

## **Environmental factors and diversity of epiphytic communities on the trunks of the Mediterranean beech forest in Hayedo de Montejo (Madrid, Spain)**

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**Abstract** – The influence of exposure on the trunk, the height above ground, the bryophytes cover and the location in the forest affects the epiphytic lichen diversity in the Natural Reserve of the Hayedo de Montejo, a relic beech forest in the Mediterranean Region located in the central part of the Iberian Peninsula. A total of 76 species collected throughout 32 relevés was analyzed by multivariate statistical techniques (Cluster analysis, PCA-GLMs, CCA). The humidity provided by the N-exposures and trunk bases allows the presence of many threatened lichens of the *Nephrometum resupinati* community. Also, the areas with N-exposure and close to water courses help the development of this community and higher lichen diversity. There are big microclimatic differences between the exposures in the epiphytic habitats and the different location into the forest, which is reflected in the composition and cover of the epiphytic lichen species. The detection of the microclimatic differences in the forests of the Mediterranean Region is important in the conservation of the threatened lichens and in its applications as bioindicators.

**Epiphytic lichens / *Lobarion* / *Nephrometum resupinati* / multivariate analysis / communities ecology / Spain**

**Resumen** – La orientación en el tronco, la altura al suelo, la cobertura de briófitos y la posición en el bosque afectan a la diversidad de líquenes epífitos en la Reserva Natural del Hayedo de Montejo, un bosque de hayas relicto situado en la Región Mediterránea en el centro de la Península Ibérica. Un total de 76 especies recolectadas a lo largo de 32 inventarios han sido analizadas por técnicas estadísticas multivariantes (Cluster analysis, PCA-GLMs, CCA). La humedad proporcionada por las orientaciones norte y zonas bajas de los troncos permite la supervivencia de los líquenes amenazados de la comunidad *Nephrometum resupinati*. También las áreas con orientación norte de ladera y próximas a los cursos de agua ayudan al mantenimiento de esta comunidad y dan una alta diversidad líquénica. Existen grandes diferencias microclimáticas entre las diferentes orientaciones en los hábitats epífitos y en la diferente localización dentro del bosque, lo que se refleja en la composición y cobertura de las especies de líquenes epífitos. La detección de las diferencias microclimáticas en estos bosques en la Región Mediterránea es de gran importancia en la biología de la conservación de los líquenes amenazados y en sus aplicaciones como bioindicadores.

**Líquenes epífitos / *Lobarion* / *Nephrometum resupinati* / análisis multivariante / ecología de comunidades / España.**

## INTRODUCTION

The study of the lichen communities contributes as important information for the conservation of the forests and its management (McCune, 2000; Will-Wolf, 2002). In spite of the great increase in the last years of the number of studies of lichen ecology thanks to its applications as bioindicators (Gombert *et al.*, 2005; Will-Wolf *et al.*, 2006), still exists a great difficulty in estimate the complex interactions between the species and the abiotic factors that take place in the lichen environment, e.g. historical factor (Giordani, 2006). In this sense, in the Mediterranean Region, aside from the studies of diversity and vegetation, few works had deal explicitly with multivariate analysis of factors influencing lichen distribution (Burgaz *et al.*, 1994a; Fuertes *et al.*, 1996; Fos, 1998; Zedda, 2002; Brunialti & Giordani, 2003; Casas-García *et al.*, 2005; Giordani, 2006; Belinchón *et al.*, 2007).

The use of lichens as bioindicators needs previous studies to establish the interactions of environmental parameters with the lichen vegetation to know the effects of some factor that affects to the stability of the ecosystem (Gomberth, 2005). These studies have a direct application in the experimental design of the works of bioindication (McCune, 1993; McCune *et al.*, 1997). In the case of studies related to atmospheric pollution in great areas (Herzig *et al.*, 1989; Herzig & Urech, 1991) the effect of ecological factors is negligible as long as sampling is well standardized (Nash, 1996).

*Lobarion pulmonariae* is a well-known epiphytic community and as well as their associations count on diverse works of ecology and conservation (Barkman, 1958; Rose, 1988 Burgaz *et al.*, 1994b; Zoller *et al.*, 1999). The association *Nephrometum resupinati* grows in the continental areas of the western Mediterranean basin and is coincident with the southern beech forest area in the Mediterranean Region of the Iberian Peninsula (Burgaz *et al.*, 1994a). Most of the species are threatened for the forest management and agricultural practices (Amo & Burgaz, 2005; Burgaz *et al.*, 1994a), and also Burgaz *et al.* (1994b) point out that moisture is a limit factor for this lichen community.

The aim of this paper is to describe the differences of species diversity and cover in the habitats of the trunk into the beech of the Hayedo de Montejo forest, aiming at the possible environmental factors that affect its distribution. With this analysis we try to clarify which factor must be considered in the design of later studies of conservation biology of this threatened lichens and its applications as bioindicators in the Mediterranean Region.

## MATERIAL AND METHODS

### Data recording

Relevés were carried out in the Hayedo de Montejo, a Natural Reserve of 122.5 ha., locate in the central part of the Iberian Peninsula (Fig. 1). It belongs to the Carpetano-Ibérico-Leonesa chorological province and the Ayllonense district (Rivas Martínez *et al.*, 1987). The area is located between 1250-1600 m altitude, the annual average temperature is 11°C, and annual average rainfall is 750 mm with two months of summer drought. It is included in the termoclimatic supra-submediterranean belt, with a submediterranean oceanic template bioclimate

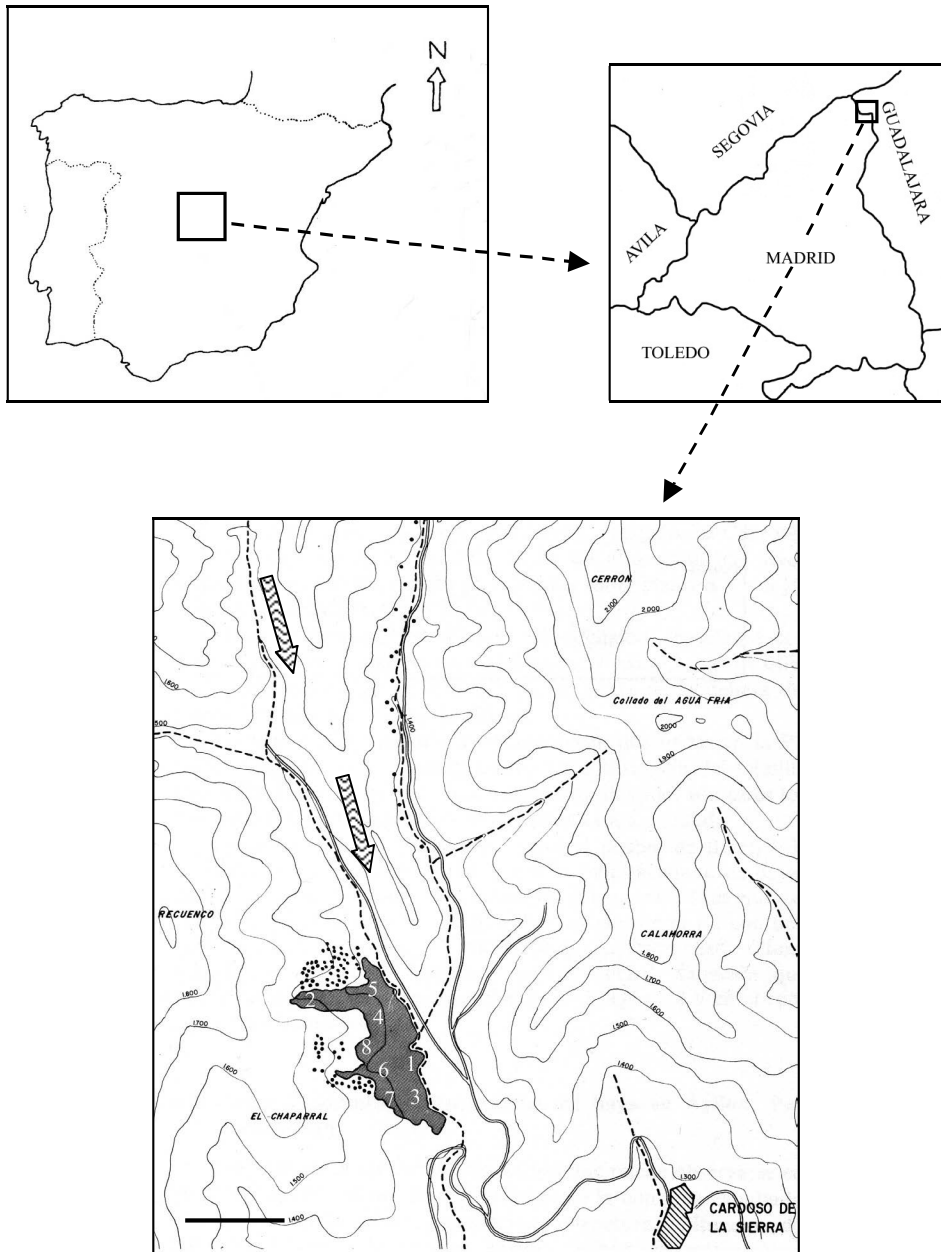


Fig. 1. Location of the Hayedo de Montejo Natural Reserve in the Madrid Province. Shaded area shows the area of the beech forest. Numbers show sampled areas. Broken line marks rivers course. Arrows show humid winds direction. Map following Hernández Bemejo & Sainz Ollero (1984). Scale bar = 1 km.

(Rivas Martínez *et al.*, 2002). The vascular vegetation belongs to the series *Galio rotundifolii-Fageto sylvaticae* (Rivas Martínez *et al.*, 2001). The mature stage is characterized by *Fagus sylvatica*, *Galium rotundifolium*, *Luzula lactea* and *Carex fragilis* mixed with isolates *Quercus pyrenaica* and *Q. petraea*. In the limits of the forest and in small open areas are heathland of *Halimio-Ericetum aragonensis*, marked up of *Adenocarpus hispanicus*, *A. complicatus* subsp. *commutatus*, *Genista florida*, *Calluna vulgaris*, *Erica arborea* and *E. australis* (Hernández Bermejo *et al.*, 1983; Rivas Martínez *et al.*, 1987). This forest represents one of the southern biogeographic limits of *Fagus sylvatica* in Europe and stand out because the Mediterranean climate of the place is influenced directly by damp winds (NW-SE direction) which increase the environmental moisture that compensates the deficit of the summer rain (Hernández Bermejo & Sainz Ollero, 1984).

The study is based on 32 relevés recorded on *Fagus sylvatica* trees with perimeters between 1.5-3 m, and includes 72 epiphytic lichen taxa. The relevés were recorded in S and N-exposures on the trunk, at 25 and 150 cm (DBH) above ground, and in homogenous zones being the dominant tree in the forest. Numbers in Fig. 1 show the location of sampled areas, with four relevés for area. Sampling was arranged according Braun-Blanquet's scale (1964). The original cover values were transformed according to the scale suggested by van der Maarel (1979).

### Data analysis

A cluster analysis to see the relationship between the relevés was carried out. A Principal Component Analysis (PCA) was used to reduce the species to a smaller number of factors, species with only one appearance were removed for this analysis. Original data (number of contacts) were normalised by means of square root transformation. The initial factorial solutions were rotated by the Varimax procedure (Nie *et al.*, 1975). Each PC score in relation to the independent variables studied (exposure, height and bryophytes abundance) and with the position of the relevés in the forests were analyzed. The interaction between both variables in the model was included. We used PC scores to define groups of species that appear together in relationship to the independent variables. CCA analysis was used to evaluate species and their arrangement to the environmental vectors. The analyses were carried out using STATISTICA 6.0 (Cluster analysis, PCA, GLM and ANOVA) and PC-ORD 4.0 (CCA) programs.

Nomenclature follows Llimona & Hladun (2001). Elix (1993) and Blanco *et al.* (2004) for parmelioid lichens. Samples were studied morphological and chemically in the laboratory.

## RESULTS

A clear trend of change in the diversity according to the exposure and height on the trunk can be observed (Fig. 2). **Cluster b1** embraces relevés carried out in the N-exposure of the trunk and **Cluster b2** includes the relevés recorded in the trunk base-N. **Cluster a1** and **a2** include the majority of the S-exposure relevés.

The relationship between exposure, height on the trunk and species diversity of each relevé is showed in the CCA analysis (Fig. 3). The Total

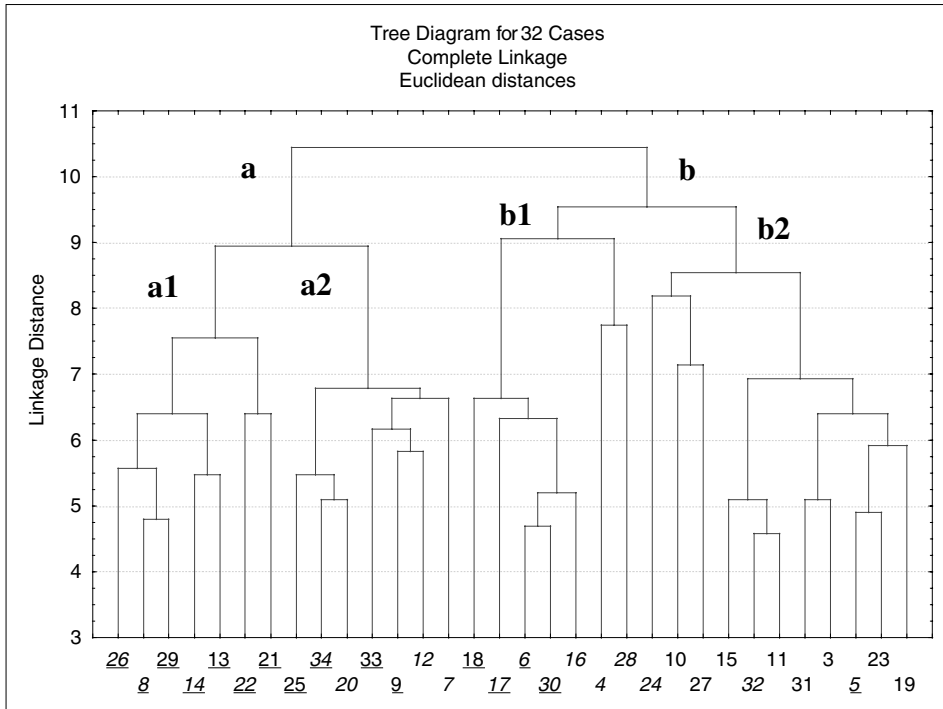


Fig. 2. Classification of the relevés made on diversity and species abundance. S-exposure relevés are underlined and N-exposure relevés without underlined. DBH relevés are italicized and base relevés without italicized.

Variation Explained (TVE) has values of 13.3, obtained in the CCA analysis is significant for the axis 1 ( $p < 0.001$ ) and significant in Pearson correlation with the environmental vars ( $p < 0.05$ ). The relevés included in the group-**A** are located in the trunk base-N, and we can link this first axis with a humidity gradient where the positive values of axis 1 represent the most humid conditions, e.g. close to the ground (height), less sunshine and therefore less leak of water (exposure). These environmental conditions are reflected by a higher bryophytes cover (Bryo. Cov.). The group-**B** is constituted with the majority of the trunk base-S relevés, and shows a decrease in bryophytes cover related to the lower humid conditions (negative values of axis 1). Owing to the sun exposure, the ground humidity would not be completely compensated by the loss of water. The influence of the N-W wet winds would add the humid condition of N-exposure (Fig. 1). The influence of the wind would have more clear influence in the topography exposure of the mountain inside.

The relevés 18 and 5 are characterized by the low species diversity and a high cover of less frequent species in the analysis such as *Ochrolechia pallescens* and *Collema furfuraceum*. The group-C includes the N and S-trunk DBH relevés with species influenced by dryer conditions and lower bryophytes cover (the lowest values in the axis 1).

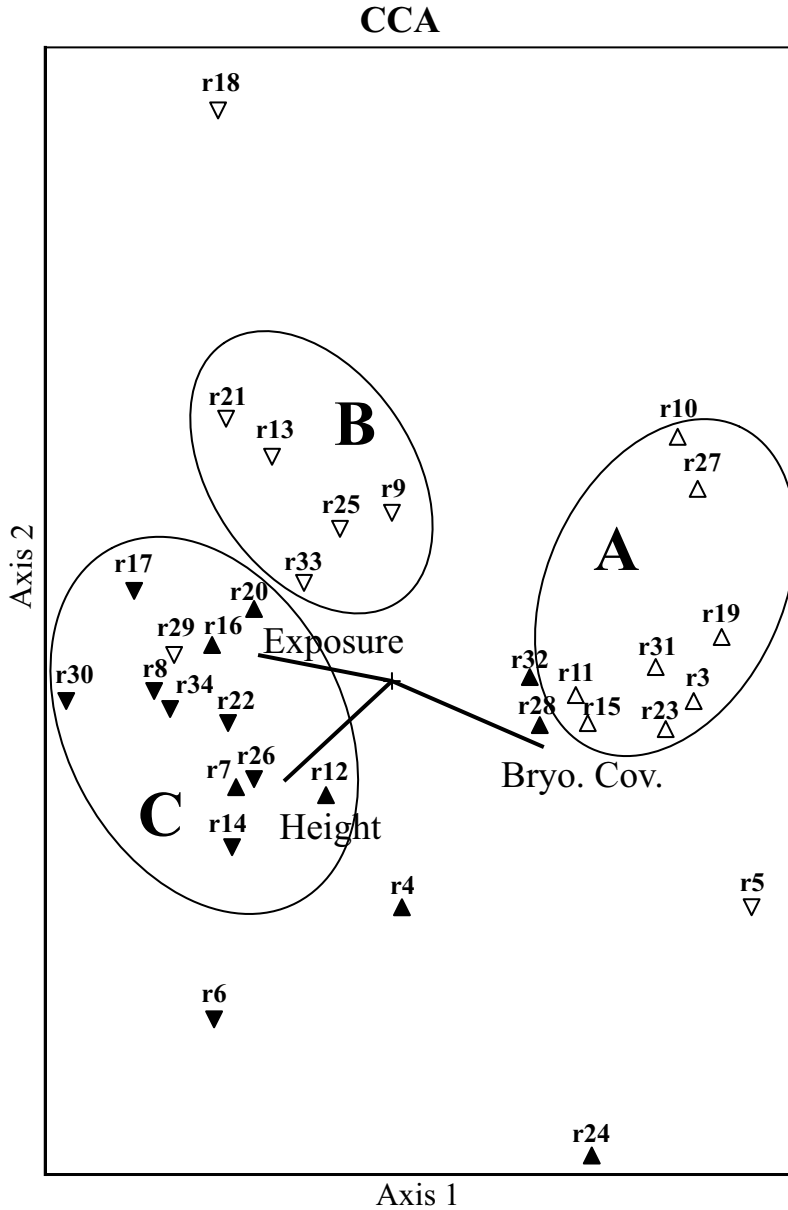


Fig. 3. CCA analysis of diversity in the relevés and enviromental vars.  $\triangle$  Relevés located in N-exposure and bases of the trunks.  $\blacktriangle$  Relevés in N-exposure and DBH.  $\nabla$  Relevés in S-exposure and bases of the trunks.  $\blacktriangledown$  Relevés in S-exposure and DBH.

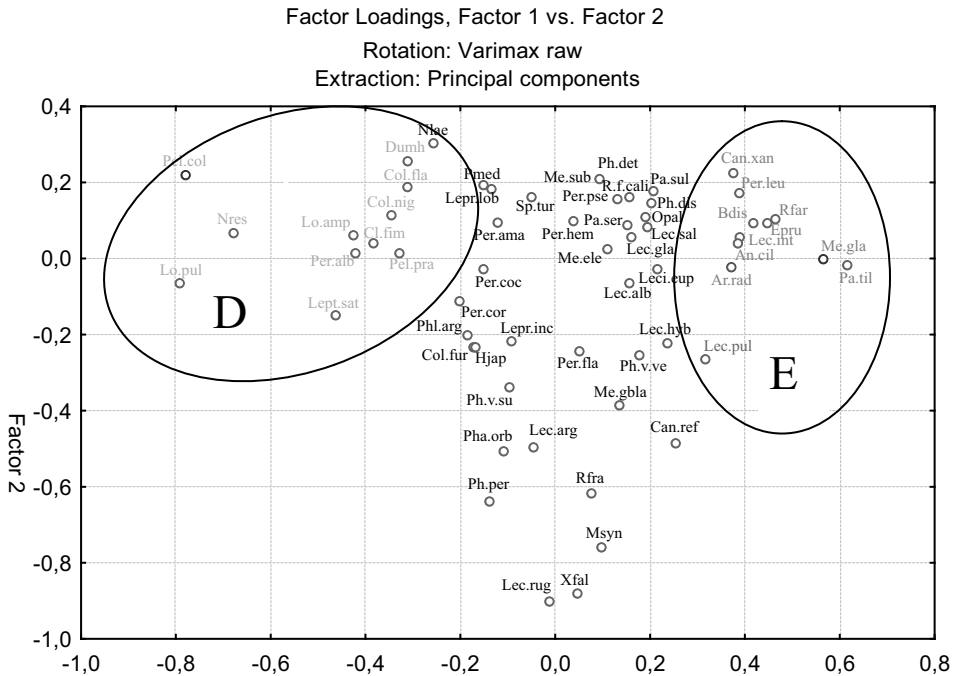


Fig. 4. PCA analysis of the species data. First factor show two groups of species: **D** – species of the community *Nephrometum resupinati*. **E** – species of the community *Orthotrichetum lyelii* subas. *parmelietosum subauriferae*.

The PCA analysis (Fig. 4) shows a group of species (**D**) which was described by traditional floristic methods as the climax association *Nephrometum resupinati* (Fuertes & Burgaz, 1989), constituted by large foliose or gelatinous thalli, mainly formed with cyanobacteria such as *Nephroma resupinatum*, *Peltigera collina*, *Leptogium saturninum*, *Collema flaccidum*, *C. nigrescens*, *Lobaria amplissima* (= *Dendriscoaulon umhausense*) or *Lobaria pulmonaria* with green algae, all of them considered skiophytic and hygrophytic elements. The namely group (**E**) described as the preclimax association *Orthotrichetum lyelii* subas. *parmelietosum subauriferae* (Fuertes *et al.*, 1996), is characterized by small foliose or crustose lichen and some fruticose thalli, all of them with green algae such as *Parmelina tiliacea*, *Melanelixia glabra*, *Evernia prunastri* and *Lecanora intumescens* which are considered photophytic and mesophytic elements. Nevertheless, in this group there are two different ecological contingents, because the binomial sun exposure and moisture is not easy to divide. The fruticose *Evernia prunastri* and *Ramalina farinacea* are considered aerohygrophytic elements while *Parmelia saxatilis* and *Physconia venusta* are considered photophytic and nitrophytic elements.

In the CCA analysis data (Fig. 5) more information of the two ecological contingents in DBH and trunk base-S relevés is shown. The group-**G** is characterized by species such as *Parmelia saxatilis*, *P. subaurifera*, *Heterodermia japonica*, *Pertusaria coccodes*, *P. hemisphaerica*, *P. coronata*, *P. flavida*, *Sphictrina*

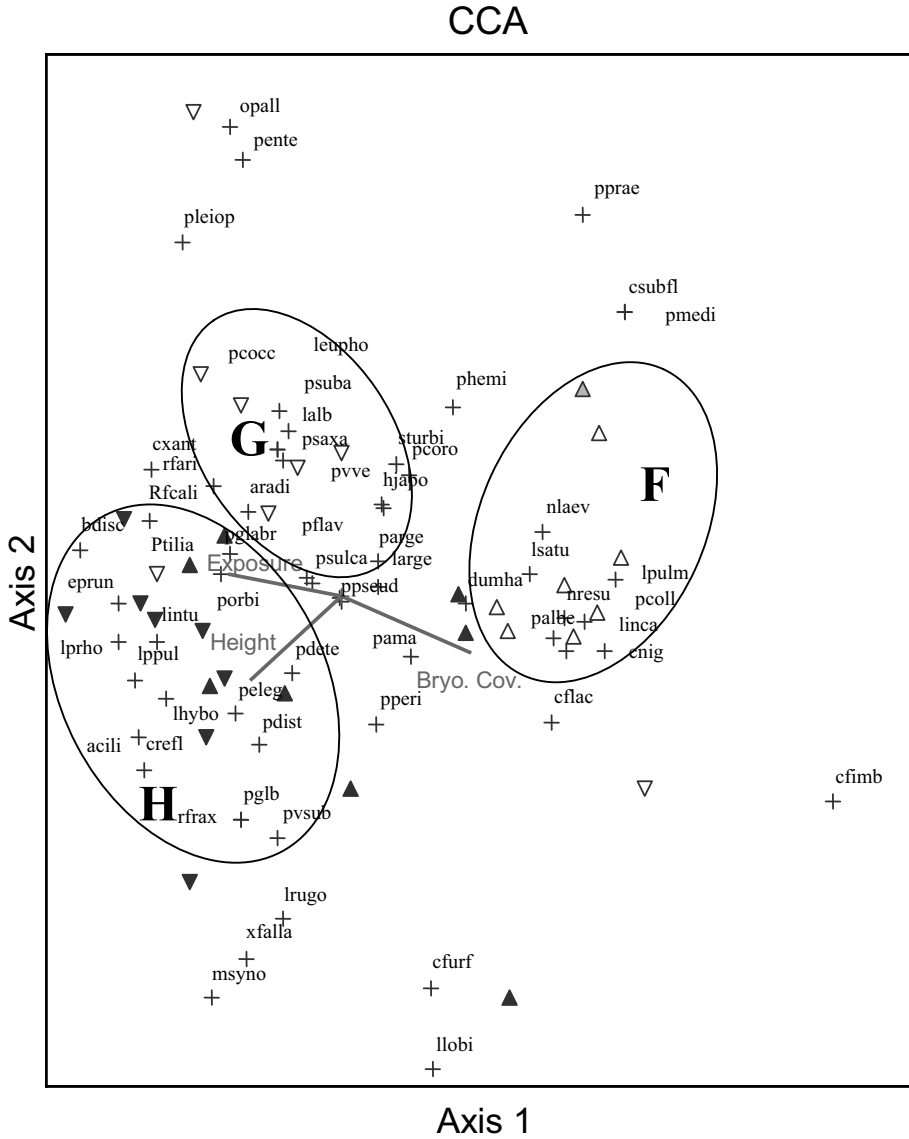


Fig. 5. CCA analysis showing the position of species, relevés and enviromental vars. △ Relevés located in N-exposure and bases of the trunks. ▲ Relevés in N-exposure and DBH. ▽ Relevés in S-exposure and bases of the trunks. ▼ Relevés in S-exposure and DBH.



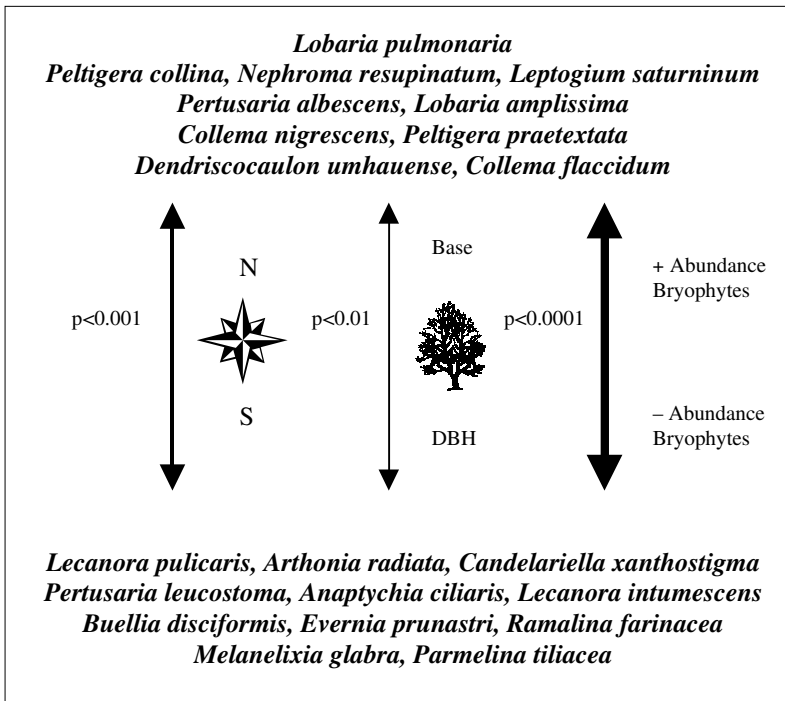


Fig. 6. Representation of results of GLM analysis of first factor PCA and the abiotic vars (exposure and height in the trunk and bryophytes cover).

*turbinata*, *Arthonia radiata* and *Physconia venusta*. These species need the moisture from the ground and sun exposure. The group-**H** is characterized by species such as *Anaptychia ciliaris*, *Lecanora hybocarpa*, *L. intumescens*, *L. pulicaris*, *Melanohalea elegantula*, *Parmelina tiliacea*, *Ramalina farinacea* and *R. fraxinea*. This group is constituted by aerohigrophytic and pioneer species. The group-**F** is constituted by *Lobaria pulmonaria*, *Nephroma resupinatum*, *N. laevigatum*, *Peltigera collina*, *Leptogium saturninum*, *Lobaria amplissima* (= *Dendriscoaulon umhausense*), *Collema nigrescens* and *C. flaccidum*, belongs to the group-**D** in the PCA analysis (Fig. 4).

The accumulated variance of the three first axis of the PCA analysis is 27.69 %. The first axis, related with the moisture and exposure, was analysed with the environmental vars by GLM (Fig. 6) where is shown the relationship between the moisture, exposure and the lichen distribution patterns in the trunk. The community of *Nephrometum resupinati* is well characterized in our relevés, but in the case of DBH and trunk base-S relevés, the species are grouped in the group-**E** (Fig. 4). The axes 2 and 3 have no correlation with the measured environmental vars (height, exposure and bryophytes cover) but have significant relationship with the location of the relevés in the forest (Fig. 7). The localities 1 and 5 are characterized by high cover of *Lobarion* species and high diversity, with few species only located at these areas.

The ANOVA of the lichen diversity is significant for the area in the forest where the relevés were carried out (Table 1). The relevés located in 1,

5 and 6 have high diversity (**Area A**) while the relevés in 2, 7 and 8 have low diversity (**Area B**) (Fig. 8). The subareas Ab and Ba are intermediate stages in the diversity gradient which exist between the bottom of the valley and the medium mountain slope.

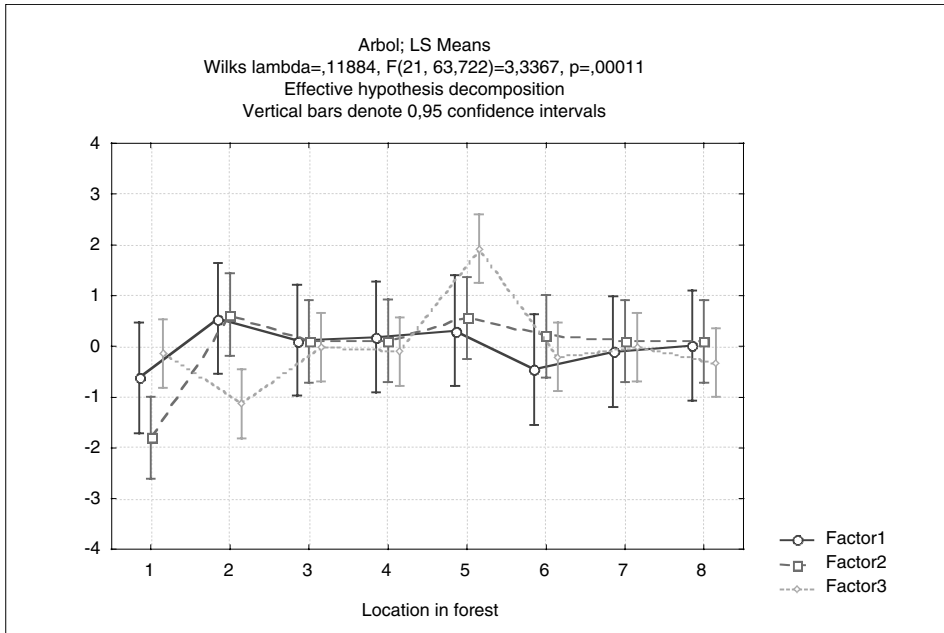


Fig. 7. Graph shows the result of the GLM analysis of the three first factors of PCA analysis and the position of the relevés in the beech forest.

Table 1. ANOVA of richness in the considered vars ( $p$  = significance level; \* =  $0.01 < p < 0.05$ ; ns = no significance). Right column show the area represented in Fig. 8-2.

	ANOVA ( $p$ )	Mean of richness ( $\pm$ Std. Dev.)	Area (Fig.8-2)
Exposure	ns	N- 12.06 ( $\pm$ 3.75) S-11.18 ( $\pm$ 3.72)	–
Height	ns	BASE- 11 ( $\pm$ 3.32) DBH- 12.25 ( $\pm$ 4.05)	–
Location of the relevé	*	1.- 13.75 ( $\pm$ 6.68) 2.- 10 ( $\pm$ 1.41) 3.- 11.75 ( $\pm$ 2.98) 4.- 10.25 ( $\pm$ 2.98) 5.- 15.25 ( $\pm$ 2.75) 6.- 14.25 ( $\pm$ 1.5) 7.- 9.75 ( $\pm$ 1.5) 8.- 8 ( $\pm$ 1.15)	B A B (Ba) A (Ab) B B A A

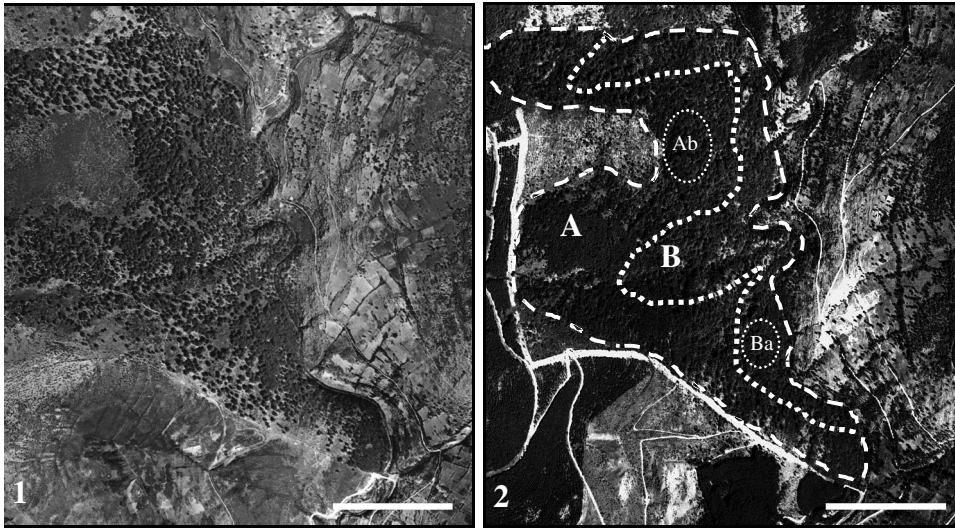


Fig. 8. Aerial photographs of the Hayedo de Montejo: 1. Photograph shows the forest in 1956. 2. Photograph shows the forest in 2001. Zone A includes the areas with less richness and zone B includes the area with major richness. Zones Ab y Bc show clear transition areas of the richness gradient. Scale bar = 500 m.

## DISCUSSION

Our results show the existence of at least two communities of lichens on the beech trunk in the Hayedo de Montejo. Against as we expected, these differences are higher between the communities growing on the trunk bases, that the differences between bases and the trunks DBH. Traditionally in the trunks are recognized mainly 10 different habitats (Hilitzer, 1928; Barkman, 1958). These authors already considered that the exposed zones and the sheltered zones of the different niches present a different microclimate.

According to our data of diversity and cover the trunk base-N matches a well-defined niche where the climax community *Nephrometum resupinati* take refuge. This habitat differs significantly to the rest of habitats (trunk base-S and DBH). The microclimatic differences between the trunk base-N and S exposures are higher than between the trunk base-S and the DBH habitats (Figs. 3 and 4). The present microclimatic conditions on the trunk bases-N allows the establishment or continuity of a climax community (Fig. 3, group **A**) as opposed to the rest of habitats that present pre-climax communities (Fig. 3, groups **B** and **C**). Between the trunk base-S and the DBH habitats also are differences, although of little rank. There are species characteristics to the trunk base-S (Fig. 5, group **G**). The DBH habitats have more elements in common. Nevertheless, the ascent of *Lobarion* species in exposure-N (relevés 1, 6, 7 and 8) or the presence in the trunk base-S (relevés 1 and 2) is frequent. On the other hand the abundance of *Anaptychia ciliaris* in open areas suggests the preference to the exposure-S.

According to our results, the loss of humidity due to the high light intensity in the trunk base-S could be the principal factor of the differences between the southern and northern trunk bases exposures. The relation between light intensity and drying was indicated by several authors (Barkman, 1958; Eversman *et al.*, 1987). The trunk base-N will give refuge against the high light intensity and the loss of humidity. This fact could be due to the open pasture woodland of the forest in last decades and whose effect still today continues in the age of the trees and in the differences in the forest canopy (Fig. 7). Even so, the general pattern of distribution of the environmental factors that determine the lichen microclimate in the “Hayedo de Montejo” does not correspond with the Barkman (1958) and Jonescu’s model (1970) for isolated trees. According to the species data, the adapted lichens to receive little light intensities are located in the trunk bases and not in high parts of the trunk next to the crown as those authors show. Probably this difference is due to the high light intensities reached in the Mediterranean Region. Although this fact cannot separately be analyzed of the rest of environmental factors that affect the lichen species (humidity, nutrients or pH) as also Jonescu (1970) show. In our case, the moisture of the ground in the beech forest plays a great role in sheltered microclimate conditions.

These results complemented the data of the ecology of the cryptogamic epiphytes and their communities in deciduous forests in Mediterranean Spain (Burgaz *et al.*, 1994a, b) because it shows the exposure like an important level of the limiting factor humidity. Also these results point to moisture of the ground as important factor for the damp conditions. Other factors, as the closeness to the river or the mountain slope facing north, cause a high moisture conditions. In this place the microclimate for the threatened species of the *Nephrometum resupinati* community is more favourable, with the presence of *Degelia plumbea*, *Pannaria conoplea* and old-grown forest lichen such as *Gyalecta ulmi*, *Sclerophora nivea*, *S. peronella* or *Lecanora quercicola* (Amo & Burgaz, 2004, 2005).

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