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Use of edaphic microinvertebrates to characterize soil quality in the forest Monte el Viejo, Palencia

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ABSTRACT

Conservation of increasingly degraded natural resources is an actual environmental priority and related to soils, they are the least focused on. This situation urges to develop fast and economic methodologies to evaluate the state of these resources in order to protect them. The aim of this study was to assess the soil quality of a Mediterranean forest located in Palencia (North Spain) using the QBS-ar index (Soil Biological Quality, adapted to arthropods). For this, three plots with different habitats in a managed ecosystem (secondary grassland, shrub community, and Mediterranean oak young forest) were selected and characterized in terms of stand composition and soil. Three soil samples were taken in each plot and the captured arthropods were identified. Moreover, six ecological indicators were calculated: QBS-ar, observed taxonomic richness (Sobs), diversity (Hsw), evenness (Ep), dominance, and specificity-fidelity. Variations of QBS-ar, observed taxonomic richness, diversity and evenness by by type of habitat and stand variables were evaluated computing generalized lineal models (GLMs). Analyses showed that QBS-ar, Sobs or Hsw did not vary between the different vegetation covers. Interestingly, taxonomic evenness showed significant variation either according to habitat -being higher in the shrubland- or by shrub height. This study addresses the relevance of belowground diversity in managed and restored environments.

Key words: soil bioindicators, arthropod mesofauna, QBS-ar, soil conservation, Mediterranean shrub importance.

RESUMEN

La conservación de los recursos naturales, cada vez más degradados, es una prioridad medioambiental actual y, en relación con los suelos, estos son a los que menos atención se presta. Esta situación urge a desarrollar metodologías rápidas y económicas para evaluar el estado de estos recursos con el fin de protegerlos. El objetivo de este estudio fue evaluar la calidad del suelo de un bosque mediterráneo situado en Palencia (norte de España) mediante el índice QBS-ar (Calidad Biológica del Suelo, adaptada a artrópodos). Para ello, se seleccionaron tres parcelas con diferentes hábitats sobre un entorno manejado (pastizal secundario, comunidad arbustiva y bosque joven de roble mediterráneo) y se caracterizaron en términos de composición de la masa y del suelo. Se tomaron tres muestras de suelo en cada parcela y se identificaron los artrópodos capturados. Además, se calcularon seis indicadores ecológicos: QBS-ar, riqueza taxonómica observada (Sobs), diversidad (Hsw), uniformidad (Ep), dominancia y especificidad-fidelidad. Las variaciones del QBS-ar, la riqueza taxonómica observada, la diversidad y la uniformidad según el tipo de hábitat y las variables del rodal se evaluaron mediante modelos lineales generalizados (GLM). Los análisis mostraron que QBS-ar, Sobs o Hsw no variaron entre las coberturas vegetales. Curiosamente, la uniformidad taxonómica mostró una variación significativa en función del hábitat -siendo mayor en el matorral- o de la altura de la cubierta arbustiva. Este estudio aborda la relevancia de la diversidad del suelo en entornos manejados y restaurados.

Palabras clave: bioindicadores del suelo, mesofauna de artrópodos, QBS-ar, conservación del suelo, importancia del matorral mediterráneo.

1.- INTRODUCTION

The society is becoming aware of the importance of environmental quality and the negative impact of diversity threats at global scale, such as climate change, habitat degradation or soil loss. The conservation of natural resources and services they provide (ecosystem services) is becoming a priority in the sustainable exploitation of these resources. Thus, the non-market benefits of the environment as economic, social or environmental goods and services are increasingly relevant in the management and planning of natural resources use, both in rural areas and at the urban interface (Jeffery, 2010). The study of the environment with a view to its sustainable management is becoming more widespread. Specifically, soils have not received most attention at global scale due to the lack of knowledge because they are made up of individuals that are practically invisible to the human eye and are difficult to know and quantify as part of the global biodiversity (Jeffery, 2010; Havlicek, 2012), they are overlooked (George *et al*., 2017). However, at least a quarter of the species on planet Earth live in the soil (European Comission, 2010) and it has been known for years that preserving the good soil health status is necessary for human prosperity. On the contrary, future generations will not have long-term resources (Jeffery, 2010; Nielsen, 2019).

Soils are dynamic, heterogeneous and biologically active systems that constitute a multifunctional natural resource. Soil biota is responsible for decomposing organic matter, thus regulating the movement of carbon and water; it also contributes to control plant pests and diseases by regulating microorganism composition in soil and root-interface, assimilating pollutants and serving as a source of natural resource as well as being nutrient reservoir for agriculture. Furthermore, soil provides products used as environmental, commercial or industrial new applications based on edaphic organisms (De Los Santos *et al*., 2018; Thiele-Bruhn, 2021; Gupta, 2022). Although most soils are composed of small organisms, they can hold up to 7 g dry weight/m^2 of soil fauna biomass (Heděnec *et al*., 2022). Terrestrial ecosystems are probably the most relevant natural resource for the survival and development of people. Due to this, Havlicek (2012) points out that little by little more importance to soil conservation at a scientific and political level is being given. Furthermore, more value is still given to the direct and indirect products obtained from the soil than to its intrinsic qualities and functions. Maintaining the heterogeneity in terms of complexity, diversity and connectivity of ecosystems at the landscape scale allows for greater biodiversity of soil dwelling, resulting in improved soil conditions and increased benefits (Nielsen, 2019). Jeffery (2010) mentioned that soils with higher biodiversity are more resistant to stress conditions and, in turn, are more resilient to disturbances. The first step in achieving soil conservation must necessarily begin with the knowledge of soil quality. To reach this, traditional studies of soil quality considered physical, chemical and biological-derived parameters as enzymatic activity or soil organic carbon (Bueis, 2018; Gómez, 2020), but new methods using animal communities as bioindicators have been successfully developed in soil quality determination by now (Parisi *et al*., 2005).

Soil fauna is a good bioindicator of the state of degradation of forest and agricultural soils as their populations vary according to the physical, chemical and biological components present. Based on this integrative approach, indices such as the so-called QBS-ar (Menta *et al*., 2018a; Menta *et al*., 2018b) have emerged, which are able to characterize soil quality by attending to the diversity of microarthropods that inhabit it according to their ability to adapt to the degraded environment. This index facilitates the diagnosis of the edaphic state and guides decision-making for its conservation and improvement. This methodology, which combines community ecology and agroforestry management, has been proved as useful and reliable in in different European agricultural ecosystems (Parisi *et al*., 2005), and has not ceased to attract attention in forest environments in the last years (Doran and Zeiss, 2000; Jeffery, 2010; Havlicek, 2012; Fusaro *et al*., 2018). The main advantages provided by the use of bioindicators of soil quality status are: (1) they allow standardizing methodologies and have reference values in addition to being able to compare with other natural, semi-natural or anthropized sites, (2) long-term monitoring is possible, (3) soil organisms respond earlier to disturbances than physical and chemical factors, (4) the results are easy to interpret and their values represent the state of the environment, (5) low economic investments, (6) establishment of limits that distinguish the current state of quality of the soils, as well as to know if the progress they are undergoing is positive or negative, and (7) it is complementary to other environmental indicators.

Ecological succession, understood as the development of degraded environments towards a climax community, can also be applied to soils. Thus, the edaphic community present in the soil changes according to the conditions of each state. Consequently, the implementation of this methodology in the evaluation of soils in forest environments, but at different stages, represents a new field of work in the context of environmental restoration, as well as in other related disciplines in the natural environment such as natural resource management or the promotion of diversity. The results obtained will allow us to know the variations due to the type of plant mass as well as their interaction, being able to draw conclusions about the quality of the soils evaluated and the ecological complexity of the invertebrate community studied.

1.1. Main faunistic groups in soil quality assessment

Vertical stratification in soil creates distinct layers inhabited by organisms adapted to specific conditions and occupying unique niches (Jeffery, 2010). Therefore, these animals are quite sensitive to changes primarily because many of them develop their entire life cycle in the soil and they are adapted to specific conditions. Soils harbor an extraordinary variety of organisms from all domains of life, within the animal kingdom; they are further classified according to their size in micro-, meso- and macrofauna (Figure 1) (Giller *et al*., 1997). According to Nielsen (2019) microfauna (*i.e*. body width >0.1 mm) lives mainly in water-filled soil micropores, mesofauna (*i.e*. body width between 0.1 and 2 mm) lives in air-filled soil macropores and macrofauna (*i.e*. body width >2 mm) usually lives in soil surface. Dominant groups belonging to microfauna are protozoans (Protozoa), nematodes (Nematoda), rotifers (Rotifera) and tardigrades (Tardigrada), as they have a limited impact on the soil structure, however, most microfauna have an important role as microbial feeders, so they are very relevant in the carbon and nutrient cycling of the soil. The mesofauna most important groups are mites (Acari), springtails (Collembola) mainly, and others less abundant such as proturans (Protura), diplurans (Diplura), symphylans (Symphyla), potworms (Enchytraeidae), and palpigrades (Palpigradi) among others. As microfauna, they essentially participate in carbon and nutrient cycling, but they also contribute to control pests by selective feeding and dispersal of microbes. Finally, the macrofauna group integrates more widely known animals like earthworms (Annelida), termites (Isoptera), ants (Formicidae), beetles (Coleoptera), millipedes (Diplopoda), and centipedes (Chilopoda). Part of the groups belonging to the mesofauna and macrofauna is common. Macrofauna helps in organic matter decomposition, nutrient cycling, water infiltration (soil engineering) and control of other groups density populations. Mesofauna and macrofauna are responsible for much of the litter decomposition.

This study was focused only on identification of the mesofauna organisms, specifically microarthropods (*i.e*. invertebrates with jointed legs, exoskeleton, segmented body and that cannot be seen without precision instruments). Although microarthropods are not able to modify the physical properties of the soil like other soil organisms *e.g*.: earthworms, ants (Hymenoptera; Formicidae) or beetles (Jeffery, 2010), they are found living with them on the surface and in the pores of the soil (Nielsen, 2019). They contribute in a very important way to maintain all the soil functions and to the environment, such as regulating nutrient cycling, organic matter decomposition or helping soil particle movement. They are good bioindicators of soil quality status, due to their abundance of species and the different forms in which they are part of the ecological community of each soil (Manu, 2019). Microarthropods include specimens belonging to three Subphylum: Chelicerata, Myriapoda and Hexapoda. Mites (Arachnida; Acari) and ants (Hexapoda; Hymenoptera) have a crucial role in soil renewal and nutrient cycling (Bernard, 2023), while other insect groups like Diptera or Coleoptera have a very varied range of functions. Both springtails (Hexapoda; Collembola) and mites are the most abundant taxonomic groups in the mesofauna that form part of soil. See Supplementary Material 1 for a schematic taxonomic classification.

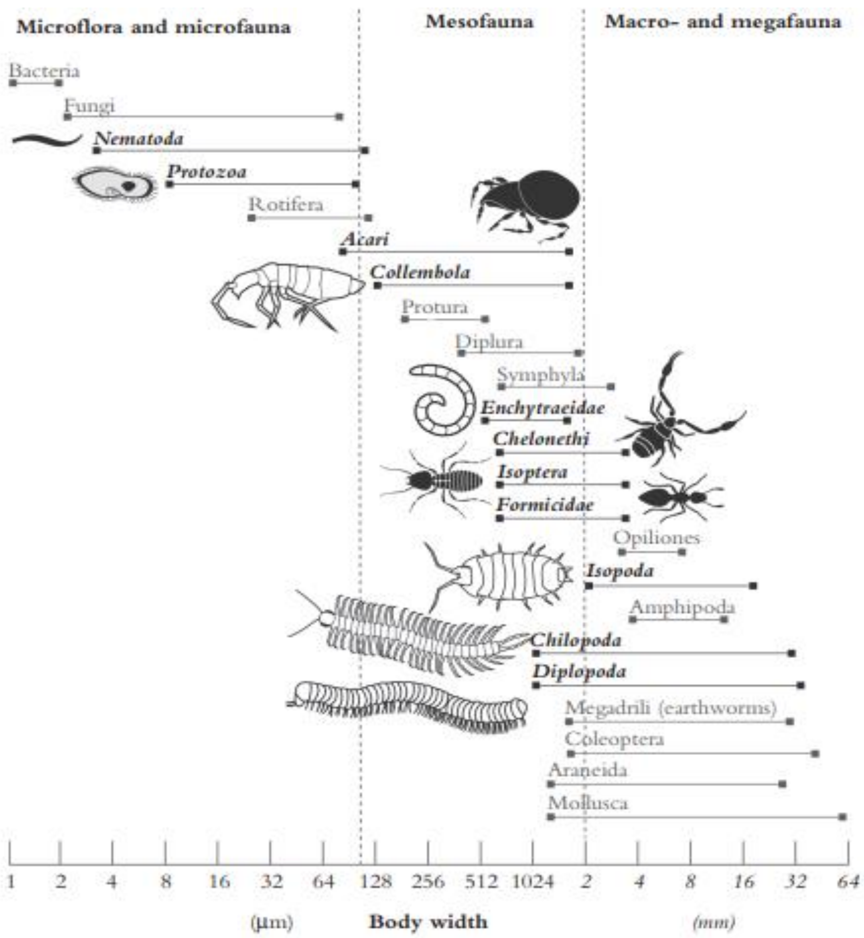

Figure 1. A schematic classification of soil biota based on body width (Nielsen, 2019).

2.-OBJECTIVES

The main aim of this research was to evaluate soil quality with QBS-ar index in a natural Mediterranean forest considering 3 different habitats (*i.e*. secondary grassland, shrub community, and Mediterranean oak young forest).

The specific objectives were (1) to characterize the microinvertebrate community from the point of view of community ecology, (2) to evaluate what the soils of higher quality are and how diverse are the hosted edaphic communities and (3) to investigate how quality soil and ecological indicators vary regarding the habitat.

3.- MATERIAL AND METHODS

3.1. Study site

The study area is located in the forest area "Monte el Viejo" in Palencia (North Spain; UTM coordinates: X:370265, Y:4646195; Figure 2), which covers around 1500 ha at an altitude of 865 m. This forest is mainly formed by stands of holm oak (*Quercus ilex* L.) and gall oak (*Quercus faginea* Lam.) with patches of aromatic shrubs such as thyme (*Thymus* sp. L.), sage (*Salvia* sp. L.), rosemary (*Salvia rosmarinus* Spenn.) or rockrose (*Cistus laurifolius* L.) that represent the continental Mediterranean forest understory. There are also mosses and lichens settled on the ground. From a geological point of view, Monte El Viejo belongs to the Duero River basin, it was formed by sediments originated in the Tertiary, where endorheic water masses evaporated. Their natural origin of the upper part is hard and compact calcareous soil made of limestone and marl rocks. The surface water currents are mostly absent in the area except for the Valdesanjuan creek in the southwest of Monte el Viejo.

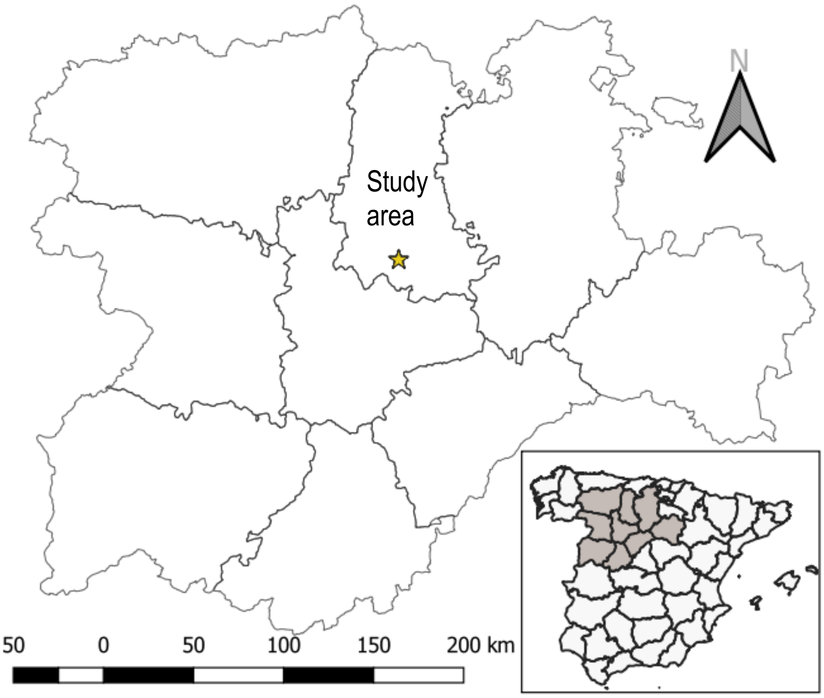

Figure 2. Location of the study area.

Regarding local climate, Köppen-Geiger climate classification is Csb (type C: average temperature of the coldest month between 0 ºC and 18 ºC; subtype s: existence of a dry season during the hottest months; subtype b: summer average temperature below 22 ºC and average monthly temperature above 10 ºC for more than 4 months in the same year (Rodríguez, 2021)), characterized by summer drought (73 mm average) and cold winters with little rainfall (134 mm average), the mean annual temperature is 11.1 ºC and the mean annual precipitation is around 471 mm (AEMET, 2023).

3.2. Plot selection and characterization

The methodology to select each soil sample followed a systematic sampling in which, each point was selected in equidistant straight lines. The plots have to be large enough to take all the samples, according to Menta *et al*. (2018) to collect at least 3 replicates of each sampling plot. The plots were separated 10 m from the edges of the roads and the distance from anthropic constructions was taken into account. Three samples were selected for each plot to achieve the greatest possible heterogeneity of edaphic microhabitats. Each sample was also taken several meters away from the previous one (10 m within the same plot). The minimal extension of plots was 40x20 m (Figure 3). The approximate average distance between plots was 140 m.

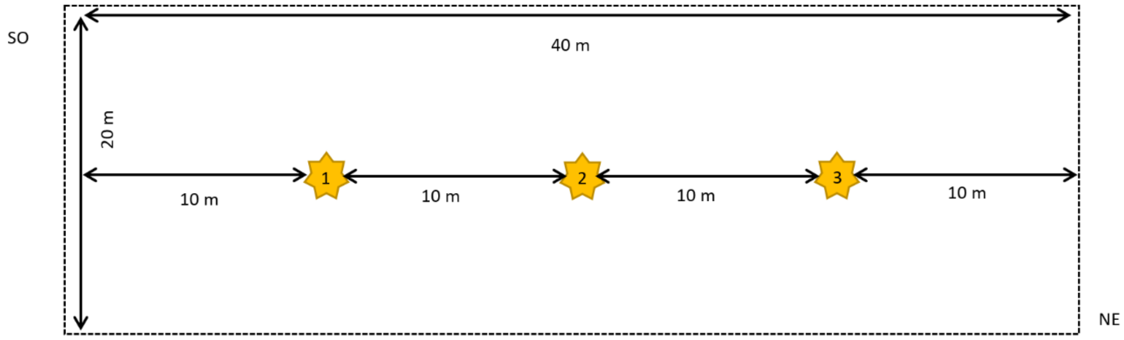

Figure 3. Sampling scheme for each plot. Stars represent the position in the plot for each soil sampling.

Sampling was carried out in April 2023, a period with moderate temperatures around 15 ºC at the beginning of the rainy season. The sampled plots were selected in order to represent different vegetation cover and habitat typology: (i) secondary grassland, (ii) shrub community and (iii) Mediterranean oak young forest. Grassland came from abandoned croplands located on very low slopes that over time have been spontaneously colonized by an herbaceous community; shrubland was a hillside community that lies midway between herbaceous layer at the bottom of the slope and a more settled holm oak (*Q. ilex*) woodland at higher altitudes. The forest was a semi-open forest stand of consolidated and low anthropized managed *Q. faginea* complemented with some regrowth, considered as shrub, and some grasses that have grown in a scattered manner (Supplementary Material 2).

Sampling points were used as reference for stand variable measurement (see below), and soil sampling (physical-chemical traits and mesofauna community characterization). Sampling plots were characterized assessing the following attributes: Sample point: first two letters of habitat and number of point Gr (grassland), Sh (shrubland) and Fo (Forest). Geolocation: X and Y coordinates in ETRS89 UTM 30N system obtained from the app QGIS (QGIS Development Team, 2023); altitude (A) at each sampling location obtained with the app QGIS through Digital Terrain Model (Source: IGN), Slope (S) of each point taken from the Digital Terrain Model (Instituto Geográfico Nacional, 2023) in QGIS; Orientation (O) in respect of north of the sampling points obtained from the app QGIS v 3.28.6.; Fraction of tree canopy cover (Fcc) estimated with the most recent LiDAR (Light Detection and Ranging) coverage layer (2nd coverage in 2019). More specifically, the plugins FUSION (McGaughey, 2020) and LASTOOLS (Isenburg, 2023) were used to calculate a grid surface (DTM) of 10x10 m cells over the study area and on this grid the forest canopy model was created using classes 2 (terrain), 3, 4 and 5 (vegetation) of the DTM that allows to know the percentage of vegetation cover; Average normal diameter of the 5 nearest trees to each sampling point (D) measured with a manual forceps; Average height of the 5 nearest trees (H) measured with Suunto PM-5/1520 PC Clinometer; Average height of the 5 nearest shrubs (Hs) measured with tape measure, and Herbaceous cover (Hc) expressed as live herbaceous cover on the substrate (%), taking into account only live species in direct contact with the soil. This last variable was visually estimated on 1 m^2 of soil made with metal rods 1 m long and facilitating its estimation with subdivisions as close as possible to the sampling point in a representative area (Supplementary Material 3).

3.3. Arthropods collection, identification, and QBS-ar index calculation

Soil mesofauna sampling followed the protocol described by Parisi *et al*. (2005) and Menta *et al*. (2018a) with minor modifications. Firstly, soil samples (10x10x10 cm) were taken after removing the litter layer with a garden trowel and separately introduced in labelled bags. Samples were brought to the laboratory the same day of the collection and placed separately in Berlese-Tüllgren extraction devices subjected to natural light period and constant heating for 7 days through 60 W incandescent infrared lamps (GiganTerra) placed 30 cm above each sample (Figure 4). In this process, the invertebrates tend to migrate deeper into the soil sample, to finally pass through a 2 mm light rectangular metal mesh and fall into the collecting vessel (a plastic tray where they were preserved in a mixture of 2/3 ethanol 96% v/v and 1/3 glycerol 99% v/v).

Taxonomic identification of edaphic mesofauna was performed after 7 days of samples incubation in Berlese-Tüllgren devices using taxonomical guides (Barrientos *et al.,* 2004). Briefly, the collector liquid from the trays was transferred to 4.50 cm diameter Petri dishes for subsequent extraction of the invertebrates to facilitate their observation under stereomicroscope. Identification was carried out with a MOTIC SMZ-168 SERIES binocular magnifying glass with magnifications between 7.5-50x and photographed with a Moticam 580 5.0 MP camera connected to the magnifying lens. With the aid of a Pasteur pipette and entomological tweezers, the extracted invertebrates were preserved in 2 mL Eppendorf tubes filled with 70% v/v ethanol and properly labelled. Finally, the liquid with the specimens was poured into zooplankton counting chambers (Aquatic BioTechnology S.L.) with five elongated grooves of 2 mL capacity each one, where the specimens were observed under binocular magnification to be classified using the main morphological characters and to be included in the taxonomical levels of Order or Class.

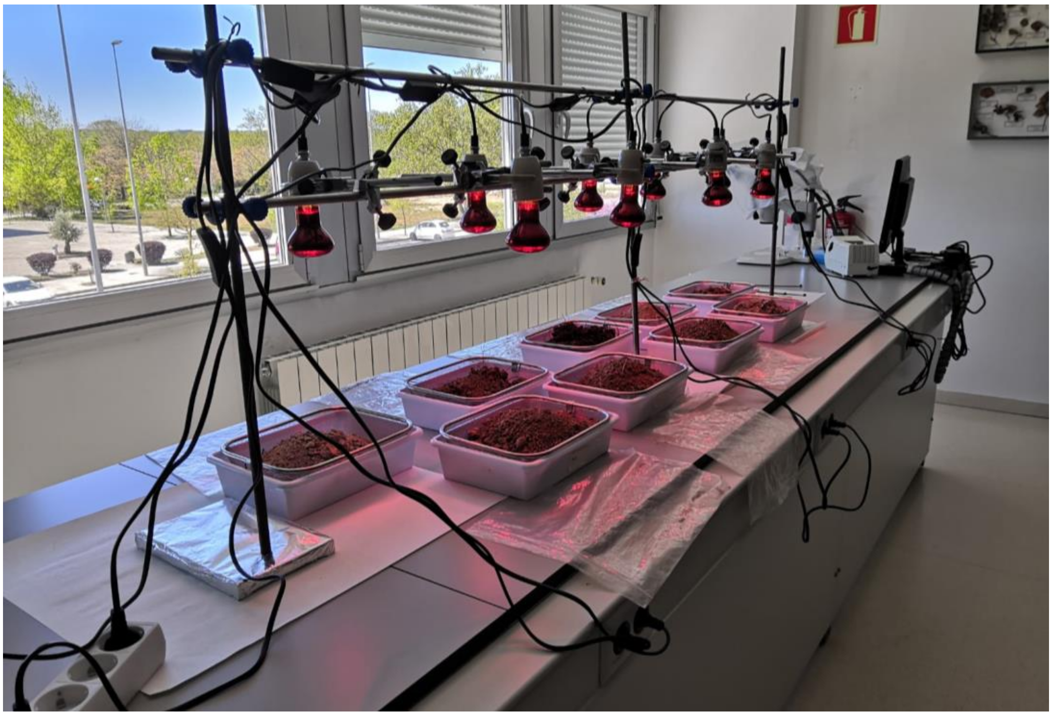

Figure 4. Berlese-Tüllgren devices used in this study.

Table 1. EMI value assigned to each found taxon.

Taxa	Description	EMI
Acari	-	20
Araneae	> 5 mm	1
	< 5 mm, scarcely pigmented	5
Pseudoscorpionida	-	20
Chilopoda	> 5 mm well-developed legs	10
	< 5 mm geophilomorphs	20
Diplopoda	> 5 mm	10
	< 5 mm Polixenida	20
Pauropoda	-	20
Protura	-	20
Collembola	> 2 mm	
	Developed appendages Pigmented Strong sclerotization	2
	Short appendages Pigmented Weak sclerotization	3
	Normally developed appendages Light coloration with transparent areas	4
	Normally developed appendages Transparent with colored areas	6
	Normally developed appendages Transparent with colored antennae	8
	Developed appendages Transparent	10
	Absent or short appendages Transparent	20
Psocoptera	-	1
Hemiptera	Epigeous or root-feeders	1
	Cicada larvae	10
Lepidoptera	Adult	1
	Larva	10
Diptera	Adult	1
	Larva	10
Hymenoptera	-	1
	Ant	5
Coleoptera (adults)	Epigeic forms	5
	4 parameters to be considered, 5 points are added for each one met: - Dimension (< 2mm) - Pigmentation (tan-brown colour) - Reduced or absence of eyes - Reduced or absence of wings	5
		10
		15
		20

The soil quality index QBS-ar was calculated according to the presence or absence of certain taxonomic groups of interest, assigning different scores (*i.e.* EMI values) to each group depending on their degree of adaptation to the edaphic environment (Menta *et al.*, 2018a). This value became 20 (maximum) in eu-edaphic taxa (Table 1), that is animals completely adapted to belowground lifestyle, and from here according to their specialization in the edaphic environment; lower values are assigned for hemi-edaphic organisms up to the minimum (1) in epi-edaphic ones (Mantoni *et al*., 2020). Whenever individuals from the same taxonomic group and different degree of adaptation to belowground lifestyle were recorded, the highest EMI value was used for index calculation (Parisi *et al*., 2005).

Finally, the QBS-ar was calculated as the addition of all the EMIs obtained for each sample. In the case of Collembola specimens and, due to the lack of match with the EMI classification developed by Menta *et al*. (2018a) another classification was elaborated by us in order to integrate the different morphological types found in the general EMI values (see Supplementary Material 4).

3.4. Soil characterization

Physical features of each sampled soil were evaluated in parallel to the collection of samples for mesofauna. First, the surface parameters *in situ* were taken by placing 4 steel rods of 1 m in length in a representative area forming a square of 1 m^2, and 2 metallic rods were placed in crosses forming 4 small squares in order to facilitate the estimations. Inside this square, we estimated surface stoniness (SuS); leaf litter weight (Lw) in a quadrant of the square, and leaf litter cover (Lc) as percentage of the soil surface covered by litter and dead matter.

The soil analysis was carried out with two types of samples to be examined in the laboratory: (i) unaltered sample; (ii) altered sample. The first samples were taken in the first 10 cm of the soil, randomly selected as one per each side (in 3 sides) of the fauna pit in order to have a more representative sample (3 per sample point, 9 per plot). These samples were extracted following the cylinder method by Blake (1986b) modified introducing the sampling cylinder by the side with the cutting edge a few cm from the surface so as not to catch the top layer of organic matter horizon. It was pushed with a mallet and a piece of wood, being careful with the thick elements, the soil that protrudes from the cylinder was leveled with spatula or field knife and the sample was stored in a closed labelled bag.

In this kind of samples (*i.e*. unaltered ones) two variables were calculated: apparent or bulk density (Bd) including the pore space according to Blake (1986b) modified, and humidity (Hu) as quantity of water retained in soil including gravitational water (water in large cracks and pores that moves within the soil by gravity draining freely), capillary water (retained in the capillaries of the soil and moves by cohesive forces or capillary effects), and hygroscopic water (that which remains attached to the soil particles by adhesion forces) calculated following MAPA (1994) with modifications.

Moreover, the altered samples (ii) were taken in the first 10 cm using a small shovel obtaining quite disturbed soil together with the coarse elements, but without the top layer of leaf litter (1 per sample point, 3 per plot). After the preparation of this samples in laboratory to obtain fine soil (explained in Supplementary Material 5) the following variables were obtained: soil stoniness (SoS) as percentage of stoniness of the soil profiles taken in altered sample; texture (T) as percentage abundance of soil particle size composition in terms of sand (< 2 mm y > 0.05 mm), silt (<0.05 µm y > $2x10^{-3}$ mm) and clay (< $2x10^{-3}$ mm) that make up the soil according to USDA (1999). Texture was determined by qualitative method (feel procedure) taking a fine soil sample and moistening with a few drops of water to form a pliable mass, assessing malleability of soil by forming balls and flat ribbons with the hands and feeling its softness or roughness (USDA, 1999). Real density (Rd) was also calculated as total mass of the solid particles with respect to the total volume they occupy following the pycnometer method modified by Blake (1986a), in addition, porosity (P) was measured as soil volume occupied by the pores by difference between Bd and Rd. Finally, organic matter (OM), the organic fraction of the soil that includes plant and animal residues in different states of decomposition, was calculated with modified method of Schulte (1996) (see abbreviations in Supplementary Material 3).

3.5. Statistical analysis

All the statistical analyses were performed in R programming environment (R Core Team, 2023).

3.5.1. Ecological description of edaphic community

Ecological calculations as described by Muñoz-Adalia (2022) were performed. To characterize the edaphic mesofauna community, the number of detected taxonomic groups (taxonomic richness; Sobs) was firstly calculated. Then Shannon-Weaver diversity index (Hsw) and Pielou´s evenness index (Ep) were calculated using the package “vegan” (Oksanen *et al*., 2022) in R. Dominance of each taxonomic group in each plot was determined using the Camargo index (Camargo, 1993). Finally, the IndVal index (Dufrêne & Legendre, 1997) was calculated in order to detect whether any taxonomic group played a role as specialist of sampled habitats (*i.e.* grassland, shrubland or forest). This index is defined as the product of site fidelity and specificity of each detected taxonomic group. The index was calculated using the package “indicspecies” (De Cáceres and Legendre, 2022) in R with 999 permutations.

3.5.2. Principal Component Analysis (PCA)

To study whether stand and soil characterization variables influenced the calculated response variables (ecological indicators), statistical modelling was used (see section 3.5.3). In order to reduce the number of variables to be considered in model computation, a principal component analysis (PCA) was performed using “FactoMineR” (Lê *et al*., 2023) and “factoextra” (Kassambara and Mundt, 2020) packages in R. Firstly, categorical variables were transformed into binary variables by factor level (0/1), then PCA was performed to detect tendencies that cluster sampling plot therefore highlighting variables that could explain the variability found in field data. The percentage of explained variability was evaluated by dimension, and the relative contribution of each variable in explained variability of the corresponding dimension were also investigated using the mentioned packages of R.

3.5.3. Generalized Linear Models (GLM)

Before beginning to evaluate whether ecological indicators are altered by soil and stand variables, possible bias caused by sample weight was evaluated computing a generalized linear model (GLM; Gaussian distribution of errors) in R where the four main ecological indicators (*i.e*. Sobs, Hsw, EP, and QBS-ar) were used as response variable and sample weight as explicative one. Then, variations in ecological indicators Sobs, Hsw, EP, and QBS-ar by plot (that is kind of habitat) by environmental variables were performed by computing GLMs. Eighty-four models were computed (21 per indicator). Accordingly, the ecological indexes and the QBS-ar were taken as response variables individually, the factor habitat and variables with those previously selected in PCA (Height of shrubs -Hs-, tree canopy cover -Fcc- and height of trees -H-), were used as explanatory variables alone, in combination and as interaction whenever it retained biological meaning. Computed models were compared according to Akaike's Information Criteria (AIC) (Anderson, 2008) using the “AICcmodavg” package (Mazerolle, 2023) among them and against a null model (that is a simple model were the ecological indicator was explained only by the factor habitat). Final selected model of each indicator was the most parsimonious one (that is, the model that explains more data variability with less number of variables), which had the lowest AIC value. Selected model fitness quality was evaluated using the package "DHARMa" (Hartig and Lohse, 2022) and predicted values were plotted with "effects" package (Fox, 2022). When significant differences were detected by an explicative factor, LSD Fisher test was performed using "agricolae" package in R (Mendiburu, 2021) in order to get pairwise comparisons as *post-hoc* test.

4.- RESULTS

4.1. Characterization of the sampled plots and soil

The description of plots (Supplementary Material 6) revealed a wider variability of stand and soil parameters between habitats. The grassland stood out for its low slope compared to the other two plots. In addition, tree size, although young, was larger in the forest than in the shrubland. Shrubs in shrubland were noticeably larger in size than forest. In terms of edaphic characteristics, it is noteworthy that Lc (Litter cover), Hu (Humidity), P (Porosity) and OM (Organic matter) values resulted considerably higher in the forest.

4.2. Relevance of each sampled faunistic taxa

The following is a brief description of the mesofauna taxa found in this study and their importance in maintaining soil functions. Moreover, the taxonomic classification of the sampled animals as well as some morphological and ecological description is showed in Supplementary Material 7.

Order Acari

Mites are widely recognized as the most abundant and diverse group of invertebrates found in soils, their density population can range from thousands to hundreds of thousands (Pacek *et al*., 2020). Most of them inhabit leaf litter, although the more mobile predators often live on plant stems waiting for their next animal prey to appear, or to feed on the plants themselves (Bernard, 2023). According to Iraola (1998) they are found in all ecosystems all around the world, probably because of their small size which facilitates their dispersion by the wind and by phoresy.

Due to their wide dispersion and abundance, they also serve as food for other faunal groups (Nielsen, 2019) such as beetles, spiders or pseudoscorpions. In addition to controlling the populations of their prey and serving as food for other animals, they play an important role in shredding organic matter and decomposing it. Iraola's study (2001) shows that microphytophages feed on hyphae and release enzymes that break down organic materials releasing nutrients and energy used by other organisms in the ecosystem. They also play an important role by providing pelleted excrement containing nutrients and compounds that can serve as food for other organisms, thus contributing to improve soil chemical conditions. Their high density, species richness, their susceptibility to changes in soil conditions, especially those belonging to the Order Mesostigmata and Oribatida, makes them good bioindicators (Manu, 2019).

Order Araneae

Their abundance is not as high as that of other groups such as springtails and mites, being between 50 to 150 individuals per m^2 (Marc, 1999; Menta and Remelli, 2020). Spiders are generalists, predators of animals helped with their venomous glands to poison preys (Rahmani *et a*l., 2014), especially other invertebrates like myriapods or insects and present in all habitats (Melic *et al*., 2015). Due to their low mobility, they are highly adapted to the specific habitat in which they live (Bernard, 2023) and can be a good soil quality bioindicator because of their sensitivity to soil changes (Melic *et al*., 2015; Menta and Remelli, 2020).

Order Pseudoscorpionida

They prey on collembolans, mites, diplurans, ants, dipterans, psocopterans, larvae and other small arthropods, mainly alive, but also dead. Their density can range from 50-300 individuals/m^2 in litter and soil depending on environmental conditions (Liebke *et al.*, 2021).

Its use as a bioindicator of soil quality is not widespread (Menta and Remelli, 2020) due to the difficulties in their species identification and the little knowledge of the relationship of the occurrence of a species to site conditions. Nevertheless, they can be used as an indicator of the environmental conditions of the place where they live, especially in arid areas (Zaragoza, 2015) because they seem to be quite sensitive to disturbances.

Class Chilopoda

Centipedes are carnivorous generalists as stated by Menta and Remelli (2020), responsible for regulating the populations of organic matter decomposers, so prey mainly on invertebrates despite they also feed on small vertebrates.

Bernard (2023) points out that the specimens belonging to the order geophilomorphs reach densities of about 1.000 individuals/m^2, although other groups are less dense.

In their research, Menta and Remelli (2020) concluded that centipedes have been used as indicators of soil quality, due to the variation in their populations according to the densities of their preys and changes in the pH of the environment in which they live.

Class Diplopoda

Their populations tend to concentrate in forest environments (Bernard, 2023), where they can reach densities of over 1.000 individuals per m^2 (Melic, 2015b), as leaf litter is abundant and the environment is more stable (Menta and Remelli, 2020).

Millipedes are not pioneering migrants; their presence gradually increases when a location becomes stable enough after a disturbance. Thanks to this characteristic, the quality of restored spaces can be distinguished based on the amount of time elapsed. As a result of this, Menta and Remelli (2020) think diplopods act as good soil quality indicators.

Class Pauropoda

They live in the soil, where their legs are not used for digging but for moving through the pores. They are generally abundant in the upper soil layers, provided that temperature, humidity, porosity, and the amount of decomposing organic matter are suitable. They may seek better thermal and humidity conditions at deeper soil depths.

The population density of these organisms is usually very low, often less than 100 individuals/m^2 (Coleman *et al*., 1996; Jeffery, 2010). As a result, they often represent undisturbed soils, acting as bioindicators of soil quality (Menta and Remelli, 2020).

Order Protura

They are generally found in upper soil horizons, especially in organic horizons in agricultural or forest environments with certain humidity and abundant organic matter in decomposition coming from leaves and wood (Bernard, 2023). They can reach 40.000 individuals per m^2, these large numbers occur mainly in secondary oak forests, due to abundant ectomycorrhizal fungi (Krauß and Funke, 1995), but according to Melic (2015a) the range of abundance is lower, down to 18.000 individuals per m^2.

They are good soil indicators according to Menta and Remelli (2020) and Pass and Urban (2011) because their communities are very sensitive to physicochemical and are related to the presence of mycorrhizal fungi.

Order Collembola

Springtails are more frequent in the soil, being able to reach 200.000 individuals/m^2 (Mayvan *et al*., 2022), both in the superficial and deeper layers (Baquero and Jordana, 2015). In these layers, they play their main role in the soil environment through the decomposition of organic matter and the formation of soil structure at the microscopic level (Arbea and Blasco-Zumeta, 2001), in addition to controlling the populations of the individuals on which they feed.

As Rusek (1998) indicates, their roles in ecosystem are to degrade the organic matter, feed on leaf tissue or animal excrement, provide the largest amount of droppings that form part of the soil humus by arthropods, distribute microbiota propagules in soil and provide nutrients in the middle and long periods of their life cycles. The same author also indicated that collembolans are not normally soil engineering making tunnels, but in the first stages of succession, they are responsible for modifying soil microstructure.

They serve as bioindicators of soil quality according to Baquero and Jordana (2015) due to their modifications in the populations that may be due to climate change or others of anthropic origin like intensive use of the soil through intensive management is carried out (Rusek, 1998).

Order Psocoptera

Their presence indicates places where organic matter decomposition occurs since their food source consists of microorganisms that appear because of degradation. Fungal spores are also part of their diet. They tend to choose more open forests (Baz, 1991). In fact, they are often difficult to find in more humid areas. They contribute to the trophic network as prey for other arthropods such as hemipterans, beetles, or spiders. According to Socarrás (2013), their population density is around 2.000 individuals per m^2 in agricultural ecosystems.

They are considered the first colonizers of bare areas in the early stages of ecological succession, contributing to soil development. However, some species within this group are associated with mature and stable forests, which can indicate the conservation status of the environment. (Socarrás, 2013; Alexander *et al*., 2015).

Order Hemiptera

In this study only Heteroptera specimens were collected. According to Fauvel (1999), Heteroptera are sensitive to chemical changes in the soil and feed on various organisms, making their presence a good indicator of soil quality (De la Mora-Estrada *et al.,* 2017).

Order Lepidoptera

Some studies may suggest that using Lepidoptera as bioindicators are less relevant due to their lack of correlations with the presence of other taxa (Gerlach, 2013). Nevertheless, the natural soil and its associated elements, such as tree trunks or leaf litter, serve as shelter and food for the larvae, where they will later pupate. Their partial life cycle in the soil creates favorable conditions for them to detect any soil disturbance caused by environmental or anthropogenic changes (Legal, 2023). Furthermore, thanks to their habitat specificity they can clearly act as bioindicators (Legal *et al*., 2020), even indicating the slightest alterations in those habitats.

Order Diptera

According to the study by Frouz (1999), soil dipterans can be classified into groups: a) those that complete their life cycle in the soil, for which they have undergone morphological adaptations such as wing loss; b) those that only spend their larval stage in the soil, and c) those that only pupate in the soil. The density of dipteran populations in soils varies greatly, ranging from a few tens to thousands of individuals per m^2 (Frouz, 1999).

For a long time, dipterans were not used as bioindicators due to difficulties in identification and their complex ecology. However, they are highly useful as they are widely spread and indispensable in their interactions with other living organisms due to the varied roles they play within the ecosystem, including their role as prey, as noted by Frouz (1999) and Menta and Remelli (2020). Also, larvae are very sensitive to soil changes and to chemical products and contamination.

Order Hymenoptera

The study only obtained ants specimens, After Menta and Remelli (2020), groups like ants are essential in ecosystem engineering, as they modify the soil by increasing porosity, drainage, and available nutrients.

This species richness also allows them to serve as indicators of ecosystem quality based on the presence or absence of higher trophic level groups, which indicate suitable environments for the development of their prey. Specifically, Menta and Remelli, (2020) points out thar ants are good indicators because their populations change depending on habitat management, successional stage, and physicochemical soil conditions.

Order Coleoptera

Like other insects, beetles play a significant role in ecosystems, establishing symbiotic relationships with other organisms such as fungi, nematodes, mites, and microorganisms (Alonso-Zarazaga, 2015). Together with spiders, they are the most representative predators in soil fauna (Bernard, 2023).

According to Menta and Remelli (2020), beetles are widely used as bioindicators of soil quality due to their sensitivity to environmental changes caused by various factors. Their wide range of habitats and their diet, which includes various organisms, provide valuable information to assess the natural conditions based on their presence or absence in different environments.

4.3. Ecological indexes of the sampled faunistic soil community

Soil invertebrate surveyed after Berlese-Tüllgren incubation resulted in 14 taxonomic groups identified (Figure 5): Acari were the most abundant (44%), followed by Hymenoptera (34%) (epigeic ants) and Collembola (15%); and other taxa were much less abundant (<2%). In addition to the above taxa, nematodes and annelids were also found, but they are not considered in the index and therefore were excluded from the study.

The groups that were always present in all habitats were: mites (33.24 ± 6.26 individuals/kg sample, mean and standard error), springtails (11.97 ± 4.34 individuals/kg sample) and beetles (mean 1.33 ± 0.50 individuals/kg sample). However, springtails with a higher degree of adaptation to edaphic life (unpigmented with poorly developed appendages) were especially abundant in the forest (23.70 ± 9,14 individuals/kg sample). On the other hand, Hymenoptera, although also abundant with a mean abundance of 18.60 ± 9.98 individuals/kg sample, were almost exclusive to more opened areas (grassland and shrubland). On the contrary, proturans and the only Lepidoptera specimen appeared only in the forest. Associations between the other groups and habitats are not too homogeneous either. Specimens of Psocoptera were found in the grassland and in the forest. Pseudoscorpions, chilopods and dipterans were found both, in the forest and in the shrub habitat. In addition, Hemiptera were the only group recorded in both grassland and shrubland. Finally, the least represented groups (mean abundance per group ≤ 0.15 individuals/kg sample) were Araneae, Pauropoda and Diplopoda with one specimen per group, the first two were only found in the grassland, while the Diplopoda was in the shrubland. Absolute abundances are shown in Table 2.

Table 2. Absolute abundances by taxonomic groups (in bold dominant taxa according to the Camargo´s index) and IndVal index [*: p-value < 0.05 (999 permutations)]. Sample point/Habitat tipology: Gr/G: Grassland; Sh/S: Shrubland; Fo/F: Forest.

Sample point	Habitat typology	Acari	Araneae	Pseudoescorpionida	Chilopoda	Diplopoda	Pauropoda	Protura	Collembola	Psocoptera	Hemiptera	Lepidoptera	Diptera	Hymenoptera	Coleoptera
Gr1	G	**68**	1	0	0	0	1	0	22	1	2	0	0	**109**	3
Gr2		**12**	0	0	0	0	0	0	1	0	0	0	0	**19**	0
Gr3		**25**	0	0	0	0	0	0	5	1	2	0	0	**3**	3
Sh1	S	**10**	0	1	0	1	0	0	2	0	2	0	1	**10**	1
Sh2		**18**	0	0	0	0	0	0	1	0	0	0	0	**11**	0
Sh3		**13**	0	0	1	0	0	0	6	0	0	0	0	**16**	1
Fo1	F	**20**	0	2	0	0	0	0	**20**	0	0	1	1	0	0
Fo2		**25**	0	0	1	0	0	0	7	0	0	0	1	0	2
Fo3		**28**	0	0	0	0	0	3	**8**	1	0	0	1	1	0
IndVal		1.00	33.33	33.33	33.33	33.33	33.33	33.33	1.00	50.00	50.00	33.33	75.00	99.41*	55.56

The IndVal index revealed ants as specialists in secondary grassland and shrubland habitats (1.00; p-value=0.01) (Table 2). Finally, once the groups were known, the EMI value contributed by each group in the QBS-ar was assigned (Table 3).

Table 3. Maximum EMI assigned to each found taxa per sample plot. Gr: Grassland; Sh: Shrubland; Fo: Forest.

Taxa	Gr1	Gr2	Gr3	Sh1	Sh2	Sh3	Fo1	Fo2	Fo3
Acari	20	20	20	20	20	20	20	20	20
Araneae	5	0	0	0	0	0	0	0	0
Pseudoscorpionida	0	0	0	20	0	0	0	0	20
Chilopoda	0	0	0	0	0	20	0	20	0
Diplopoda	0	0	0	20	0	0	0	0	0
Pauropoda	20	0	0	0	0	0	0	0	0
Protura	0	0	0	0	0	0	20	0	0
Collembola	20	3	20	20	10	20	20	20	20
Psocoptera	1	0	1	0	0	0	1	0	0
Hemiptera	1	0	1	1	0	0	0	0	0
Lepidoptera	0	0	0	0	0	0	0	0	10
Diptera	0	0	0	10	0	0	1	10	10
Hymenoptera	5	5	5	5	5	5	5	0	0
Coleoptera	5	0	5	5	0	5	0	5	0

Figure 5. Taxonomic groups found in soil samples. A1, A2: Acari, B: Araneae, C: Pseudoscorpionida, D: Chilopoda, E: Diplopoda, F: Pauropoda, G: Protura, H: Collembola, I: Psocoptera, J: Hemiptera, K: Lepidoptera (larva), L: Diptera, M: Hymenoptera; N: Coleoptera.

4.4. Effect of environmental variables in ecological indicators

PCA revealed a first dimension which explained 97.3% of the variability (Figure 6) followed by a dimension 2 with explained 1.7% of variability. First dimension included H (trees height) as the variable that represents 98.2% of this variability explaining the rest of considered variable less than 1.8% of variability each one. On the other hand, dimension 2 explained only 1.7%, where the greatest contribution of Hs (shrub height) is 73.9% and 8.3% of Fcc (tree canopy cover). Accordingly, the variables H, Fcc and Hs were further investigated as explicative variables in data modelling (see below).

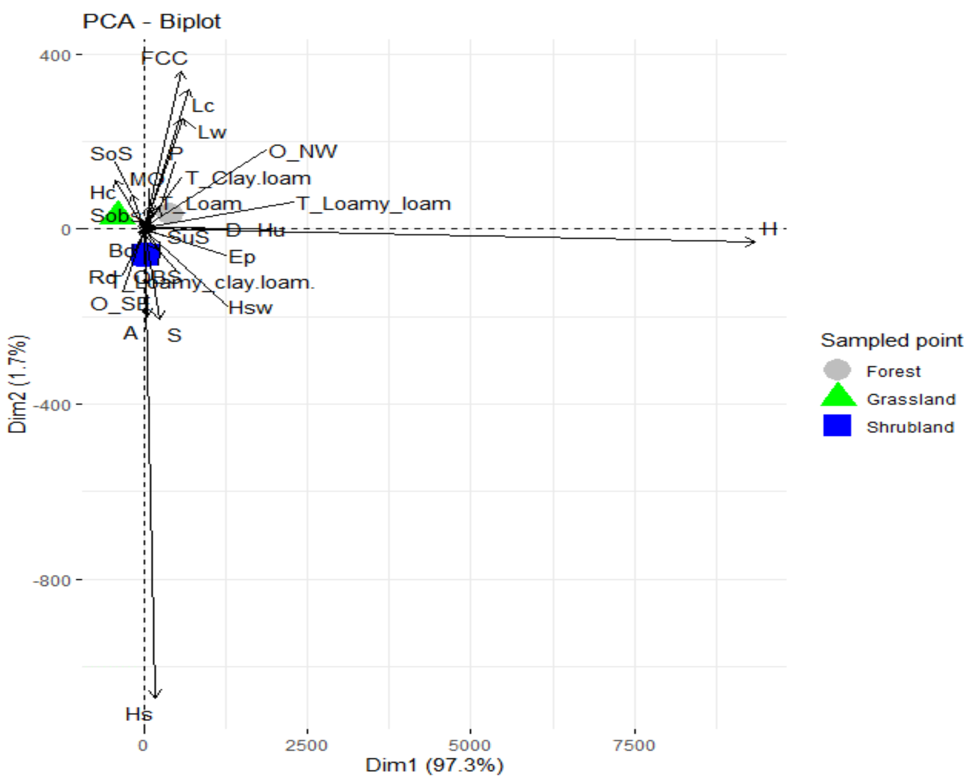

Figure 6. PCA of plot and soil characterization.

The sampled soil mesofauna communities were characterized through four ecological indicators: QBS-ar, Sobs (observed richness), Hsw (diversity) and Ep (evenness). Variations of these indicators by habitat, H, Hs and Fcc variables were evaluated computing GLMs. Previously, variations of the indicators by sample weight did not reveal any significant variation of QBS-ar, Sobs, Hsw and Ep (p-value ≥ 0.57 in all cases) therefore excluding this variable from models computation.

The highest QBS-ar value was found in a shrubland plot (QBS-ar = 101) and the lowest (QBS-ar = 28) in grassland. The greatest variation of this value between sampling points within the same habitat has occurred in the shrubland with a difference of 66 points, while the forest is the most uniform across all its plots with a range of only 8 points. The highest mean QBS-ar was in forest habitat, decreases slightly in the shrubland and the grassland shows a remarkable decrease. Despite these variations, the GLM QBS-ar showed no significant differences between habitats (p-value = 0.28). Moreover, tree height-H-, tree canopy cover -Fcc- and shrub height-Hs- did not show significant effect in QBS-ar variation (p-value ≥ 0.28).

The mean values of Sobs and Hsw were very close among the different habitats (5.44 ± 0.60 and 1.08 ± 0.08 points, respectively), reaching their maximum and minimum values both in grassland and shrubland, being quite homogeneous in forest. The variables H, Hs and Fcc did not significantly affect Sobs and Hsw (p-value ≥ 0.50 in all cases).

Significant differences of this variable were neither found between habitats (p-value = 0.72 for Sobs and p-value≥0.50 for Hsw).

Ep values, although similar between sampling points and between habitats, have shown the highest range of values in the grassland. The highest mean value by habitat was that of shrubland, 0.75 points higher than that of forest, the lowest one. According to GLM 2 variables significantly affected the variation of evenness (Ep): shrub height (Hs; model M76; p-value =0.01) and type of habitat (model M64; p-value = 0.01). Model M76 resulted the most parsimonious according to AIC (Table 4) showing lower value than null model and M64. According to the model M76, the value of Ep increases as Hs increases (Figure 6). The latter model (null model) was selected for further analysis because of it considered habitat type despite it showed higher AIC than M76. *Post-hoc* analysis of M64 revealed significant differences in Ep between grassland and forests (Figure 7).

Table 4. Results of models (GLMs) describing variation of ecological indicator Ep (Pielou's evenness) with Hs (average height of the 5 nearest shrubs), Fcc (Fraction of tree canopy cover) and habitat (type of habitat in each plot). Df: degrees of freedom, AIC: Akaike's Information Criteria, ΔAIC: the difference in AIC score between the best model and the model being compared. Selected model in bold.

Ecological indicator	Model	Description	Df	Loglike	Deviance	AIC	ΔAIC
Ep	**M76**	**Ep ~ Hs**	**8**	**13.34**	**26.68**	**-15.87**	**0.00**
	M79	Ep ~ Fcc	8	11.28	23.6	-11.77	4.11
	M64 (null model)	Ep ~ Habitat	8	14.79	29.58	-11.57	4.30

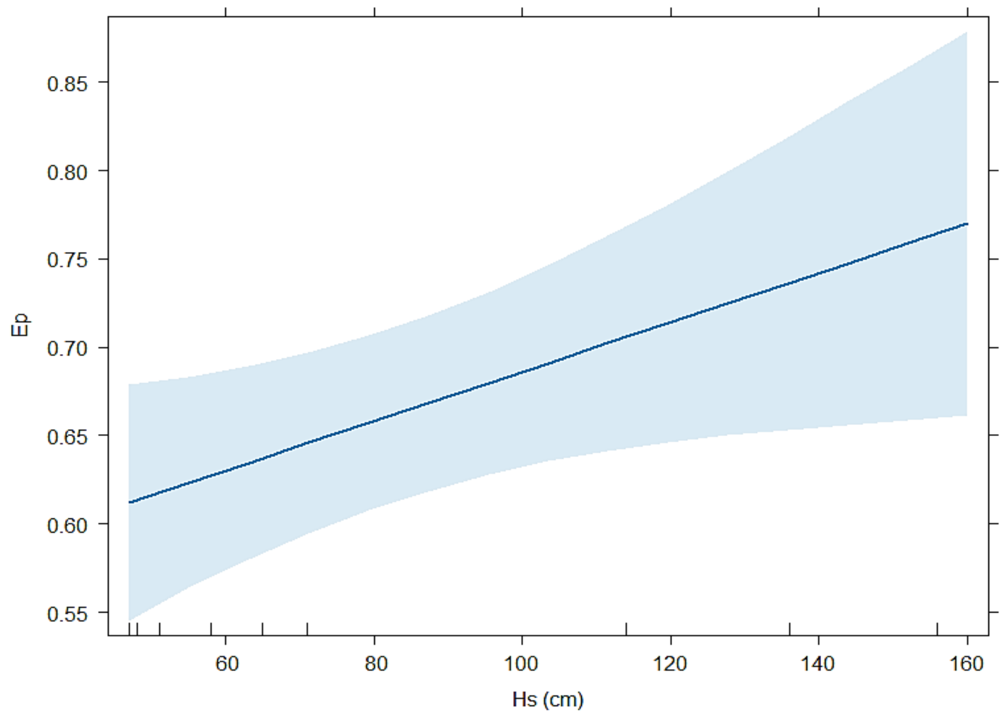

Figure 7. Predicted values of Model M76. Hs: average height of the 5 nearest shrubs. Ep: Pielou's evenness. Shaded area represents 95% confidence interval of predicted values.

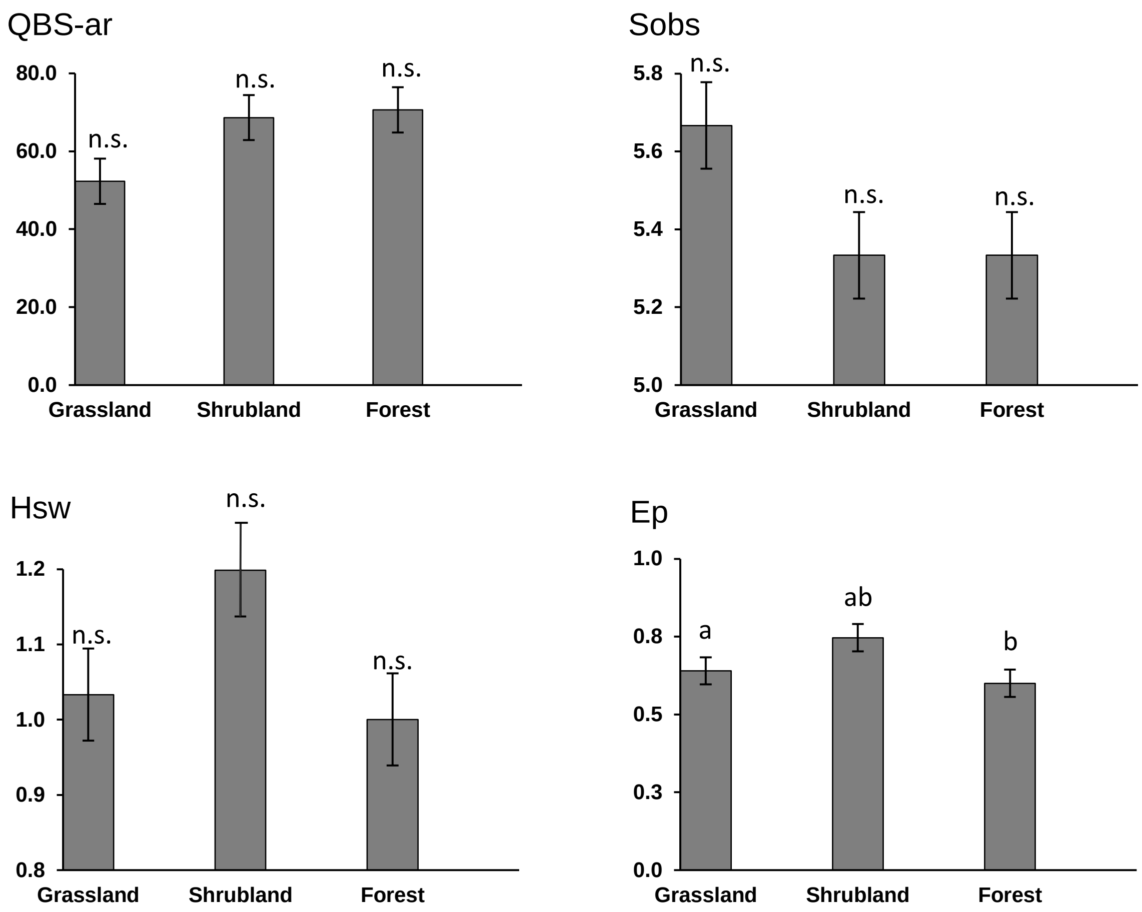

Figure 8. Observed values of ecological indicators by habitat. Mean values and standard error are shown. Sobs: Observed taxonomic richness, Hsw: Shannon-Weaver´s diversity index, Ep: Pielou´s evenness index. Small letters (a-b) denote significant differences according to LSD Fisher *post-hoc* analysis (p-value < 0.05). n.s.: not significant differences.

5.- DISCUSSION

In this study, we evaluated the soil microarthropod community in the forest Monte el Viejo regarding three habitats from secondary grassland and shrubland to *Quercus faginea* young forest. Almost half of the recorded microarthropods were mites (Acari); what may be due to his generalist habits; which allows them to be numerous and well distributed in different habitats and environmental conditions (Evans, 2013). The groups that are usually more widely distributed in soils (mites and springtails) were among the most abundant along with ants, and the other groups were much less represented. Jeffery (2010) observed that mites and springtails represent 75% of all forest soils arthropods, within these two groups the abundance is variable according to the physicochemical conditions and site management. Curiously, the second most numerous group was not the springtails as reported in other studies (Mussury *et al*., 2002; Behan-Pelletier, 2011), but ants (Hymenoptera), highly abundant and also dominant in the grassland and shrub according to Camargo index. This fact has been previously reported by Menta and Remelli (2020) since they point out that Hymenoptera can become as dominant as mites and springtails. Rusek (1998) and Rocha de Lima *et al*. (2017) that found that springtails were more abundant in more stable spaces with a greater variety and density of plants mentioned a possible explanation for this low Collembola density. Contrary to this, our study showed that the plots with the greatest variety of vegetation in terms of structure and plant species, the shrubland, had the lowest number of springtails. This apparent underrepresentation may be due to Collembola species are more associated to either closed or open spaces (Szigeti *et al*., 2022), medium coverage environments are less frequented by these arthropods. This distribution pattern, as reported by Petersen (1982), is typical in temperate climates. Nonetheless, despite their abundance, mites and springtails seem to be ubiquitous (specially collembolans in grassland and forest), while Hymenoptera is restricted to shrub and herbaceous areas according to the IndVal index. Schultheiss *et al*. (2022) and Menta and Remelli (2020) reported that the abundance of ants is not so high in rather dry forests, which may be due to thermal factors that affect the species according to their heat tolerance regime (Sánchez-García, 2022), which, in this case, would be conditioned by vegetation cover. On the other hand, other authors show that ants are more abundant in anthropized environments and that is why they tend to appear in the early stages of succession (Rocha de Lima *et al*., 2017). These authors also mentioned that ants are usually more frequent in forest edges than in its interior.

Chauvat *et al*. (2011), de Groot *et al*. (2016), and Nielsen (2019) pointed out that springtails or mites tend to increase their diversity and abundance as ecological succession advances. In this study, we did not observe this in shrubland despite it could be considered as an intermediate stage in progression of grassland-forest. The statement mentioned by Jeffery (2010), Andrés *et al*. (2011) and Nielsen (2019) claims that it does seem these two groups are more abundant in soils with high organic matter (OM) content. This edaphic variable did not show any effect in the indicators evaluated here, nevertheless, the abundance of these taxonomic groups in our grassland and shrubland plots was scarce with lower levels of OM and forest show higher abundances in both cases corresponding with higher levels of OM. In consequence, the abundance of the three most common groups in the edaphic fauna (Acari, Collembola and Hymenoptera) seems to be highly variable according to habitat conditions.

Regarding other less represented taxonomic groups*, i.e.* Myriapoda (Chilopoda, Diplopoda and Pauropoda), although the abundance has been very low, all the specimens have contributed a maximum value of EMI (20). In the case of proturans and, according to Nielsen (2019), these animals live in soils rich in organic matter, that is clearly remarkable because they only appeared in the unique sample point with highest OM. In addition, dry conditions recorded in the sampled year could be also affecting the amount of these groups, usually adapted to humid environments. Strikingly, we observed some specimens of Psocoptera in the grassland plots, where the light reaching the ground is much higher, as well in the forest plot with low tree canopy cover (40 %). These insects seem to be more frequent in areas without vegetation cover (Socarrás, 2013) and in forests leaf litter (Bernard, 2023). In the same way, Heteroptera have only been found in grassland and shrubland; apparently species of this group prefer open spaces and can colonize new environments rapidly, especially natural herbaceous canopies (Fauvel, 1999; Frank & Künzle, 2006; De la Mora-Estrada *et al*., 2017), in either undisturbed or disturbed areas (Bröring and Wiegleb, 2005). On the other hand, Diptera specimens appeared more frequently in the forest, a fact that agrees with Frouz (1999), which mentions a greater abundance and diversity of species in forests and grasslands. We have found this relationship for the forest, also coinciding with the values of higher organic matter and humidity. Unlike the aforementioned groups, Coleoptera appeared in all habitats, but they were noticeably more abundant in the grassland. Taboada *et al*. (2011) reported that beetles are mostly abundant in semi-natural grasslands since they have more open spaces, with better temperature, light and low humidity compared to forests.

Abandoned agricultural fields, which harbor hardly any arthropods, once converted to pasture increase microarthropod biodiversity over time to the values of natural grasslands, but some management such as controlled grazing and mowing tends to increase these values (Siepel, 1996; van Eekeren *et al*., 2022). Since the abundance of microarthropods in the pasture is the highest in this study (6.04 ± 2.73 individuals/kg sample), it is possible that before the sampled crop field was abandoned, it was used for cattle grazing. In addition, with respect to pesticides, soils with previous agricultural use usually have residues of these chemicals that can influence arthropod populations. In this case, the abundance of microarthropods in the secondary pasture is slightly higher than those in young oak forest (5.82 ± 2.16 /kg sample) where grazing, a plausible use of agro-chemicals, is expected to have been low, so it is seen that they have not been negatively affected by human uses the past short term. The (scarce) use of the *Quercus faginea* stand and its young age are probably the reason why the edaphic community has not developed as much as in stable forests without long-term management and differences between habitats are less marked.

The QBS-ar index obtained from the taxa present in the samples seem to increase as the woody plant community develops (grassland<shrubland<forest), being notably higher in the young forest (Figure 6). Note that the EMI values are higher in the shrubland and more markedly in the forest with a lower number of taxa, *i.e*. more specialized soil groups, while in the grassland, despite having a greater diversity of taxa, its EMI values are lower. Forested and grassland habitats seem to harbor higher numbers of arthropods, although it does not imply high differences in ecological indices and QBS-ar. The higher QBS-ar value in the shrub and forest can be partially explained because, although the number of taxa has not been significantly higher in these areas, the taxa present have more specific soil-living conditions, (Parisi *et al*., 2005; Menta *et al*., 2018a). It is precisely in the forest where the maximum EMI value is repeated most often in Acari, Pseudoscorpionida, Chilopoda, Protura and Collembola, thus supporting this interpretation of the index. Nonetheless, the soil is a highly variable medium throughout the seasons and the years (Gardi, 2008; Havlicek, 2012), so this index is influenced by seasonal changes and therefore longer sampling time is required to see changes (Tabaglio *et al*., 2009). A greater abundance of microarthropods does not always mean that the quality of the soil environment is better; the quality depends on the adaptations of the fauna it hosts.

Community evenness has proven to be a key indicator in understanding the differences between the three habitats. Although the abundance of arthropods observed in the shrub habitat was the lowest, it appears that taxa are more evenly distributed. In this, the dominant taxa (*i.e.* Acari and Hymenoptera) are much less abundant than in the other studied habitats, while the richness remained similar, implying that the infrequent groups are better represented. Specifically, all sampling points in shrubland have the highest evenness values. Our results revealed that these differences can be attributed to the height of the shrubland (*i.e.* Hs, Table 3), which was considerably higher in the shrubland compared to the herbaceous and forest stands. This disparity in vertical structure has direct implications for the distribution and relative abundance of species in each habitat, thus conditioning the dominance of the groups.

In relation to soil physical and chemical parameters, the biodiversity in this study does not respond to changes as plant biomass increases and therefore nutrient availability and organic matter increase as well. This event does not corroborate what was pointed out by Nielsen (2019), that the ecological succession towards the climax stage progressively increases these parameters and with it species richness. Moreover, Jeffery (2010) mentioned that forests are generally the system with the highest levels of soil biodiversity but taking into account that the community is influenced by many other factors. Remarkable similarities have been observed between grassland and shrubland, where Pielou´s evenness index resulted relatively high due to a more uniform distribution of biomass among species. In the same way, similarities are found between shrub and young forest, although in the latter, the evenness may decrease slightly due to the presence of tree species and the difficulties of growth that this implies for the incipient vegetation. Mediterranean shrublands are typically sclerophyllous, adapted to drought conditions and nutrient scarcity. Our shrubland environment had richer plant community, and with a greater heterogeneity in its structure that give rise to patches dominated by shrubs that create different microconditions within the same habitat like a 'resource reserve island' (Doblas-Miranda *et al*., 2009). Some of these patches are more closed, mainly due to the presence of these thickets, the correlation of which is positive with respect to improved environmental conditions for microarthropods (Liu *et al*., 2013). The larger and more luxuriant thickets increase shade and thus soil moisture due to their leaf litter, which retains soil water for a longer period, especially important in this semi-arid Mediterranean climate with severe droughts (Liu *et al*., 2022) as the dry period recorded during the sampled period of this study.

Shrublands also form part of the resource sinks for this fauna through the reservoir of organic matter to the soil from the leaf litter and by the exudations of the rhizosphere (Sardans and Peñuelas, 2013), this increases fertility thanks to the presence of more nutrients available in the trophic network, which favor the presence of more specialized edaphic animals. Mycorrhizal fungi also play an important role in improving the conditions of a resource-limited habitat in that they associate with different groups of plants (the more plant variety, the more mycorrhizae) and thus improve their resistance to environmental stress (droughts, nutrient shortages or soil alterations). Shrublands also provide other types of resources such as more shelters, or potential oviposition sites for a wide variety of fauna (Liu *et al*., 2013). On the other hand, other patches leave more open spaces for generalist species that require less demanding conditions of food, humidity and light. All these microclimatic factors determine the distribution of the edaphic community. This mosaic-like heterogeneity allows the availability of varied resources, which results in greater development for the less abundant taxonomic groups that find here a favorable place to live. On the other hand, in the grassland or forest, under the same conditions of limited resources, they have lower complex compositions and the dominance of microarthropod taxa is much more unequal.

6. - CONCLUSIONS

[1] Soil quality according to QBS-ar was not significantly different in any of the habitats, but it tends to improve with the degree of development of the woody plant community. Forest and shrubland soils, present organisms adapted to more specific soil conditions with higher EMIs and therefore can be understood as a more stable ecosystem with better physicochemical conditions. On the other hand, the herbaceous cover seems to harbor more arthropods, but more generalist and adapted to poorer soil qualities.

[2] Despite the dominance between faunistic taxa can be variable according to the environmental conditions, mites were always the dominant group in all the habitats, followed by Hymenoptera (ants) that were exclusive from grassland and shrubland. Other taxa such as Chilopoda, Diplopoda and Pauropoda had a low abundance but a very important contribution to the final value of the EMI because they contribute maximum values.

[3] The ecological indicators of richness and diversity did not indicate differences between habitats. However, the evenness revealed significant similarities between grassland-shrub and shrub-forest, observing a positive correlation between the height of the scrub and the existence of more suitable conditions for the organisms. This information highlights the importance of considering the role of vegetation, its heterogeneity and complexity in shaping soil quality and emphasizes the need for sustainable land management practices to preserve and improve soil health in natural forest environments.

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8-. REFERENCES

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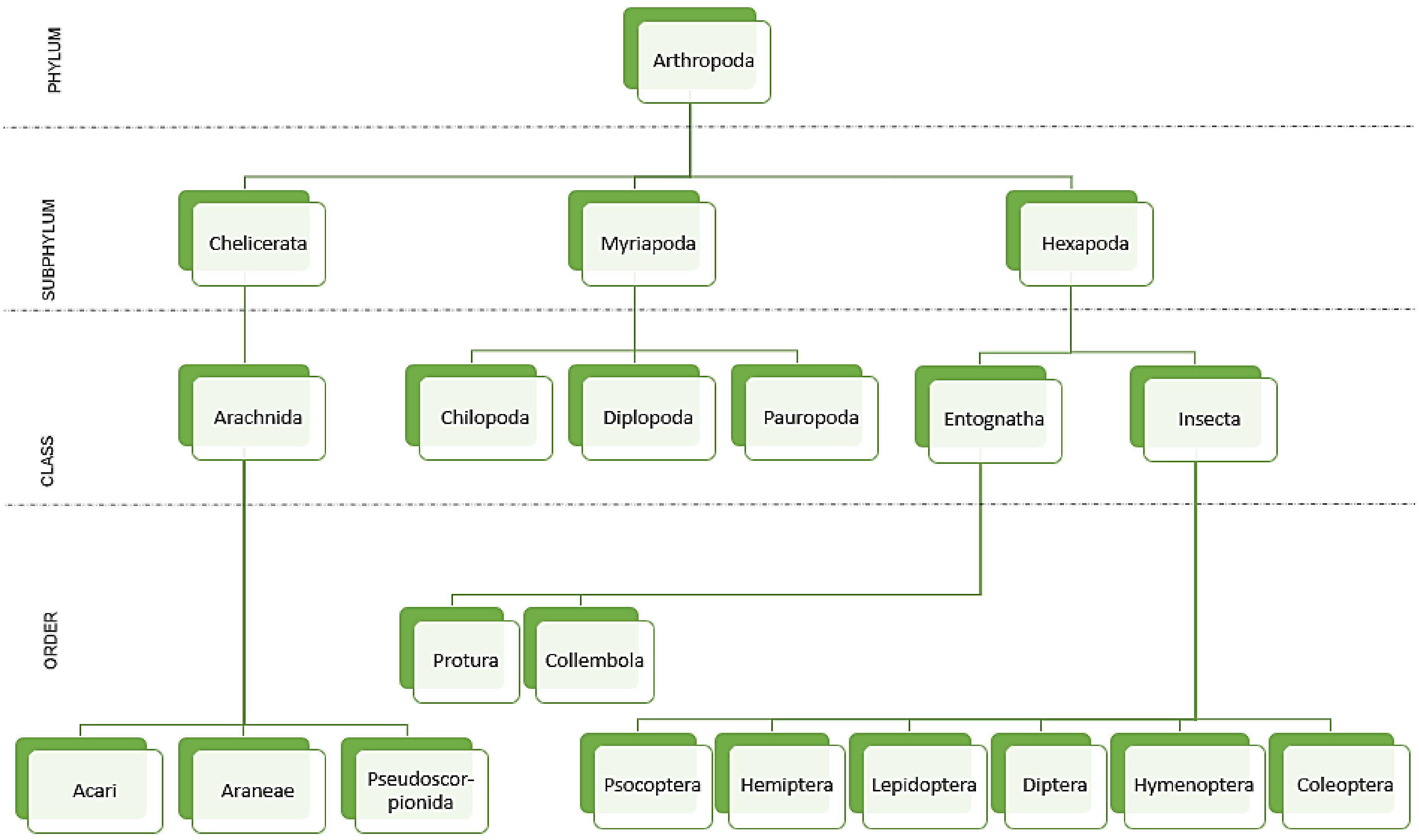
SUPPLEMENTARY MATERIAL 1 – TAXONOMY OF THE STUDIED MESOFAUNA GROUPS



SUPPLEMENTARY MATERIAL 2 – PHOTOS OF THE SAMPLING AREA

Figure S.M.2.1. Grassland plot 2.

Figure S.M.2.2. Shrubland plot 2.

Figure S.M.2.3. Forest plot 2.

Figure S.M.2.4. Soil sampling in shrubland plot 1.

Figure S.M.2.5. Quadrant to measure leaf litter and herbaceous cover in forest plot 3.

Figure S.M.2.6. Soil sampling and labelling samples in shrubland plot 1.

Figure S.M.2.7. Locating the sampling points of the forest plot.

Figure S.M.2.8. Measuring tree height in forest plot 1.

SUPPLEMENTARY MATERIAL 3 – VARIABLES, ABBREVIATIONS AND UNITS

Table S.M.3.1. Variables with their corresponding abbreviations and units.

Variable	Abbreviation	Unit	Variable	Abbreviation	Unit
Grassland Shrubland Forest	Gr Sh Fo	-	Surface stoniness	SuS	%
Coordinate X Coordinate Y	X Y	-	Leaf litter weight	Lw	g
Altitude	A	m	Leaf litter cover	Lc	%
Slope	S	%	Bulk density	Bd	g/cm^3
Orientation	O	-	Humidity	Hu	%
Fraction of tree canopy cover	Fcc	%	Soil stoniness	SoS	%
Average normal diameter of 5 nearest trees	D	cm	Texture	T	-
Average height of the 5 nearest trees	H	cm	Real density	Rd	g/cm^3
Average height of 5 nearest shrubs	Hs	cm	Porosity	P	%
Herbaceous cover	Hc	%	Organic matter	OM	%

SUPPLEMENTARY MATERIAL 4 – COLLEMBOLA'S EMIS ASSIGNED AND PHOTOS

Table S.M.4.1. EMI for collembolans less than 2 mm (larger specimens EMI value = 1).

Colour	Sclerotization	Furca	Antennae	EMI
Colored (brown, marbled...)	Strong	Visible and developed	Long	2
Colored (pink, purple, gray, ...)	Medium /weak	-	Short	3
Light coloration (yellowish, greenish...) with transparent areas	-	-	Normally long	4
Transparent with colored areas (head, legs, antennae)	-	-	Normally long	6
Trasparent only with colored antennae	-	-	Normally long	8
Transparent	-	Visible and developed	Long	10
Transparent	-	Absent or short	Thick, triangular, not very developed	20

Table S.M.4.2. Collembolan morphotypes observed in Monte el Viejo and its EMI assigned.

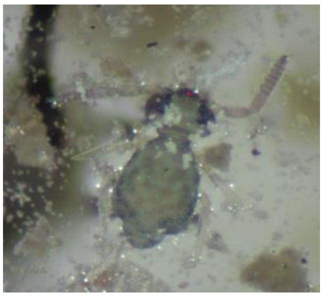
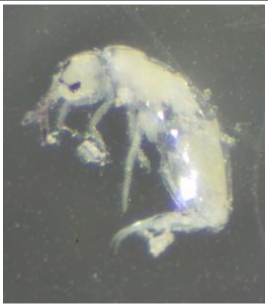
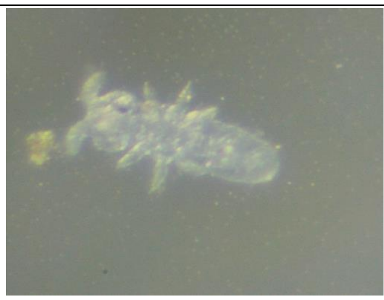
EMI	
2	   
3	  

Table S.M.4.2. (Continuation). Collembolan morphotypes observed in Monte el Viejo and its EMI assigned.

4		
6	 	
8		
10		
20		

SUPPLEMENTARY MATERIAL 5 – SOIL ANALYSIS METHODOLOGY

1. Bulk density:
    - Determine the volume of the sample cylinder measuring the diameter and the length with a caliper.
    - Transfer the sample in the laboratory to a beaker.
    - Introduce in the oven to 105 ºC for 24 hours to lose moisture.
    - Take the beaker to the desiccator until it cools and weigh.
    - Weigh the beaker with the dried sample.

$$bulk\ density\ (g/cm^3) = \frac{dry\ soil\ weigth\ (g)}{volume\ (cm^3)}$$

2. Humidity: Weight the bulk density sample before and after the oven

$$Humidity\ (\%) = \frac{wet\ soil\ weight\ (g) - dry\ soil\ weight\ (g)}{dry\ soil\ weight\ (g)}$$

3. Preparation of the sample
    - Spread the sample on a tray arranged so that the thickness does not exceed 2 cm. Break the soil aggregates with the fingers and let dry 48 hours.
    - Weight the dry sample.
    - Remove separately both, coarse (MAPA, 1994) and organic elements (stems and roots) and leave them in trays.
    - Break up the soil aggregates again with the fingers or a glass bottle.
    - Pass the soil through a sieve of 2 mm of light moving it closed horizontally and before opening, it wait so as not to lose sample when lifting the lid.
    - Remove mineral and organic elements, break up soil aggregates and sieve again. What has passed the sieve is the fine soil.
    - Weigh the fine soil (<2 mm), the coarse elements (>2 mm) and organic elements (>2 mm) and also note the abundance, size and shape of coarse elements

4. Soil texture: USDA, 1999 method of determining texture by feel procedure showed in Figure S.M. 2.1.

5. Organic matter: was calculated with modified method of Schulte (1996) introducing 3 g of soil sample in the oven at 105 ºC for 24h, weight and introduce in the muffle 400 ºC for 4 h and weight again

6. Real density following the pycnometer method modified by Blake (1986a):

➢ pycnometer calibration:
    - Weigh the empty pycnometer and fill with distilled and degassed water and weigh again
    - Measure the temperature of the water to know its density and empty the pycnometer

➢ determination of real density of the soil
    - Add 10 g of dry fine soil in the empty pycnometer and weigh
    - Add degassed water to the pycnometer with the soil up to approximately half of its volume. Introduce the pycnometer in a vacuum desiccator for 15 minutes. Apply a negative tension, to facilitate the elimination of the air that is trapped between the soil particles.

- Remove from the desiccator, finish filling with degassed water, level it and weigh.

$$total\ volume(mL) = \frac{water\ weight\ (g)}{water\ density\ \left(\frac{g}{mL}\right)} + \frac{soil\ weight\ (g)}{soil\ density\ \left(\frac{g}{mL}\right)}$$

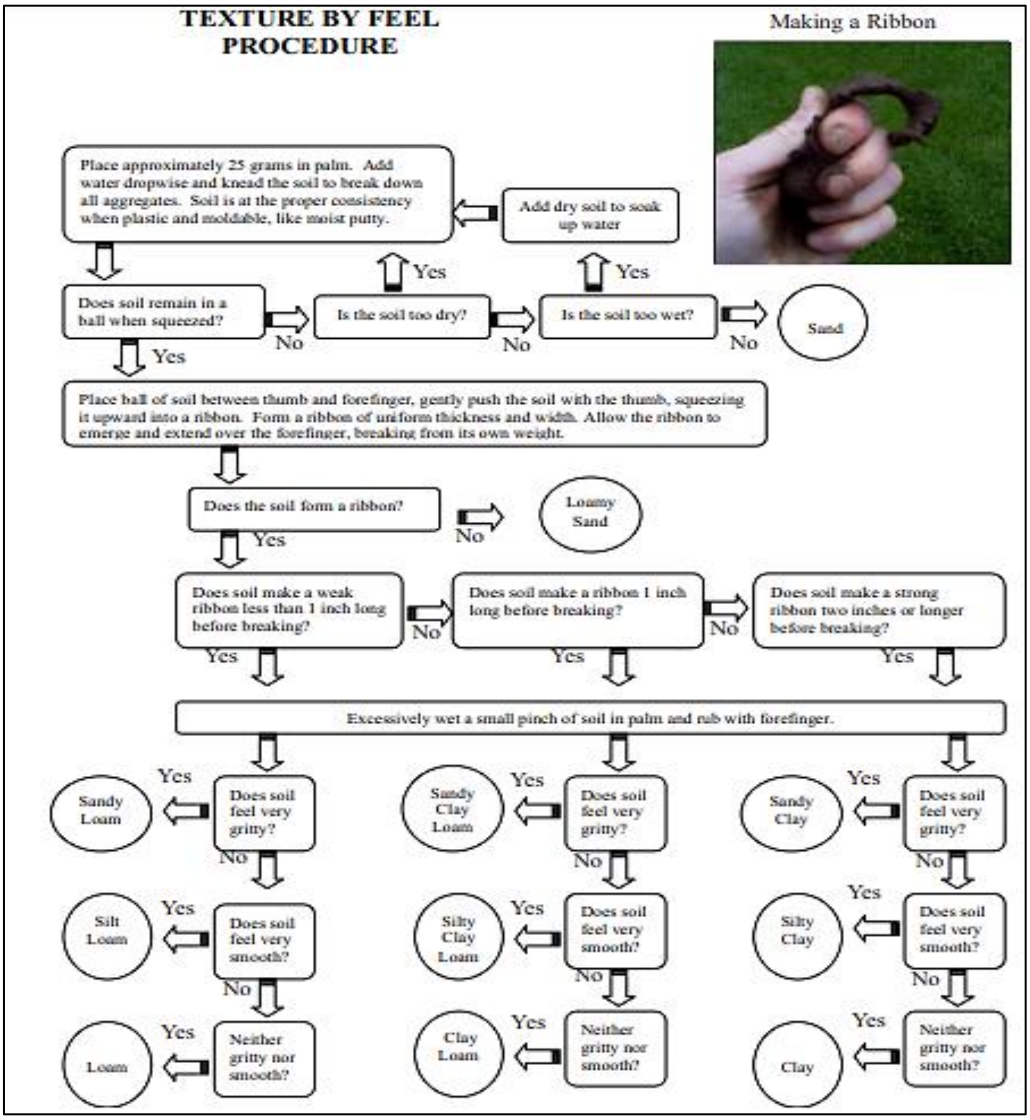


Figure S.M.5.1. Texture by feel procedure (USDA, 1999).

7. Porosity:

$$Porosity\ (\%) = \frac{pores\ volume}{total\ soil\ volume} * 100 = \frac{real\ density - apparent\ density}{real\ density} * 100$$

8. Organic matter:

$$Organic\ matter\ (\%) = \frac{organic\ matter\ weight\ (g)}{dry\ soil\ weight\ (g)} * 100$$

SUPPLEMENTARY MATERIAL 6 – PLOT CHARACTERIZATION AND SOIL ANALYSIS RESULTS

Table S.M.6.1. Results of plot characterization. Sample: code assigned to each sample (Gr: grassland, Sh: shrubland, Fo: Forest); X/Y: X/Y coordinate in ETRS89 UTM 30N system; A: altitude; S: slope; Fcc: tree canopy cover; D: average normal diameter of the 5 nearest trees; H: average height of the nearest 5 trees; Hs: average height of the nearest 5 shrubs; Hc: live herbaceous cover.

Sample	X	Y	A (m)	S (%)	Fcc (%)	D (cm)	H (cm)	Hs (cm)	Hc (%)
Gr1	370446	4647717	816	6.00	0	0	0	47	40
Gr2	370440	4647709	816	6.73	0	0	0	48	45
Gr3	370433	4647702	816	4.51	0	0	0	58	50
Sh1	370341	4647638	832	32.00	1.90	6.6	450	136	15
Sh2	370334	4647630	833	32.67	0	11	400	156	20
Sh3	370327	4647623	833	31.21	0	10.75	450	114	10
Fo1	370535	4647691	818	29.21	71.67	18.7	780	71	5
Fo2	370528	4647684	819	25.68	39.29	15.3	750	65	10
Fo3	370522	4647676	819	20.59	40.91	20.9	870	51	5

Table S.M.6.2 Results of soil analysis: W: soil sample weight (g; arthropods sampling); Hu: soil humidity; Bd: bulk density; Rd: real density; P: porosity; SuS: surface stoniness; SoS: sample stoniness; Lw: weight of litter; Lc: percentage of litter; T: soil texture, and OM: organic matter.

Sample	W (g)	Hu (%)	Bd (g/cm^3)	Rd (g/cm^3)	P (%)	SuS (%)	SoS (%)	Lw (g)	Lc (%)	T	OM (%)
Gr1	1139.08	10.11	1.15	2.26	49.31	5	30.29	0.00	0	Clay loam	6.18
Gr2	1067.82	7.65	1.15	2.14	46.14	5	28.08	0.00	0	Clay loam	6.47
Gr3	930.17	8.03	1.09	2.17	49.59	5	40.68	0.00	0	Clay loam	7.90
Sh1	745.94	15.45	1.03	2.18	52.72	5	15.02	11.98	15	Silty clay loam	6.33
Sh2	642.09	17.24	0.96	2.33	58.85	5	19.86	7.97	15	Silty clay loam	6.05
Sh3	885.17	7.38	1.05	2.21	52.39	5	22.07	3.67	5	Silty clay loam	5.48
Fo1	477.96	11.10	0.88	2.65	67.00	5	22.59	44.82	90	Loam	10.62
Fo2	550.76	16.20	0.83	2.20	62.26	5	21.41	57.29	85	Silty loam loam	8.89
Fo3	483.07	35.68	0.36	1.63	77.86	5	7.98	50.84	15	Clay loam	19.56

SUPPLEMENTARY MATERIAL 7 – DESCRIPTION AND RELEVANCE OF EACH SAMPLED FAUNISTIC TAXA

SUBPHYLUM CHELICERATA

Class Arachnida

Arachnida include different orders of which, in this study only have appeared three of them: Acari, Araneae and Pseudoscorpionida. In a general way, animals belonging to this Class have their bodies are divided in the prosoma (with a pair of chelicerae, a pair of pedipalps and 4 pair of legs), and the opisthosoma.

Order Acari

The ecosystems in which they live are as varied as their morphology and habits they present. They have a very varied feeding regime and their body and cheliceral morphology changes accordingly. They are predators, phytophagous, mycophagous, saprophagous, coprophagous, necrophagous, detritivorous or can be phoretic or parasites on other species (Bernard, 2003; Evans, 2013).

Order Araneae

Spiders measure between 0.5 mm to 9 cm (Levi *et al.*, 2023) and have mostly, 4 pair of eyes and have their organs specialized to their habit. Spiders are solitary (Melic *et a*l., 2015; Bernard, 2023)

Order Pseudoscorpionida

Pseudoscorpions are morphologically like scorpions but smaller (0.8 mm to 1 cm according to Zaragoza (2015)) and without their tail with the sting. They have pincer-shaped chelicerae and big chelate pedipalps, with poisonous glands and setae very sensitive to air movements (Bernard, 2023). They are widely distributed in all continents of the planet. For their dispersal, they generally use phoresy with insects, on which they do not feed. Usually present in stable grasslands and forests as stated by Menta and Remelli (2020).

SUBFILUM MYRIAPODA

The body of the Myriapoda specimens is divided into a head and a trunk. They are characterized by numerous pairs of legs that are inserted in many segments along their trunk. This group includes four Classes: Chilopoda, Diplopoda, Pauropoda and Symphyla (Bernard, 2023), of which, we only found the first three in our soil samples.

Class Chilopoda

Chilopods have a flattened and whitish body (between 1 mm and-30 cm long), and a pair of forcipules that they use to poison their prey (Giribet, 2015; Bernard, 2023;). As for the place where they live, they do not distinguish between wet or arid places, usually under natural elements like leaf litter or rocks.

Class Diplopoda

The elongated body of diplopods can be cylindrical, hemispherical or flattened, depending on the species, normally sclerotized, except in the Order Polyxenida (Melic, 2015b). One of their most notable characteristics is that they roll up on themselves when they feel threatened, thus protecting their most vulnerable parts. Their size varies from less than 1 mm to as large as 30 cm (Bernard, 2023). However, unlike the chilopods, the diplopods or millipedes have two pairs of legs per segment due to the fusion of the segments two by two (Melic, 2015b), which allows them to have a more robust body with which to dig in the leaf litter or in the soil as Bernard (2023) indicates. They feed primarily by herbivory of dead plant material; however, they also resort to coprophagy, necrophagy, fungivory or predation.

This arthropod usually lives solitary, found associated with decaying plant material, in the soil itself or under rocks. The evolution of this taxonomic group has allowed them to be ubiquitous (Menta and Remelli, 2020). Depending on the site in which they live, they have morphological adaptations such as less pigmentation or absence of eyes (Melic, 2015b). Diplopods can also be found in more opened areas like grasslands or agricultural ecosystems (Melic, 2015b; Menta and Remelli, 2020), but less often. Different millipede species may have preferences for specific types of vegetation, and their populations may be more abundant in areas where their preferred plant species are dominant.

Class Pauropoda

This small myriapod, measuring between 0.4 and 2 mm, are blind creatures that also avoid light. Their body is generally wide, whitish, and covered in setae. They have a thin and usually smooth cuticle, occasionally granulated. They have 9 to 11 pairs of legs that help them move in short bursts, after which they briefly stop (Domínguez, 2015).

Their diet is based on being detritivores and fungivores (Domínguez, 2015; Bernard, 2023). Much is still unknown about the ecology of this group. Their morphological characteristics indicate their adaptation to predominantly humid environments. Temperature is also a determining factor, as species richness significantly decreases in very cold environments. They can be found in a variety of ecosystems with different types of vegetation and soil. Although they are also common in agricultural areas, they tend to appear more frequently in forest environments, taking refuge under fallen plant material, on trees, or in crevices (Bernard, 2023).

SUBFILUM HEXAPODA

Class Entognatha

This class includes primitive insects, such as those without developed wings (apterygotes) and those without visible jaws (entognatha). We have found specimens of this group belonging to the orders Protura and Collembola.

Order Protura

Protura are characterized by having an elongated (less than 2 mm) and thin body, with a little sclerotized cuticle and little color, whitish or yellowish. They lack antennae and eyes, but they are replaced with the front legs that have been enlarged and placed at the front, while the second and third pair of legs are responsible for the movement. The mouthparts are styliform to feed on fungi, but this is not clearly studied yet (Bernard, 2023; Menta and Remelli, 2020), are also believed to feed on liquids from decaying organic matter and bacteria (Melic, 2015a).

They live in places without light because they are lucifugous, but are not troglodytes (Melic, 2015a). Although proturans are not able to travel long distances, they are distributed in many habitats (Pass and Urban, 2011).

Order Collembola

Springtails have small bodies between 0.12 mm and 17 mm (Baquero and Jordana, 2015) divided in 3 tagma (head, thorax and abdomen). They are characterized by having an odd appendage; the furcula or furca, that they use to flee from predators. Their morphology changes depending on their location on the ground in vertical stratification: whitish body and smaller or lack of appendages in deeper layers (Rusek, 1998); or long appendages and colored bodies in shallower and leaf litter layers (Bernard, 2023). Their mandibular characteristics also differ according to their feeding habits: most of them have chewing-type mouthparts, while others, have piercing-sucking mouthparts, this allows them to have a wide range of feeding habits.

They are generally defined as fungivores (Bernard, 2023), but according to Arbea and Blasco-Zumeta, (2001), they also prey on some edaphic pathogens such as Protozoa, Nematoda, Rotifera, Enchytraeidae, Trematoda, some bacteria, algae as well as on other springtails.

Class Insecta

This Class of Hexapoda includes the group of pterygota (insects with wings) in which we can find several orders, of which we find in this study: Psocoptera, Hemiptera, Lepidoptera, Diptera, Hymenoptera and Coleoptera.

Order Psocoptera

These insects have a globular head and abdomen, and well-developed eyes (Alexander *et al*., 2015). They are divided into two types according to their size; the specimens captured in this study belong to the smaller body size group (< 2 mm), commonly known as book lice, which are wingless. They are free-living insects and do not act as parasites.

They typically take refuge in vegetative environments, such as leaves or tree trunks, although some species have been found in caves. Some species are particularly abundant on herbaceous plants, while others prefer forest leaf litter or even bare rocks.

Although they have a high capacity of movement, they typically remain in the same habitat as long as it has not undergone significant alterations over extended periods of time (Socarrás (2013). They are widely distributed throughout the world.

Order Hemiptera

The adults can vary in size, ranging from 1 mm to over 10 cm (Froeschner, 2023). They are characterized by their elongated mouthparts in the form of a stylet, and the presence of membranous wings in the distal region but thickened at the basal region hemelytra. Most specimens belonging to the Suborder Heteroptera inhabit terrestrial ecosystems, living on plants, under tree bark, in leaf litter, on rocks or soil crevices, and in the soil itself (Fauvel, 1999; Weiraucha *et al.*, 2019). Regarding their diet, they feed on plant parts, fungi, animals (Frank and Künzle, 2006). Others are saprophagous and feed on decomposing organic matter or are omnivorous.

Order Lepidoptera

This group is widely known for its visibility due to its flight, in which its pair of colorful membranous wings stand out, covered with scales (modified setae). The larvae are eruciform, meaning they have a caterpillar-like shape, possess 5 pairs of false legs, and have a strongly sclerotized head. Adults and larvae have very different sizes. The pupa or chrysalis, in which the metamorphosis characteristic of their holometabolism occurs, usually has the appendages fixed to the body without mobility. The main feeding stage is caterpillar, terrestrial larvae feed on all parts of plants, although some species are also known to be predators or saprophagous (García-Barros *et al*., 2015). Lepidoptera are insects with wings covered by scales; they have a crucial role in the food chain in terrestrial ecosystems (Legal, 2023). Their habitat varies greatly depending on the species, occupying all possible environments, some of them are specialists of a specific habitat (García-Barros *et al*., 2015; Legal, 2023).

Order Diptera

Dipterans, which include well-known groups such as flies and mosquitoes, insects that have transformed their second pair of wings into halteres, small structures that help them balance while flying, although there are some wingless species. Carles-Tolrá (2015) indicates that their size varies to less than 1 mm from 8 cm.

Regarding feeding habits, they exhibit extremely varied behaviors, including phytophagy, coprophagy, saprophagy, necrophagy, xylophagy, predation, decomposition, parasitism, parasitoidism, and fungivory. (Carles-Tolrá, 2015). While they are present in any ecosystem, temperature and humidity conditions influence their distribution and abundance.

Order Hymenoptera

Hymenoptera is a large holometabolous group that includes well-known insects such as wasps, bees, and ants, among others, with very different sizes. Their size ranges from 0.2 mm to 5 cm (Lindauer, 2023). Many of these insects feed on plant nectar, while others feed on fungi, leaves, (Britton, 2023) or other body parts of other insects (Fernández and Pujade-Villar, 2015) and some of them are parasitoids. Some are solitary, while others are gregarious. These aggregations can lead to high population densities (Menta y Remelli, 2020). Hymenopterans play various roles in ecosystems, which are distributed among different groups. Bees and wasps, for example, are involved in pollination and controlling populations of other insects and arachnids, particularly when they become pests (Fernández and Pujade-Villar, 2015).

Their species diversity has allowed them to colonize all type of habitats (Fernández and Pujade-Villar, 2015).

Order Coleoptera

Beetles is the most diverse order of living organisms (Carles-Tolrá, 2015; Menta and Remelli, 2020) characterized by the presence of the elytra. Although they have common characteristics, such as being highly sclerotized, with membranous wings hidden beneath hardened forewings called elytra (Alonso-Zarazaga, 2015). They range in size from less than 1 mm to over 12 cm (Gressit, 2023), and exhibit a wide range of coloration.Their feeding habits vary greatly depending on the species. There are carnivorous, phytophagous, omnivorous, mycophagous, and saprophagous beetles, among others as Alonso-Zarazaga points out (2015).

Terrestrial species are associated with a wide variety of vegetation, ranging from herbs to shrubs and trees, as well as lichens and mosses. Some are predators, while others are parasites (Alonso-Zarazaga, 2015). There are beetles that specialize in a single habitat type, while others are more ubiquitous (Menta and Remelli, 2020).