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# Soil centipedes (Chilopoda, Geophilomorpha) in the Val Camonica forests (Southern Alps): species composition and richness

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## Abstract

Soil centipedes (Chilopoda, Geophilomorpha) are a widespread group of predators in the forest soils of the European Alps. While in the Eastern and Western Prealps important efforts were devoted to better understand geophilomorph fauna, little is known about species composition of the central part of Southern Prealps. In this work, 5 sites located in the Val Camonica were surveyed by hand searching between November 2021 and July 2022. Species richness was estimated applying non-parametrical statistical methods (Chao-1 and Abundance-based Coverage Estimator) to account for incomplete detection. A total of 18 species were found. A maximum of 12 species were recorded in a single site, while estimates suggest that other 1-3 species were likely undetected. By comparing the communities, species composition were found variable also between sites with similar species richness.

## Keywords

Chilopoda, Southern Prealps, community ecology, species richness

## Introduction

Soil centipedes (Geophilomorpha) are a widespread component of the soil fauna (Jeffery et al. 2010, Orgiazzi et al. 2016). In particular, there are more than 40 morphologically distinct species recorded just in the European Alps (Bonato et al. 2014). As a matter of fact, temperate forests could hold the richest communities of geophilomorphs, and more in general chilopods (e.g., Petersen and Luxton (1982), Bonato et al. (2017)).

In comparison with other major groups of soil predators, many facets of the diversity of geophilomorph communities and their ecology are almost unknown (Bonato and Minelli

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2009, Bortolin et al. 2018). As many other soil invertebrates, geophilomorphs are strongly affected by local environmental factors: species composition and abundance of populations could change on a short spatial scale (e.g., Purchart et al. (2013)). In addition, most studies on community composition reported only the number of species found, which is usually lower than the real number of species present in a given site because of the well known problem of incomplete species detection (Gotelli and Chao 2011). As a matter of fact, carrying out exhaustive sampling is a hard task: most of geophilomorphs are endogeic and perform seasonal migrations between different soil strata to survive unfavorable environmental conditions (Voigtländer 2011). However, suitable methods have been developed to overcome the problem of incomplete species detection (Gotelli and Chao 2011).

This paper presents the results of the first investigation on the geophilomorph communities in the forests of an area of the Southern Alps, namely Val Camonica (Fig. 1). As a matter of fact, very few data have been published so far on geophilomorphs from this area: only one record of *Eurygeophilus pinguis* (Brölemann, 1898) (Manfredi 1948) and one of *Himantarium gabrielis* (Linnaeus, 1767) (Zapparoli and Minelli 2005). More in general, no targeted surveys have been carried out so far on geophilomorphs in the central part of Southern Prealps, unlike Western Alps (Minelli and Zapparoli 1992) and Eastern Prealps (Minelli 1987, Zapparoli 1989, Erhard 1996, Ravnjak and Kos 2015).

The aim of the study was (i) to contribute to fill in the gap on knowledge of geophilomorph fauna of Southern Prealps by focusing on the Val Camonica forest soils, and (ii) to estimate the species richness of selected communities with statistical models in order to adjust for incomplete detection.

# **Material and Methods**

### Study area

A total of 5 sites were studied in the Val Camonica (Fig. 1, Table 1). The minimum distance between two sites was 5.3 km, while the maximum was 22.2 km. Sites were selected within woods, selecting those currently not affected by human usage other than wood harvesting (Fig. 2). Sites were chosen to cover different parts and both sides of the valley. Each site was defined as a circular area of radius 8 m, within a continuous forest patch of at least 0.25 ha with uniform vegetation structure and at least 10 m away from forest edges, other ecotonal zones, and roads.

### Sampling protocol

The 5 sites were visited between November 2021 and July 2022, for a total of 2-7 sampling sessions for each site. Each sampling session was carried out for 1.0-1.5 hours by 1-4 researchers searching in parallel by hand on the ground, digging with a small shovel in the leaf litter and soil, going deep to about 15 cm (when possible), and turning stones and

barks on the surface. This method was chosen because it is known to be one of the most effective for endogeic centipedes, as shown by Tuf (2015).

## Species identification

All geophilomorph centipedes were collected in test tubes and fixed with 70° ethanol, and later identified to species level using a Leica DMLB microscope with magnification up to 400×, after mounting the specimens on temporary microscopic slides (Pereira 2000). When none of the two pretarsi of the second maxillae was visible, the head of the specimen was detached from the trunk (see Bonato et al. (2010), for anatomical terminology).

Identification of all specimens was conducted by means of Chilokey (Bonato et al. 2014), and, when necessary, considering the original descriptions or subsequent redescriptions of the species. For taxonomy and nomenclature, the Checklist of the Italian Fauna was followed (Bonato and Minelli 2021).

### Species composition

Differences in species composition between sites were evaluated with the Jaccard similarity index, which is based on presence-absence data. Moreover, a Correspondence Analysis was also performed in order to assess pattern of diversity between sites and between species. Since sites received different sampling efforts, the analysis was performed on presence-absence data, not on abundance data, using the FactoMineR package in R (Husson et al. 2007, Lê et al. 2008). Biplots were generated using the Factoextra package in R (Greenacre 2010, Kassambara and Mundt 2017).

### Species richness estimation

The number of species in each site was estimated using two non-parametric estimators: the Chao-1 estimator, which is based on the proportion between the number of species collected once and the number of those collected twice (Chao 1984), and the Abundance-based Coverage Estimator (ACE), which is based on the frequency of "rare" species (Chao and Lee 1992). These estimators allow one to overcome the limitations of parametric estimators, which do not assess undersampling bias (Magurran 2004).

Chao-1 and ACE were calculated using PAST 4.08 (Hammer et al. 2001) and the vegan package in R (Oksanen et al. 2017) using all parameters as default; 95% confidence intervals were computed by the bootstrap method in PAST.

In order to compare species richness among sites, rarefaction and extrapolation were integrated from the observed value of species richness, with 95% confidence intervals based on "unconditional" variance, as proposed by Colwell et al. (2012). The analysis was performed with the iNEXT package in R (Hsieh et al. 2020), which uses the bootstrap

method proposed by Chao et al. (2014). The parameters were set as default, except for the number of permutations, which was set to 150. A rarefaction analysis with 95% confidence intervals based on "conditional" variance (Magurran 2004) was also performed with PAST.

# Results

A total of 38 hours of sampling sessions allowed to collect 242 specimens, with between 31 and 85 specimens per site. All specimens were identified to species level, for a total of 18 species detected (Table 2).

## Species composition

Considering the species detected in the five communities, the pairwise values of the Jaccard similarity index were between 0.11 (between sites D and E) and 0.38 (between sites B and C), with a mean value of 0.26 (Table 3).

The Correspondence Analysis performed on presence-absence data produced three main coordinates, accounting for 38.3%, 29.9%, and 19.2% of the total variance, respectively (Fig. 3). Taking into account the first two coordinates, the community E was different from all other sites because of the presence of *Strigamia acuminata* and *Eurygeophilus pinguis*, while the community B was separate from all the others because of the presence of *Geophilus* sp., *Henia vesuviana*, *H. montana*, *H. brevis*, and *Stigmatogaster gracilis*. Communities C and D differ from the other and share the presence of *S. crassipes* and *G. impressus*. Finally, the third coordinate allows distinguishing community A from all the others.

## **Species richness**

Between 4 and 12 species were detected in each of the 5 sites (Fig. 4, Table 4): 4-6 in three sites (A, D, E) and 10-12 in the other two (B and C). In most of the sites, estimates of actual species richness (Chao-1 and ACE) excessed the observed number of species, with 1-3 species likely undetected (Fig. 4, Table 4). In the sites with the highest number of observed species (B and C), the estimators suggested that the sampling was pretty exhaustive, but the 95% confidence intervals of Chao-1 indicated the possibility of many other undetected species (Fig. 4, Table 4). PAST and vegan gave similar results, different just slightly. The two sites with the highest species richness (B and C) were also the two most similar to each other (see Table 3).

The rarefaction analysis with 95% confidence intervals based on "unconditional" variance (Fig. 5a) indicated a statistically significant difference in species richness between the poorest site (A, with 4 detected species and no estimated undetected species) and the sites B, C, and E. Moreover, the rarefaction analysis with 95% confidence intervals based on "conditional" variance (Fig. 5b) suggested that sites B and C are significantly richer than site D.

# Discussion

This study provides the first insights on the species richness and composition variation of geophilomorph communities in forest soils of Val Camonica. Moreover. the work contributes to fill a gap of knowledge on the geophilomorph fauna of a broader sector of the Southern Prealps (see above in Introduction). Of the only 2 species previously recorded in Val Camonica (Eurygeophilus pinguis and Himantarium gabrielis), only the first was confirmed, while the latter was not found, but it is expected to be strightly limited to xerothermic sites along the Southern Prealps (Zapparoli and Minelli 2005). Moreover, other 17 species were found anew (Table 2). Among these many species, two are most probably still undescribed species, belonging to the genera Henia C.L. Koch, 1847 and Geophilus Leach, 1814. In general, all the species found were expected, based on their reported occurrence on either the Western part or the Eastern part of the Southern Prealps (Zapparoli 1989, Minelli and Zapparoli 1992). On the other hand, the apparent absence of Clinopodes flavidus C.L. Koch, 1847 is notable, because it occures both in the Bergamo Prealps (Zapparoli and Minelli 2005) and the Garda Prealps (Minelli 1992).

## Species composition and relation with species richness

There are few studies that compare local communities of geophilomorphs in terms of species richness (e.g. Grgič and Kos (2005), Leśniewska et al. (2005), Leśniewska and Leśniewski (2015), Peretti and Bonato (2018)). In the study area, geophilomorph communities with similar estimates of number of species (4-7) have actually very different composition (as shown in Table 3). In the same way, also the other two sites with more numerous species estimates (10-13) were different in composition (only 5 species in common). These differences could be explained by very different habitats (Table 1). Anyhow, more studies are needed to understand which ecological parameters influence more the composition of geophilomorpha communities.

Results of this work could be affected by some methodological limits. The estimates of species richness and their comparisons between sites could be biased by different probability of detection between species, and between different sites for the same species. Despite this, the hand searching method adopted by us permitted to increase the sampling rate of geophilomorphs, and to catch also strictly endogeic species, unlike other commonly performed methods (e.g., pitfall traps), as also shown by Tuf (2015).

### **Richness estimates**

Real data as well as non-parametric estimators indicates that more than 12 species of geophilomorphs – not considering high level of uncertainty because of large confidence intervals – can regularly live in syntopy in the study area (Fig. 4).

Considering the Southern Prealps and Dinarides, a few other studies estimated centipede species richness using statistical tools to account for incomplete detection (Grgič and Kos

2003, Grgič and Kos 2005, Peretti and Bonato 2018). However, all these studies did not provide separate estimates of only the geophilomorphs. But, taking into account the absolute number of detected species, between 4 and 16 species of geophilomorphs were detected, among all the sampling sites of these studies, with a mean of 9-10 species. On the other hand, the five sites sampled in Val Camonica have a smaller mean of species present, which is 7.

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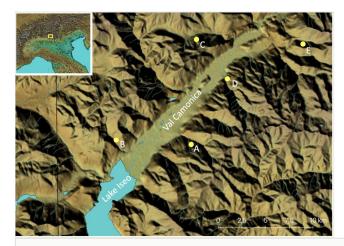
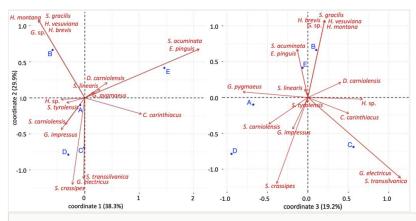


Figure 1. Sampling sites in the Val Camonica (yellow points).



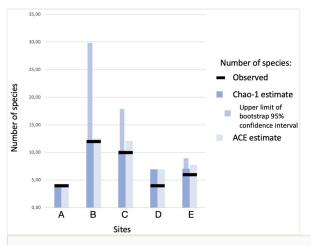
Figure 2. Sampling sites (Table 1).



#### Figure 3.

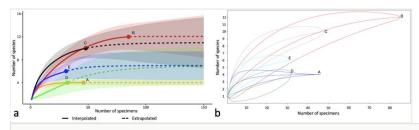
Contribution biplots of the Correspondence Analysis performed on presence-absence of species in 5 sites in the Val Camonica. Arrows correspond to species, while points correspond to the sites.

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#### Figure 4.

Observed and estimated species richness of Geophilomorpha in 5 sites of the Val Camonica.



#### Figure 5.

Comparison of the estimated species richness of Geophilomorpha between 5 sites in the Val Camonica. **a** Rarefaction curves (solid lines) and extrapolated curves (dashed lines) with 95% confidence intervals based on unconditional variance (colored areas). **b** Rarefaction analysis, 95% confidence intervals based on conditional variance.

#### Table 1.

Geographic features of the sampling sites in the Val Camonica.

Site	Locality	Latitude (°N)	Longitude (°E)	Altitude (m)	Aspect	Lithological substrate	Dominant tree species	Number of sampling se
A	Acquebone: Near Ca' de Gos	45.8368	10.1812	760	W	Schistose metamorphic	Castanea sativa, Larix decidua, Picea abies	6
В	Stramazzano: Torrente Supine	45.8420	10.0795	695	SSW	Carbonate	Castanea sativa, Ostrya carpinifolia, Picea abies, Quercus petraea	7
С	Borno: Under Fienili Mensi	45.9366	10.1906	1070	SE	Carbonate and mixed sedimentary	Abies alba, Picea abies	4
D	Sacca: Valle del Resio	45.8987	10.2319	525	WNW	Siliclastic sedimentary	Castanea sativa, Fagus sylvatica	6
E	Passo Crocedomini: Over Degna	45.9306	10.3354	1190	NW	Glacial drift	Corylus avellana, Fagus sylvatica, Larix decidua, Picea abies	2

#### Table 2.

Species of Geophilomorpha and number of specimens found in 5 sites in the Val Camonica. Families after Bonato et al. (2013). <sup>#</sup> Recently adopted name for the species previously called *Geophilus alpinus* Meinert, 1870 (see Popovici (2022)); \* Putative undescribed species

		All sites				
	А	В	С	D	Е	
Geophilidae						
Clinopodes carinthiacus (Latzel, 1880)			3		4	7
Eurygeophilus pinguis (Brölemann, 1898)					1	1
Geophilus electricus (Linnaeus, 1758)			1			1
Geophilus impressus C.L. Koch, 1847#		2	3	1		6
Geophilus pygmaeus Latzel, 1880	23	54		29	4	110
Geophilus sp.*		2				2
Henia brevis (Silvestri, 1896)		6				6
<i>Henia montana</i> (Meinert, 1870)		1				1
Henia vesuviana (Newport, 1845)		2				2
Henia sp.*		3	2			5
Stenotaenia linearis (C.L. Koch, 1835)	3	7	12		13	35
Strigamia acuminata (Leach, 1815)					1	1
Strigamia crassipes (C.L. Koch, 1835)			1	1		2
Strigamia transsilvanica (Verhoeff, 1928)			3			3
Himantariidae						
Stigmatogaster gracilis (Meinert, 1870)		2				2
Mecistocephalidae						
Dicellophilus carniolensis (C.L. Koch, 1847)		2	10		8	20
Schendylidae						
Schendyla carniolensis Verhoeff, 1902	13	2	11	1		27
Schendyla tyrolensis (Meinert, 1870)	7	2	2			11
Total specimens	46	85	48	32	31	242

Table 3. Jaccard similarity index among 5 sites in the Val Camonica.								
Sites	A	В	С	D	E			
A	1.000	0.333	0.273	0.333	0.250			
В	0.333	1.000	0.375	0.231	0.200			
С	0.273	0.375	1.000	0.273	0.231			
D	0.333	0.231	0.273	1.000	0.111			
E	0.250	0.200	0.231	0.111	1.000			

#### Table 4.

Observed and estimated values of species richness of Geophilomorpha in 5 sites in the Val Camonica.

	Sites				
	А	В	С	D	Е
Observed species	4	12	10	4	6
Estimated richness by Chao-1	4.00	12.07	10.33	6.91	6.97
Upper limit of the 95% confidence interval of Chao-1 index (9999 bootstrap replicates)	4.00	29.79	17.83	6.91	8.90
Estimated richness by ACE	4.00	12.50	12.05	6.91	7.68