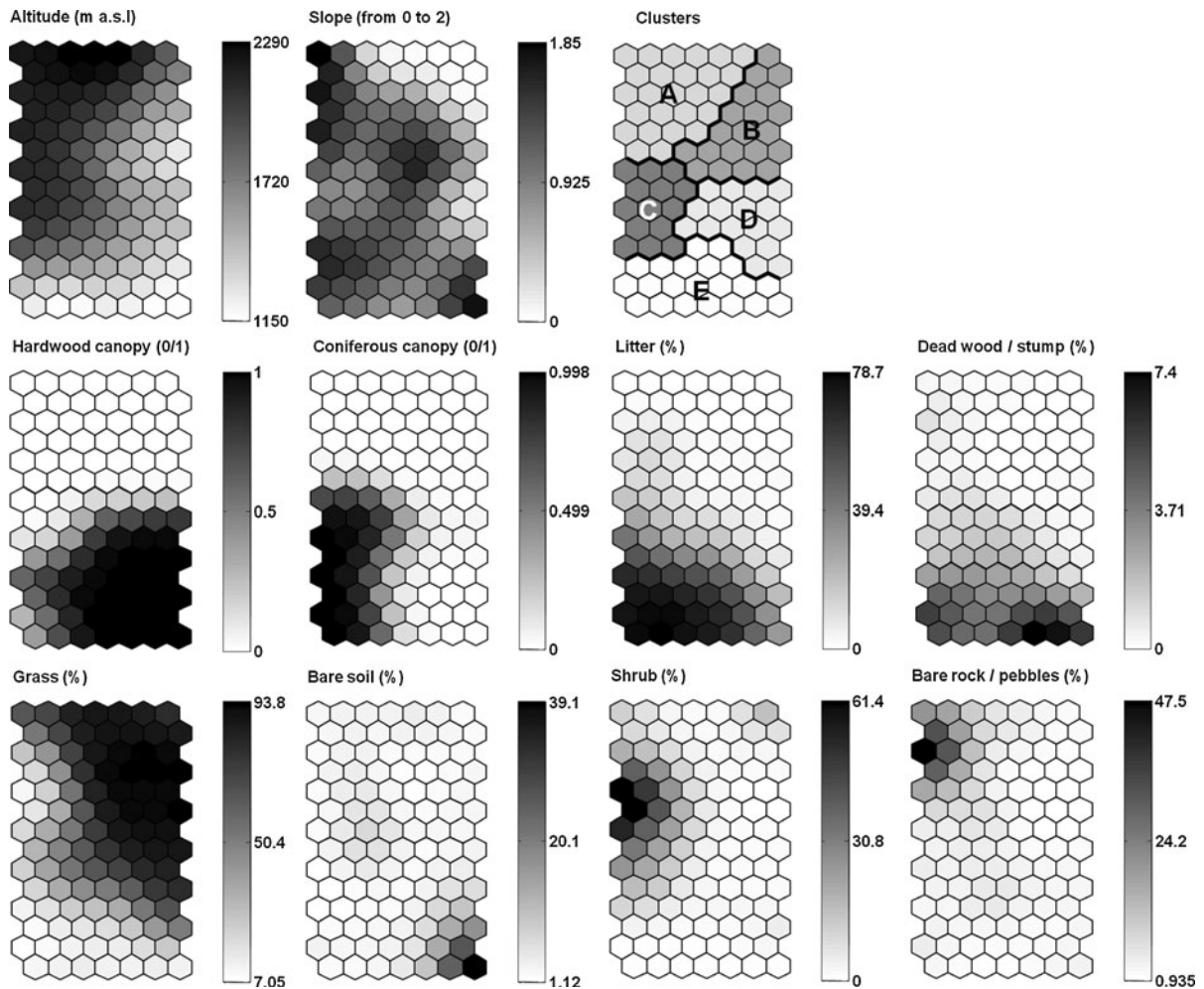
Fig. 2 Gradient distribution of each environmental variable on the trained self-organizing map. A grayscale (dark = high value, light = low values) was used to visualize the value of the variables. The SOM allows to derive two main gradients: an altitudinal gradient ranging from low (clusters *D* and *E*) to high (clusters *A*, *B* and *C*) altitudes, and a gradient of environmental closure, ranging from closed (clusters *C*, *D* and *E*) to open (clusters *A* and *B*) areas. See also Fig. S1 for the mean annual temperature and mean precipitation variables

hand collecting. There was no relationship between these values and the duration of pitfall activity for the Madriu valley (Spearman's rank correlation: $r = -0.49$, $P = 0.17$, $n = 9$) or the Pique valley (Spearman's rank correlation: $r = 0.15$, $P = 0.64$, $n = 11$) (Table S1).

The distribution of the five functional groups on the SOM previously trained with the physical and landcover variables are shown in Fig. 3. With the exception of Social Parasites, all functional groups are present in the five clusters of the Kohonen map. The Cold Climate Specialists/Shadow Habitats is the dominant functional group in most clusters (range:

44–58 %, mean: 56 %, Fig. 4a). The Opportunists is the second largest group (range: 23–47 %, mean: 30 %), followed by the Cryptics (range: 8–19 %, mean: 11 %), the Coarse Woody Debris Specialists (range: 1–15 %, mean: 5 %) and the Social Parasites (range: 0–3 %, mean: 1 %) (Figs. 3, 4a).

The distribution of species occurrences in each functional group was not homogeneous across the five clusters ($\chi^2 = 96.16$, $df = 16$, $p < 0.001$). The Coarse Woody Debris Specialists functional group was significantly and positively associated with cluster C (residuals = 6.2, Figs. 3, 4b) and negatively associated with cluster B (residual = −2.4, Figs. 3, 4b)
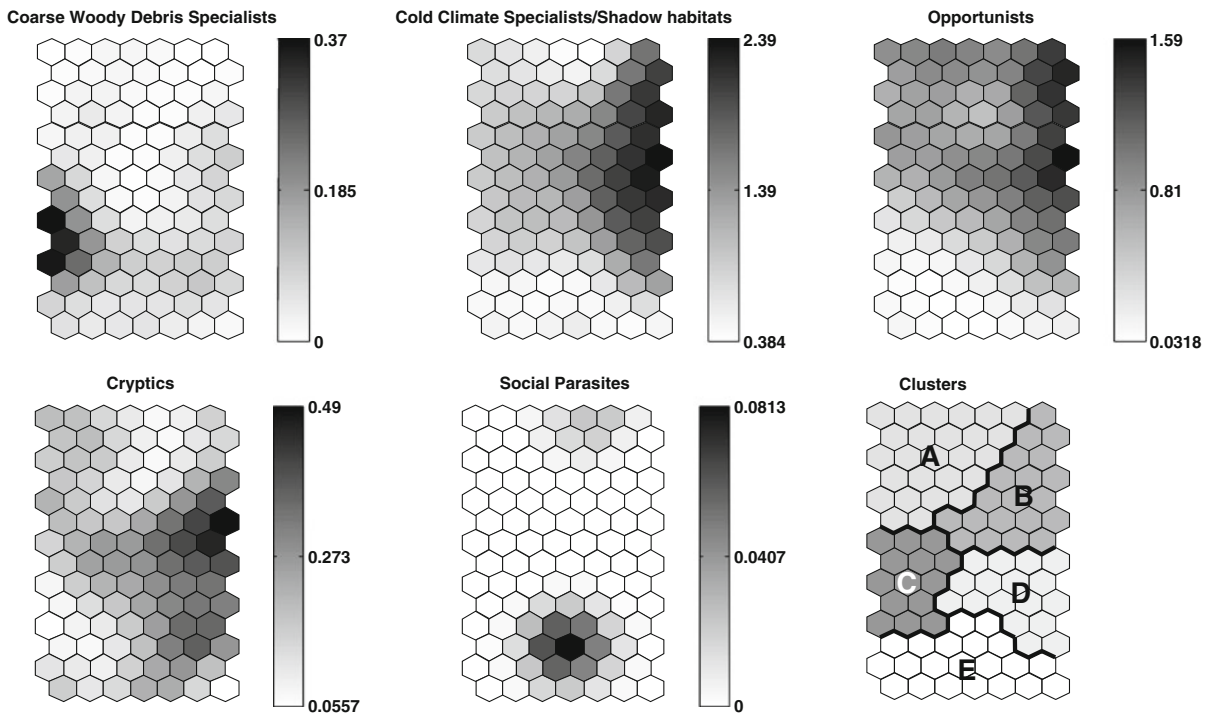
Fig. 3 Ant functional groups. Visualization of the five ant functional groups on the self-organizing map trained with the four physical and the eight land-cover variables. Each functional group has its own distribution pattern. The functional groups occupying similar zones on the map have a high probability to be associated and to be found in the same area. A grayscale (light = low values, dark = high values) was used to visualize the level of presence of each functional group. Note that the *grayscales* are different for each functional group
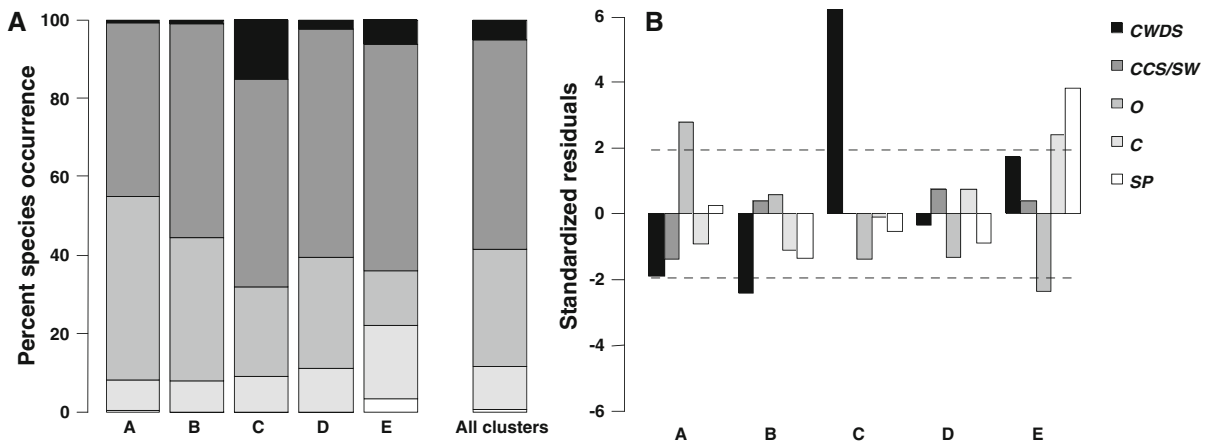

Fig. 4 Analysis of the distribution of the ant functional groups. **a** Percentage of species occurrences for each functional group found in the Madriu and Pique valley. The percentages are given for each of the five clusters identified by Ward's algorithm on the self-organizing map and for all clusters grouped together. **b** Residual values for the five ant functional groups found in the Madriu and Pique valley. The residual values are given for each of the five clusters identified on the self-organizing map. According to the sign of their residual values, the functional groups may be positively or negatively associated with the clusters. The two dotted lines represent the significance threshold at $P = 0.05$. *O* opportunists, *SP* social parasites, *CWDS* coarse woody debris specialist, *CCS/SW* cold climate specialists/shadow habitats and *C* cryptics

and to a lesser extent to cluster A (residual = –1.8, not significant, Figs. 3, 4b). This functional group is thus characteristic of woodland areas and is negatively associated with open areas. The Cold Climate Specialists/Shadow Habitat and Opportunist functional groups do not show any clear distribution pattern. They are abundant in all clusters (Fig. 4a). The Social Parasites are significantly and positively associated with cluster E (residuals = 3.8, Fig. 4b). The Cryptic group was significantly present in cluster E (residuals = 2.3, Fig. 4b) and is thus associated with the presence of litter and hardwood canopy.

The distribution of ant species richness per sampling point differed significantly among the five clusters (GLMM, $\chi^2 = 94.18$, $df = 4$, $p < 0.001$). Cluster E had the lowest ant species richness (mean ant species richness per sampling point $\pm CI_{0.95}$: 0.74 ± 0.27) while clusters B (3.77 ± 0.38) had the highest one (Fig. 5). Cluster D was only marginally significantly different from clusters A and C (GLMM, z value = 1.93, $p = 0.052$).

Discussion

In this study we used a Self-Organizing Map algorithm to categorize 400 sampling points on the basis of 12
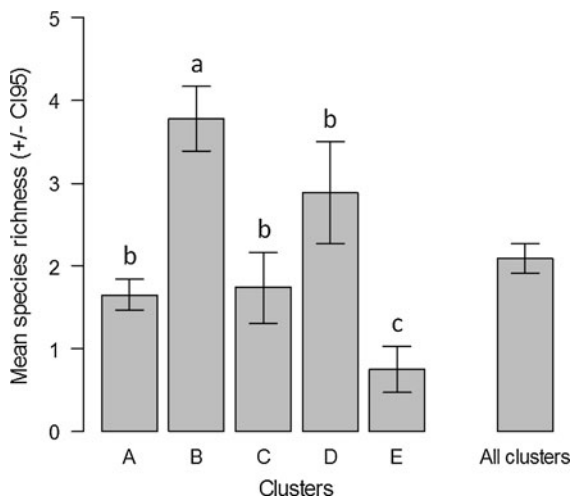

Fig. 5 Ant species richness. Mean number of ant species ($\pm CI_{0.95}$) per sampling point for each SOM cluster (*A–E*) and for all clusters grouped together. Significant differences in ant species richness between clusters were tested with a generalized linear mixed model (GLMM) with a Poisson error distribution. Different letters above the *error bars* indicate significant differences in ant species richness at $P < 0.05$

environmental variables characterizing the physical environment and the type of land-cover around each sampling point. The SOM algorithm reduced the complexity of the database to five clusters of sampling points corresponding to high and low elevation grassland areas, hardwood and coniferous forests, and a transition area between hardwood forests and grasslands. These clusters highlight two main gradients: an altitudinal gradient that mimics to a certain extent the altitudinal zonation of vegetation found in the Pyrenees (Ninot et al. 2007) and a gradient of environmental closure. We then categorized the species found at the sampling points into five functional groups and used the SOM map generated by the algorithm to study the distribution of these groups in the environments sampled. Using this method, we were able to validate the contours of most functional groups by positively or negatively correlating their distribution with the environmental variables measured. The distribution of ant functional groups changed along environmental gradients and was differently affected by environmental variables. Finally, we examined the distribution of ant species richness across the five clusters of sampling points identified by the SOM algorithm to find out the environmental characteristics associated with low or high ant species diversity. Three of the five functional groups (*Coarse Woody Debris Specialists, Social Parasites and the Cryptic*) showed a clear pattern of association with particular features of the environment such as the presence of litter and canopy. These three groups however represented only 31 % of the 42 species collected whereas the distribution pattern of the two other functional groups (*Cold Climate Specialist/Shadow Habitat* and *Opportunists*) that represented 69 % of the total number of species collected was much less clear. Our results show therefore that the distribution of the five functional groups we defined change along environmental gradients, and that they are thus differently affected by environmental variables. Assuming that species are more likely to reach neighboring areas than areas far apart and that neighboring sampling points tend to exhibit similar physical features, small-scale autocorrelations of ant assemblages were suggested from the SOM outputs (Figs. 1, 2, 3). However, almost all ant functional groups (four out of five) were present within all SOM clusters which shows that spatial autocorrelation alone cannot explain the SOM outputs.

Indeed, if spatial autocorrelation were the only factor explaining the relationship between sampling points then each of the SOM cluster would correspond to an ant functional group.

Why does the SOM analysis show such a discrepancy among functional groups in their pattern of association with environmental variables and why do in particular both the Cold Climate Specialist/Shadow Habitat and Opportunists functional groups appear to be so widely distributed in our sampling area? Two explanations could be provided. The first explanation could be that this result reflects true particular biological traits of the species belonging to these groups. According to Andersen (1995, 1997a, 2000) the distribution of the Cold Climate Specialists/Shadow Habitat species is centred on cool-temperate regions. In the two valleys we sampled, this group was represented by three genera: *Formica*, *Lasius* and *Myrmica*. Most of the species of these genera are holarctic and are characteristic of the cold regions of the northern hemisphere (Bernard 1968). Their success in these regions is mainly due to specific behavioural and/or physiological adaptive traits that allow them to resist to low temperatures (Heinze 1992; Maysov and Kipyatkov 2009). Since the whole area we sampled was located in a temperate mountainous region it should therefore come as no surprise that the Cold Climate Specialists/Shadow Habitat functional group was not found to be associated with any particular specific environmental variable. As for the species belonging to the Opportunistic functional group, we found that they occupy a wide range of habitat but were particularly present in grassland areas of high altitudes, a relatively stressful environment for ants, both because of the low temperatures, characteristic of high altitudes, and of the scarcity of food (Andersen 2000).

A second and alternative explanation to the discrepancy found among functional groups in their pattern of association with environmental variables could be linked to the criteria used to define the functional groups. The criteria used to define the Cold Climate Specialist/Shadow Habitat and Opportunists functional groups could not be relevant to obtain clear patterns with the SOM analysis. As pointed out by Andersen (1997b) the definition of functional groups is scale-dependent and one should thus be cautious in using them in community ecology studies. As a case in point Andersen (1997b) gives the example of the mound-building species of the genus *Formica*. At a local scale, these species are behaviorally dominant throughout the Holarctic and they could thus be described as belonging to the dominant species functional group in local ant fauna (Andersen 1997b; Savolainen and Vepsäläinen 1988). However, this dominance is limited to cool-temperate regions and at a global scale they would rather be considered as belonging to the group of cold-climate specialists. The importance of competition and dominance in ant community structure is thus scale dependent. The categorization in functional groups used in our study corresponds to that proposed by Roig and Espadaler (2010) to describe the ant fauna of the Iberian Peninsula and Balearic Islands. Applied at the local scale of our study area, this categorization may not be discriminative enough (Andersen 1997b) and could conceal the response of some ant species to particular ecological variables. A solution could have been found in subdividing some of the functional groups we used (Bestelmeyer and Wiens 1996; Andersen 1997b). This could have increased the discriminative power of our analysis and as a consequence, clearer successional patterns in relation to environmental variables could have been revealed.

The ant species belonging to the Coarse Woody Debris Specialists and Cryptic functional groups were significantly and positively associated with woodland areas and negatively associated with open areas. The strong response of these two groups to the presence of a canopy can most likely be explained by their nesting habit. As a caveat however one should keep in mind that SOM is a correlative analysis and thus does not convey information on the mechanisms generating the distributions observed. For example, we do not know if the presence or absence of a species in a given environment results from an effective choice of a habitat by newly mated queens during colony founding, from an impossibility to colonize a particular environment, or whether it results from competition mechanisms. For the Social Parasites, as for the Coarse Woody Debris Specialists and Cryptic groups, these species show a clear and localized pattern of distribution on the SOM. However, this result has to be interpreted cautiously. This functional group probably does not respond to physical and land-cover variables per se, but rather to the presence or absence of its hosts.

The mean number of species in the five clusters we identified ranged from 0.7 to 3.7. Clusters B and D have

a higher species richness compared to clusters A and C which have also the highest mean altitudes. The decrease in ant species richness with increasing altitude has been reported in other studies (e.g. Sanders et al. 2007, 2010; Lessard et al. 2007) and many hypotheses have been put forward to explain this pattern (see Dunn et al. 2009b). Ants do not respond to elevation per se; elevation is only a surrogate for a variety of factors that shape diversity gradients (Körner 2007; Dunn et al. 2009b). Nevertheless we introduced this environmental variable to examine how it is linked to other habitat features (e.g. litter, shrubs, etc.). Altitude at both of our field sites was correlated with steep slopes and bare rock areas, two environmental features that could negatively influence local ant species richness by limiting potential nest sites.

A case point in the results is cluster E. Although it is characterized by a high structural complexity and a low altitude it had the lowest species richness of all clusters. Habitat complexity is known to be an important factor driving species richness and community composition in ants (Lassau and Hochuli 2004). It has generally been found that species richness correlates positively with the complexity of the environment (Andersen 1986; McCoy and Bell 1991). Our results however do not seem to fit with this general observation. Clusters with greater ant diversity in our study indeed were simple from a structural point of view (see cluster B corresponding mainly to grassland areas). A similar result has been found by Lassau and Hochuli (2004) and Lassau et al. (2005) in their study of Australian ant communities. The two explanations provided by these authors to account for this result can also hold for our study. The first explanation is related to the locomotory behaviour of ants. The movements of ants are known to be more efficient and less constrained in simple than in complex environments (Kaspari and Weiser 1999). In simple environments, ants can move quickly, easily recruit nestmates, and defend/monopolize food sources efficiently against competitors and colonies can therefore develop more quickly. The second explanation is related to temperature. Sites with a dense canopy cover are likely to be cooler than sites exposed to direct sunlight. Since ants are thermophilic animals, a reduction in ground temperature could therefore reduce ant foraging activity and thus slow down or impede the development of ant colonies (Brown 1973; Cerda et al. 1998; Lessard et al. 2009).

Along with cluster B, Cluster D was also one of the clusters characterized by a relatively high species diversity. This is probably explained by the fact that it corresponds to transitional areas between hardwood forests and grassland. Ecotones are indeed known to have a positive effect on species richness (Risser 1995). An explanation for this is that an ecotone not only has its own characteristics (composition and structure) but also share the characteristics of both adjacent habitats (Risser 1995). Previous studies on "edge effect" however have led to conflicting results (e.g. in insects: Dauber and Wolters 2004) and the results of the present study would thus need to be confirmed.

SOM have already been applied successfully on ants to investigate the efficiency of sampling methods (Groc et al. 2007), the ecological impact of land use by Amerindians on ant diversity (Delabie et al. 2009), or the impact of ant-plant mutualism on the diversity of invertebrate communities (Céréghino et al. 2010). We show here that the use of SOM can in addition be useful to study the response of ant functional groups to environmental variables and land-cover features. This technique can explore large and complex datasets and thus can be used as an efficient tool in community ecology to define the characteristics of the ecological niche of each species (Groc et al. 2007; Céréghino et al. 2010). By using SOM we were able in addition to point out the sites of greater ant biodiversity in our study area. Environmental variables were used to characterize the landscape around each sampling points. However, information on coverage protected areas (see Hopton and Mayer 2006) could also have been introduced into the SOM. This could help to find out if clusters with high species richness overlap with protected areas (Hopton and Mayer 2006). This illustrates another important asset of SOM: because it provides information on the relationship between species distribution and habitat characteristics, SOM can be particularly helpful in targeting the areas in which to focus conservation effort.

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