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Evaluating and measuring biodiversity in a subterranean light-gradient

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ABSTRACT The structure and composition of the biodiversity have been analysed in a light gradient of a case-study cave in Northern Italy to evaluate the influence of light in promoting, limiting, or altering it. Minor quantitative variations have been found along the gradient but remarkable qualitative differences have been recorded and discussed on the composition of the biodiversity proceeding from the full light of the entrance toward the darkness of the deep cave. Light intensity proved to be the main limit for many troglobiont an troglophilic species migration from or to the inner part of the cave. The subterranean environment is here discussed as a model for assessing also the epigean biodiversity considering the ecological limits in conservation problems of vulnerable environments.

KEY WORDS biodiversity; biospeleology; conservation; ecology; karst.

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INTRODUCTION

Biodiversity, defined as all hereditarily based variations at all levels of organization, from the genes within a single local population or species, to the species composing all or part of a local community, and finally to the communities themselves that compose the living parts of the multifarious ecosystems of the world (Wilson, 1988), is a simple name given to a huge complexity. This complexity is probably the greatest limit for its complete understanding and its full evaluation and measuring, is almost impossible. If measuring the whole biodiversity is a limit many methods to give a good representative quantification have been proposed (Hill et al., 2005). Most of them are based on species indicators, representative for the whole community and useful for comparisons between different places, or on environmental parameters linked to the species richness (Caoduro et al., 2014), as a compromise to manage a fundamental resource for our planet without knowing it in detail. Studying the hypogean environments, usually composed by a scarce number of high specialized species and simple communities, offers the rare occasion to have a nearly complete measure of the whole local biodiversity and a good understanding of its structure. A global evaluation of subterranean biodiversity is however still scarce in literature (Culver et al., 2006) where single species indicators (or groups of) are more often used to compare different caves (Culver & Pipan, 2009; Latella et al., 2012), instead of evaluating the whole biodiversity of a single one.

Caves are not closed environments and measuring biodiversity in caves must consider contamination rates from more or less troglophilic organisms and how much cave organisms remain isolated or migrate to other places, according with the superficial underground environment concept (MSS in Juberthie et al., 1980). Understanding the parameters that influence, promote or limit the biodiversity of a cave can be important to understand how biodiversity complexity evolves in a resource-limited environment. Light gradient is here considered as the main direct limit for autotrophs' ecology, diversification and, influencing also the temperature, evaporation, humidity and other physical parameters, indirectly the key factor for all the other levels of the local food net. The case-study of the cave of Ponte Subiolo is here presented, a well known cave since historical times with an almost straight and barely sloping development with a long light gradient at the entrance which make possible to separate the main steps of the disappearing of the light and its influence on the biodiversity of the cave.

In this study the changes in the biodiversity have been evaluated in relation to the light gradient to examine the species richness, its composition and the dynamics related to a transition zone between epigean and subterranean environments. Evaluating and quantifying how the light influences subterranean communities as a limiting factor for biodiversity can be helpful in understanding how conservation measures promote stable and rich subterranean communities.

MATERIAL AND METHODS

The cave of Ponte Subiolo is located in a subalpine continental area of northern Italy (45°52'18.13"N, 11°40'8.94"E) at 175 m a.s.l. in a narrow valley covered mostly by *Carpinus-Fagus* woods. The cave is a natural part of the dolomitic karst system of the Altopiano di Asiago, partially altered by human activity and occasionally used by tourists since the XIX century. It develops almost horizontally whit a moderate sloping for 260 m from the entrance (Fig. 1). The entrance of the cave is located in the middle of an emi-circus of dolomite rocks (10 m diameter), never exposed to direct sunlight and with scarce surrounding vegetation and with a continuous gradient of light in the straight passage toward the hypogean area. The light gradient was here measured directly with a a luxmeter (1 lux resolution) and indirectly, using the presence of chlorophyll photosynthesis as an environmental parameter, from full light (photosynthesis present) to complete and permanent darkness (photosynthesis absent or not observed), for a length of 20 m and 4 m average diameter tunnel. Photosynthesis was defined by the presence of different kinds of vegetation in three different locations where a pitfall trap has been placed: entrance, in permanent shadow but with full indirect light (C1: 0.5 m from the entrance), where the last living vascular plant (*Parietaria officinalis* L.) was recorded together with non-vascular plants, mosses and algae; twilight zone, an intermediate point between traps C1 and C3 (C2: 8 m from the entrance) just after the last non-vascular plant (*Asplenium trichomanes* L.) fertile and with erect structure was recorded, together with true mosses and algae; dark zone, with a complete and permanent darkness (C3: 20 m from the entrance) just after the last photosynthetic organism (Algae). After C3 some weak indirect light was still recognizable from the human eye but no photosynthetic organisms were found and 0 lux were recorded by the luxmeter.

Temperature and relative humidity recorded have been measured in the three points during seasonal investigations. To evaluate the biodiversity of the cave three pitfall traps, containing fresh meat (chicken liver) as attractive and salt water for killing and preserve, have been placed in the three locations with increasing darkness (1, 2, 3) and left for about three continuous years (from 19/07/2011 to 18/03/2014), seasonally checked to evaluate their conditions, attractiveness and impact on the local ecosystem. To prevent damage to the deep cave ecosystem, deep cave biodiversity was evaluated qualitatively placing non-trapping meat baits in all along the cave every 50 m and checking them occasionally, recording the species observed. A small underground river located in the deepest part of the cave (about 200 m from the entrance) was investigated placing water traps (plastic bottles with meat) to check the presence of water macroinvertebrates during October 2011.

To evaluate the species exchanges between the cave an the external woodland 6 pit-fall traps have been placed in the hemicycle outside the cave during the same time-period in different environments: 3 (A1, A2, A3) at 2-5 m from C1 in a cave-like environment (rock slope with scarce erbaceous vegetation, Fig. 2) and 3 (B1, B2, B3) at 6-10 m when the rocky ground left its space for the earthly soil of the *Carpinus-Fagus* woodland (Fig. 3).

Figure 1. Perspective of the study area with the light gradient and the disposition of the three inner pitfall traps (C1, C2, C3) from the opening of the cave to the starting point of the permanent darkness, in the moment of maximum illumination.

Figure 2. Entrance of the cave of Ponte Subiolo with the maximum limit of the direct sunlight traced on the ground. Picture taken in a sunny day at 13:34, June 12, 2014.

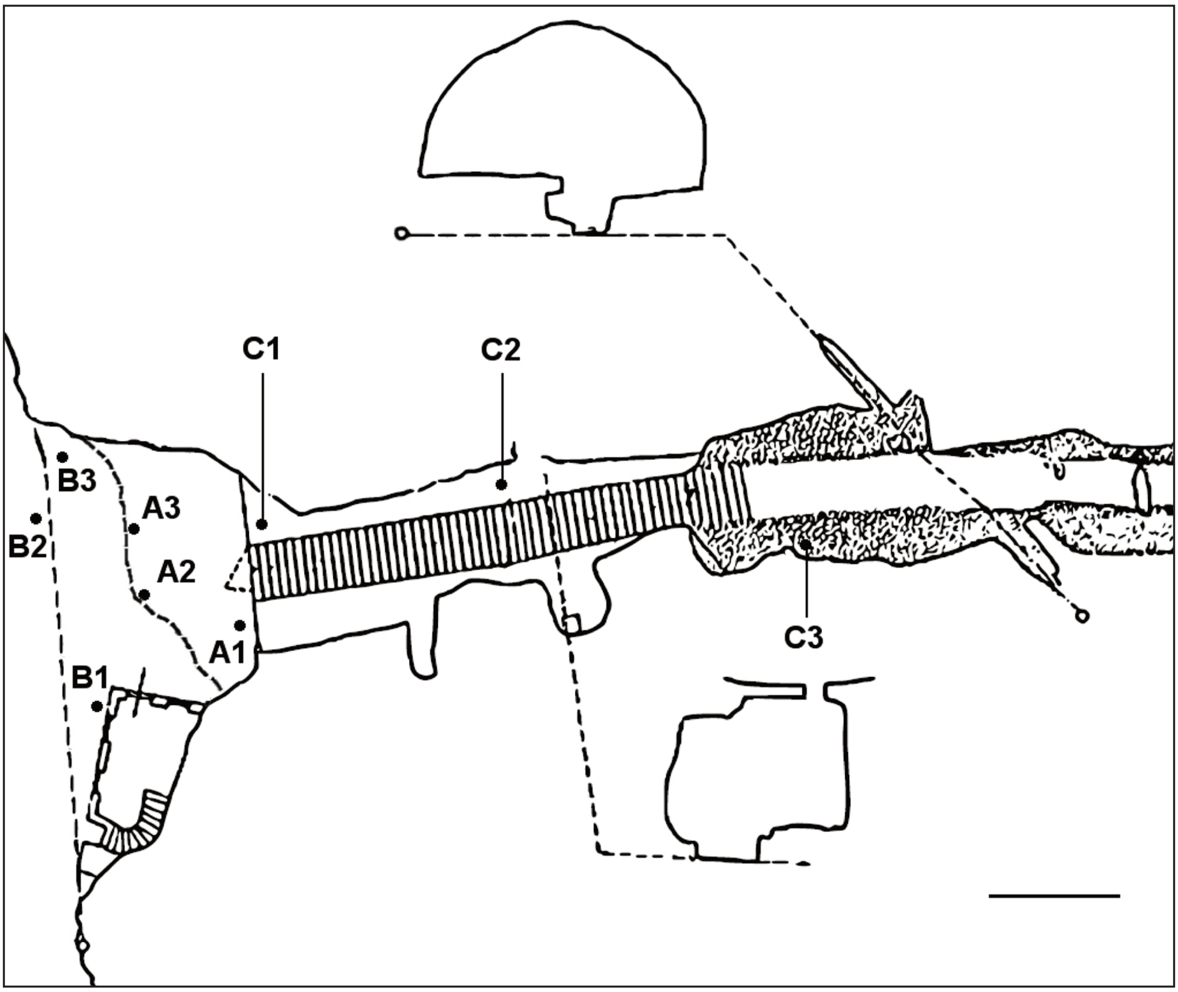

Figure 3. Land survey of the study area (courtesy Gruppo Grotte Giara Modon, modified) with the disposition of the three pitfall traps inside the cave (C1, C2, C3), the three pitfall traps under the outer cave ceiling (A1, A2, A3), the three pitfall traps on the woodland edge (B1, B2, B3). Scalebar 5m.

To compare the biodiversity in the light gradient Shannon-Wiener Index and Species Evenness were measured considering the specimens collected in the three cave pitfall traps. Mean values of Chao2 index (Chao, 1984) were measured with software EstimateS (Colwell, 2009) to evaluate the richness of unique species. Ecological categories (trophic habits and troglophily, here intended the progressive adaptation to form subterranean communities low or absent in Trogloxenes, moderate in Troglophiles, high in Troglobiont) were inferred using information available on the single species when available from literature and morphological characters (mouth parts, depigmentation, expansion of limbs or sensilla, etc.) directly observed. Since the ecology of all the species was not completely known Sket separation between eutroglophile and subtroglophile (Sket, 2008) have not been used here.

Taxonomical identification has been done here at Order level to separate main different functional strategies to live in a subterranean environment. Lower level of identification was used when necessary to better describe single units, and species level was used for all individuals collected as functional, morphological distinct units as "morphospecies" (SP1, SP2, etc...) to evaluate quantitatively their diversity and richness. Only adults or high vagile immatures (i.e. Orthoptera) have been considered to avoid bias due to direct egg-layings inside the traps occurred by some Diptera. Coleoptera have been here considered as walker as their primary moving strategy since all the species collected were linked to the subterranean environment where flight, when available, is used occasionally. The Analysis of Variance at significance 0.05 has been used to compare the specimens and species collected in the three cave traps. Biomass have been evaluated by measuring the wet weight of the specimens, grouped by taxa, with an electronic balance with sensitivity 0.1g.

RESULTS

Physical air parameters (Temperature/Humidity) attested on a annual average difference between one trap and another of 4.53°C and 18%: 4.50/17% from the external area to the entrance of the cave (C1), 4.85/18% from C1 to C2 and 4.25°C/19% from C2 to C3, reaching the nearly constant absolute parameters for the whole cave in C3 of 12°C/82%.

Maximum light measures were obtained during summer: 572 lux in the shadows of the external part of the cave, 127 lux at C1, 68 lux at C2 and 0 lux at C3.

The overall biodiversity in the three years of sampling attested on: 624 invertebrate specimens belonging to 35 different species collected in the three cave traps placed in the cave (Table 1). To these numbers must be added the autotrophs present in the study area: 4 different species belonging to: Magnoliophyta, Pteridophyta, Bryophyta, Chlorophyta, and the occasional presence Trogloxene/Troglophile vertebrates, *Vulpes vulpes* (Linnaeus, 1758), *Rhinolophus hipposideros* Bechstein, 1800, *R. ferrumequinum* Schreber, 1774, which visited the cave and rested there non-continuously for some days/months, the Troglophile spiders *Nesticus cellulanus* (Clerck, 1757) and *Pholcus phalangioides* (Fuesslin, 1775) never trapped but frequently observed in the area between C1 and C2, and at least one species of Fungi was observed occasionally. No invertebrates were collected during water samplings and no bacteria, protozoan or microinvertebrates were here considered. Deep cave observations recorded a progressive but not constant diminish in the number of species. The species collected in C3 were also observed till 80 m from the entrance, where the flyers disappeared. From 80 to 140 m only walkers have been observed and after 140 m where the floor of the cave is frequently submerged by interstitial water, no species were observed.

The total number of species recorded in the cave of Ponte Subiolo in the three years is 44.

Soil invertebrates species have been collected in almost equal numbers in the three cave traps (Table 2), not significant different in ANOVA one way test, both for the number of species (P: 0.69, 24 dof) and number of specimens collected (P: 0.32, 52 dof). A large part of the animals collected are detritivores, but the ratio with predators is close to 1 proceeding toward the darkness. Even if the trophic categories have been evaluated only by a descriptive point of view, the percent of Troglobiont species increased over the Trogloxene species from light to darkness, with a high and nearly constant number of Troglophile species collected in all the gradient, something expected for a cave transition zone, which confirms the goodness of the trapping procedure and the representativeness of the community considered in the analyses.

All these percents should be however considered in the vagility of their components which remained in nearly constant ratio (walkers/flyers) in all the cave traps (1.2, 0.9, 1.4). As expected, most of the Trogloxene species in traps C2 and C3 were flyers while all the Troglobiont species were much less vagile walkers. With the exception of one trogloxene Calliphorid fly (body parts occasionally found from the entrance to 80 m inside the cave), some Troglophile species were recorded alive in good numbers both in darkness as full light such as the flyers Diptera (Phoridae), Hymenoptera (Icneumonidae) or Trichoptera (Limnephilidae) as well as the walker millipede (Polydesmidae). Some troglobiont species, collected or observed more than twice (and supposed not to be occasional encounters), never reached the full light entrance of the cave (C1) as the Pseudoscorpion, *Neobisium torrei* (Simon, 1881), and the Isopoda, *Spelaeonethes nodulosus* Verhoeff, 1932, while the millipede, *Typhloiulus tobias* (Berlese, 1886), and the cave beetle, *Orotrechus targionii* (Dalla Torre, 1881), never passed the edge of darkness (C3).

Two invertebrates, the isopod, *Androniscus brentanus* Verhoeff, 1932, and the springtail (Collembola) were frequently observed on the rocks from the deepest of the cave to the entrance.

Trap	Taxonomic Group	Morpho-species	Individuals	Feeding	Troglophily	Vagility
C1	Aracnida	SP1	1	Predator	Trogloxene	Walker
C1	Aracnida	SP2	1	Predator	Troglophile	Walker
C1	Aracnida	SP3	1	Predator	Troglophile	Walker
C1	Acaroidea	SP1	1	Predator	Troglophile	Walker
C1	Diplopoda	SP1	5	Detritivore	Troglophile	Walker
C1	Collembola	SP1	7	Detritivore	Troglobiont	Walker
C1	Orthoptera	SP1	9	Detritivore	Troglophile	Walker
C1	Trichoptera	SP1	7	Detritivore	Troglophile	Flyer
C1	Coleoptera	SP1	1	Predator	Trogloxene	Walker
C1	Coleoptera	SP2	1	Detritivore	Trogloxene	Walker
C1	Coleoptera	SP3	1	Detritivore	Trogloxene	Walker
C1	Coleoptera	SP4	1	Detritivore	Troglophile	Walker
C1	Coleoptera	SP5	5	Detritivore	Troglophile	Walker
C1	Coleoptera	SP6	2	Detritivore	Troglobiont	Walker
C1	Hymenoptera	SP1	3	Detritivore	Troglophile	Flyer
C1	Hymenoptera	SP2	2	Predator	Troglophile	Flyer
C1	Diptera	SP1	85	Detritivore	Trogloxene	Flyer
C1	Diptera	SP2	3	Detritivore	Trogloxene	Flyer
C1	Diptera	SP3	1	Detritivore	Trogloxene	Flyer
C1	Diptera	SP4	1	Detritivore	Trogloxene	Flyer
C1	Diptera	SP5	1	Detritivore	Troglophile	Flyer
C1	Diptera	SP6	1	Detritivore	Troglophile	Flyer
C1	Diptera	SP7	1	Detritivore	Troglophile	Flyer
C1	Diptera	SP8	1	Detritivore	Troglophile	Flyer
C2	Aracnida	SP4	1	Predator	Troglobiont	Walker
C2	Aracnida	SP5	1	Predator	Troglobiont	Walker

Table 1. Specimens collected in the three cave traps with their taxonomical and ecological categories.

Trap	Taxonomic Group	Morpho-species	Individuals	Feeding	Troglophily	Vagility
C2	Pseudoscorpiones	SP1	2	Predator	Troglobiont	Walker
C2	Scorpiones	SP1	1	Predator	Troglophile	Walker
C2	Acaroidea	SP2	1	Predator	Trogloxene	Walker
C2	Diplopoda	SP1	18	Detritivore	Troglophile	Walker
C2	Hymenoptera	SP1	53	Predator	Troglophile	Flyer
C2	Hymenoptera	SP2	3	Predator	Troglophile	Flyer
C2	Diptera	SP1	126	Detritivore	Troglophile	Flyer
C2	Diptera	SP2	13	Detritivore	Trogloxene	Flyer
C2	Diptera	SP3	2	Detritivore	Trogloxene	Flyer
C2	Diptera	SP4	1	Detritivore	Trogloxene	Flyer
C2	Diptera	SP5	1	Detritivore	Trogloxene	Flyer
C2	Diptera	SP6	1	Detritivore	Trogloxene	Flyer
C2	Diptera	SP7	1	Detritivore	Trogloxene	Flyer
C2	Isopoda	SP1	4	Detritivore	Troglophile	Walker
C2	Isopoda	SP2	4	Detritivore	Troglobiont	Walker
C3	Aracnida	SP5	1	Predator	Troglobiont	Walker
C3	Aracnida	SP6	2	Predator	Troglobiont	Walker
C3	Pseudoscorpiones	SP1	1	Predator	Troglobiont	Walker
C3	Opiliones	SP1	1	Detritivore	Troglophile	Walker
C3	Diplopoda	SP1	2	Detritivore	Troglophile	Walker
C3	Diplopoda	SP2	5	Detritivore	Troglobiont	Walker
C3	Trichoptera	SP1	1	Detritivore	Troglophile	Flyer
C3	Coleoptera	SP7	7	Predator	Troglobiont	Walker
C3	Hymenoptera	SP1	70	Predator	Troglophile	Flyer
C3	Diptera	SP1	142	Detritivore	Troglophile	Flyer
C3	Diptera	SP2	14	Detritivore	Troglophile	Flyer
C3	Diptera	SP3	8	Detritivore	Trogloxene	Flyer

Table 1. Specimens collected in the three cave traps with their taxonomical and ecological categories.

Springtails always maintained their nearly-white/transparent pigmentation, and were never encountered under daylight so night habits are supposed. On the contrary isopods were occasionally observed even on the rocks, even under full indirect daylight with a slightly darker (pink) pigmentation respect the completely white-nearly transparent observed in the deep cave individuals.

Biodiversity measured in the three cave traps in terms of species evenness was almost equally distributed, with a light decreasing proceeding deeper in the cave. A bit more evident but not remarkable was the decreasing in the Shannon-Wiener Index, but significant in Species Richness halved from the light to the darkness (Table 3). This trend is maintained also when considered in a wider range of 50 computed repetitions where Chao2 slightly diverges (from 0 to 32%) from the species rarefaction curve (Fig. 4). Animal biomass from sampled invertebrates shows a non linear progression from light to darkness (Table 4) and is almost equally distributed between flyers (7.1 g) and walkers (7.2 g) but dominated by few species: the large and trogloxenic Calliphorid flies within the flyers and by the troglophilic/trogloxenic Diplopoda within the walkers.

In the external traps (A1, A2, A3, B1, B2, B3) 288 specimens were collected in 51 species, 14 of them (27%) found also in the traps inside the cave.

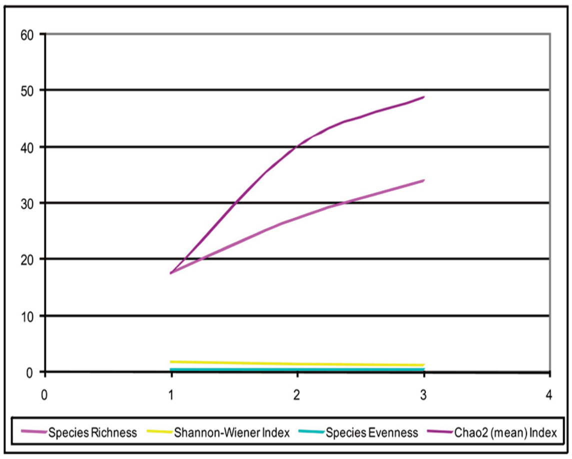

Figure 4. Comparison of the biodiversity estimators used, related to the number of individuals computed with EstimateS (x axis) in 50 repetitions, for to the three cave trap sites, and the number of species (y axis).

CONCLUSIONS

The light-gradient (together with other air parameters related to it) seems to have a moderate influence in changing on the overall subterranean biodiversity and biomass of the cave studied. Even if the length of the gradient here considered is remarkably long for a semi-natural karstic cave, the difference between the three cave traps in terms of biodiversity is quantitatively very low, but it should be considered also qualitatively. The slight decreasing of the number of species, proceeding from full light to full darkness, is the result of a replacement of species and strategies that makes the overall biodiversity almost constant: specialized deep cave species take the place of less specialized epikarst species.

In a model of a dynamic subterranean environment (Giachino &Vailati, 2010) the cave of Ponte Subiolo confirms again that some traditionally defined troglobiont species are generally not exclusive of the deep cave habitat. However it should be noted how the presence of light seems to be a real limit for others. Our results indicate that seems some species don't cross (or at least we can presume don't use to cross at day/season level) the line of permanent darkness (or limit for chlorophyll photosynthesis), forming separate subterranean communities. Chlorophyll photosynthesis stopped at 1 lux where the last alga was found and vascular plants stopped at 130 lux leaving the range between 130 to 50 lux to non vascular plants and the range between 50 to 1 to unicellular algae. If we exclude trogloxenic species collected or observed only once that may be related to occasional intrusions, very few species of the woodland habitat have been frequently found in the cave and almost all of them limited to the external part, and vice versa. We can presume that this barrier is not absolute but these records suggest that for some troglobionts, the migration rates from cave to cave are very low and should be considered in terms of many years or absent.

This is supported also by the historical records (Paoletti et al., 2009): in more than 20 years from 1992 to 2013 the invertebrate populations of the cave of Ponte Subiolo are nearly the same. Even if located in a karst area near other caves with different fauna, species contaminations and migration between caves seem to be extremely reduced. Large

Trap	Groups	Species	Specimens	Predators	Detritivores	Trogloxene	Troglophile	Troglobiont
1	9	20	142	25%	75%	33%	58%	8%
2	8	18	227	39%	61%	39%	33%	28%
3	8	12	254	42%	58%	8%	58%	33%

Table 2. Sum of the categories and abundance of the specimens collected in the three cave traps.

Trap	Species Richness	Shannon-Wiener Index	Species Evenness
C1	24	1.79	0.56
C2	17	1.50	0.53
C3	12	1.29	0.52

Table 3. Biodiversity estimators directly calculated on the sampled specimens.

	C1	C2	C3	Total
Diptera	1	3	2.3	6.3
Diplopoda	0.6	2.2	1.8	4.6
Orthoptera	1.4	n.p.	n.p.	1.4
Isopoda	n.p.	0.7	n.p.	0.7
Hymenoptera	<0.1	0.3	0.3	0.6
Coleoptera	<0.1	0.2	0.1	0.3
Aracnida	<0.1	0.2	<0.1	0.2
Trichoptera	0.2	<0.1	<0.1	0.2
Collembola	<0.1	0	0	<0.1
Total	3.2	6.6	4.5	

Table 4. Biomass measured in the three traps grouped by taxon. Values are reported in grams, "n.p." is for a taxon not present in the trap and "<0.1" is for the weights (wet) lower than the sensitivity of the balance.

invertebrates like *Troglophilus cavicola* (Kollar, 1833) or *Meta menardi* Latreille, 1804 can be commonly found in natural caves or military galleries in the surroundings (<2 km) but have never been collected in the Cave of Ponte Subiolo and vice versa large and easy to see invertebrates of this cave (*Gryllomorpha dalmatina* Ocskay, 1832 or *Typhloiulus tobias*) have never been observed or are extremely rare in the surroundings (Battiston unpublished data).

A single case does not allow any generalization but the topic structure of this cave and its history in its natural and anthropic context supports the idea that it should not be an exception.

By a methodological point of view comparing the records from the external traps with the internal traps show a remarkable abundance of specimens inside the cave represented by few species and the opposite outside. This can be due to the trap stability: more efficient in a protected environment and less in an open one subjected to rainfalls, interactions with large predators or scavengers or other unpredictable disturbing factors.

The almost gradual progression of this trend from the cave to the external area and to the woodland suggests however an increasing of dispersion and diversification of life. This should be carefully investigated in further studies and considered in its methodological implications.

The qualitative distribution of biodiversity observed under a light gradient has remarkable implications for the conservation of the subterranean environments; they seem to be stable by a qualitative point of view, but they may be not in a quantitative point of view, and any loss of species can have long term effect to the biodiversity and be a threat for the resilience of its ecological system. Dispersions and concentrations of species and individuals should be considered in assessing biodiversity both in subterranean and epigean contexts.

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REFERENCES

Chao A., 1984. Non-parametric estimation of the number of classes in a population. Scandinavian Journal of Statistics, 11: 265–270.

Colwell R.K., 2009. EstimateS: Statistical estimation of species richness and shared species from samples. Version 8.2.

Culver D.C., Deharveng L., Bedos A., Lewis J.J., Madden M., Reddell J.R., Sket B., Trontelj P. & White D., 2006. The mid-latitude biodiversity ridge in terrestrial cave fauna. Ecography, 29: 120–128.

Culver D.C. & Pipan T., 2009. The Biology of Caves and Other Subterranean Habitats Oxford University Press, Oxford, 256 pp.

Giachino P.M. & Vailati D., 2010. The Subterranean Environment. Hypogean Life, Concepts and Collecting Techniques. WBA Handbooks, Verona, 132 pp.

Caoduro G., Battiston R., Giachino P.M., Guidolin L. & Lazzarin G., 2014. Biodiversity indices for the assessment of air, water and soil quality of the "Biodiversity Friend" certification in temperate areas. Biodiversity Journal, 5: 69–86

Hill D.M., Fasham M., Tucker G., Shewry M. & Shaw P., 2005. Handbook of Biodiversity Methods: Survey, Evaluation and Monitoring. Cambridge University Press, 573 pp.

Juberthie C., Delay B. & Bouillon M., 1980. Extension du milieu souterrain en zone non-calcaire: description d´un nouveau milieu et de son peuplement par les Coléoptéres troglobies. Mémoires Biospéologie, 7: 19–52.

Latella L., Verdari N. & Gobbi M., 2012. Distribution of Terrestrial Cave-Dwelling Arthropods in Two Adjacent Prealpine Italian Areas with Different Glacial Histories. Zoological Studies, 51: 1113–1121.

Paoletti M.G., Celi M., Ciolat C., Tisat L., Faccio A., Del Re A.A.M. & Bacelli R., 2009. Cave Dwelling Invertebrates: Possible bioindicators of pollution in Italian caves. Konrad Thaler Memorial Book, Contributions to Natural History, 12: 1029–1045.

Sket B., 2008. Can we agree on an ecological classification of subterranean animals? Journal of Natural History, 42: 1549–1563.

Wilson E.O., 1988. The current state of biological diversity. In: Biodiversity, Wilson E.O (Ed.), Washington, D.C.: National Academy Press, pp. 3–18.