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Epikarst crustaceans from some Italian caves: endemisms and spatial scales

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SUMMARY

The epikarst crustacean fauna from four Sicilian caves (Conza, Entella, Molara, and Zubbia del Cavallo caves) and four caves in the Lessinian Massif in the Venetian Prealps (Covolo della Croce, Ponte di Veja, Roverè Mille, Buso della Rana caves) was recently investigated. The two groups of caves differ in their environmental conditions: the Sicilian caves are fossil except one which has an active branch; they are all fed by strongly intermittent and scarce rainfall peaking in the fall. The Lessinian caves are fed by more abundant rainfall, with two yearly peaks (May-June and October-November); two of them are active, one has a temporary stream, one is fossil. The crustacean fauna found in the epikarst drip of each of the studied caves is characterized by interesting endemic harpacticoid and cyclopoid copepods, and one bathynellacean syncarid, often collected in only one cave. Higher diversity of stygobiotic taxa was recorded for the Lessinian caves (9 species of copepods in the Lessinian, and 6 species of copepods and one bathynellacean in Sicily); most of the taxa collected in Sicily are endemic to one cave. Spatial analysis showed very different distributions over short spatial scales (tens of kilometers) and, within each cave, the distribution also varied over distances of a few meters. Our data correspond with other studies where many epikarst crustaceans showed a distribution with a linear extent of only a few hundred meters: the epikarst fauna is not uniformly distributed but rather divided in "blocks" probably characterized by different environmental conditions and, as a consequence, by different taxocoenoses. The data highlight the epikarst as a source of "hidden" biodiversity, and the importance

of management protection plans which include not only the caves, but also the epikarst overlying layer and the water sources that feed it.

INTRODUCTION

The epikarst is the topmost part of the vadose zone of karst and represents a boundary zone between the superficial unconsolidated material and the karstic habitats. The epikarst is extremely heterogeneous, with numerous cracks and crevices, and semi-isolated solution pockets (Bakalowicz 2004, Williams 2008). This mosaic of microhabitats hosts a variety of taxa, including a rich array of small crustaceans (Brancelj and Culver 2005, Pipan 2005), resulting in a diversity often rivalling the diversity of the rest of the karstic aquifer (Pipan and Brancelj 2004, Brancelj and Culver, 2005). The epikarst has been recently shown to host a high local richness and biodiversity in several European countries, Italy included (Pipan and Culver 2007a, Dole-Olivier et al. 2009, Galassi et al. 2009, Michel et al. 2009), and to be often a source of new and endemic species (Pipan et al. 2010). More specifically, the epikarst is a perched aquifer, "…partially saturated with water and capable of delaying or storing and locally rerouting vertical infiltration to the deeper regional phreatic zone of the underlying karst aquifer" (Jones et al. 2004); it is typically 3 to 10 m deep (Williams, 2008), but it can extend to a depth of up to 30 m (Klimchouk 2000). Water is transmitted mainly vertically as percolating water, which can passively or actively transport stygobiotic organisms, thus determining the alpha-diversity (within caves). However, sometimes water percolating through the epikarst moves laterally before finding a pathway deeper into the vadose zone (Pipan and Culver 2007b, and references therein), thus contributing to beta-diversity (between caves), although with increasing distance differences in epikarst diversity can be due to morphological and ecological characteristics of the entire watershed, such as size, connectivity and extension of fractures and solution pockets, epikarst development, land use, surface and subsurface capacity for water storage, degree of pollution (Moldovan et al. 2007). On a larger scale (different watersheds) historical events (colonization of epikarst habitats by surface-dwelling ancestors) and the hydrological and geological setting (isolation of aquifers) probably interact to determine gamma diversity.

The epikarst fauna is typically dominated by copepods (Pipan and Culver 2013, and references therein); at the scale of these organisms, the epikarst is highly heterogeneous in environmental parameters with many semi-isolated solution pockets whose water chemistry is also quite variable over short distances, with seasonal and even daily oscillations, due to changes in precipitations and to the geology and lithology of the vadose zone. The scale of the heterogeneity is such that, within a cave, several more or less distinct communities of copepods occur in the epikarst above it (Pipan and Culver 2007b; Meleg et al. 2011). Water exiting from the epikarst appears in caves as drip and seeps from ceilings and walls, often creating pools or rimstone pools (well above any streams that might be in the cave and well above the water table). This water represents the transition between the inaccessible epikarst together with the vadose zone and the pools of water occurring on the cave floor. Since drips are fed directly from the epikarst, the fauna of drips is the result of dislodgement of organisms which consequently drift in the water column, and the composition of the animals in drips should be an approximation of the fauna of epikarst. However, different species can have different susceptibility or propensity to enter the water column (based on their size, body shape, locomotory ability, developmental phase) and, as a consequence, the fauna of drip pools is an incomplete sample of the fauna of drips (Pipan et al. 2010).

Most research on epikarst fauna (i.e., fauna collected directly from drips) has been so far conducted in the Dinaric karst, a well-known biodiversity hotspot for groundwater fauna (Culver and Sket 2000). In Italy, studies on epikarst fauna are limited to a research covering a wide area of the Lessinian Massif in the Venetian Prealps (Galassi et al. 2009). Recently, we investigated the epikarst crustacean fauna from four Sicilian caves (two on carbonatic limestone, Conza Cave and Molara caves, and two on evaporitic gypsum, Entella and Zubbia del Cavallo caves) and four caves in

carbonatic limestone in the Lessinian Massif (Covolo della Croce Cave, Cave A del Ponte di Veja, Roverè Mille Cave, Buso della Rana Cave) (Zaupa 2008, Cottarelli et al. 2012, Bruno et al. 2017) (Fig. 1).

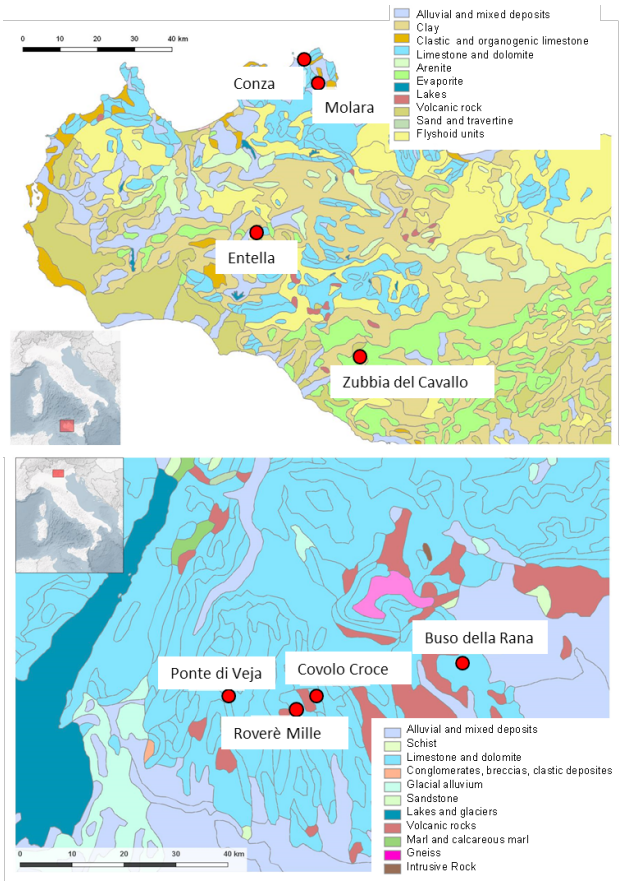

Figure 1. Location of the caves and lithological features of the study areas. Top: Sicilian caves. Bottom: Caves in the Lessinian Massif.

The scope of this paper is to investigate the epikarst fauna from Italian caves from two different areas, characterized by different climatic (rainfall and hydrological regimes), geological (limestone and gypsum) and zoogeographical (Alpine and Sicilian Faunistic Provinces sensu Ruffo and Vigna Taglianti, 2000) characteristics. We analyzed the two groups of caves separately, to assess their microcrustacean distribution over different spatial scales (from few meters, i.e., within a cave, for the Sicilian caves, to tens of kms, i.e., among caves, for the Lessinian caves), and to provide faunistic data for groundwater fauna of the studied areas which, in the case of Sicily, have been little investigated so far.

METHODS

Four caves were investigated in Sicily: Conza and Molara caves are about 5 km apart, in the same karstic limestone formation (Sciacca Formation) in the Billiemi mountain range near Palermo. Two gypsum caves are located in the Messinian evaporites successions in central and southern Sicily: Entella cave in a small evaporitic outcrop in central-western Sicily about 40 km south of Palermo, and Zubbia (Zubbia meaning "cave" in local dialect) del Cavallo in the largest Sicilian gypsum karst outcrop about 43 km southeast of the previous one (Fig. 1). The cumulative monthly rainfall for the sampled years recorded by the "Osservatorio delle Acque Regione Siciliana", and the "Unità Operativa Specializzata SIAS Sicilia Occidentale" from meteorological stations near Conza and Entella caves, respectively, shows a seasonal trend with a period of low or no precipitation extending from May to August, and peaks in rainfall in the fall and spring.

Conza Cave (38°11'13.9"N, 13°16'44.2"E, cadastral number Si Pa 60) develops within Upper Triassic limestone rock, it opens at 175 m asl, and extends for a total length of 100 m, sloping 30° upwards with a 30 m height difference, with reduced ceiling thickness (about 14 m). Although the cave is karstic, its morphology has been influenced by the presence of argillite in the numidic flysch underlying the carbonatic layers and of fractures and faults. Such discontinuities increased the permeability of the soluble rock, the water infiltration and lead to increased rock dissolution, while the impermeable argillite layer blocked the water, which was retained in the carbonatic layer, and boosted even more the karstic dissolution. The cave is strongly affected by outside thermal excursions, due to the cave small size (Mannino et al. 1986, Spena 2007). There is no running water in the area, and water input to the cave is exclusively due to percolation of rainfall into the cave. The accumulation of rainfall and percolation water forms few temporary puddles at the entrance of the cave and the drips form rimstone pools inside the cave. The rimstone pools are isolated and their structure and position prevent them from even accidentally being filled from water collecting in the cave. Monitoring of the rimstone pools was conducted in 2010-2013, showing that from late spring to the fall the rimstone pools are constantly dry; some of them are dry through the winter; samples were collected from five rimstone

pools from 2009 to 2011 (Fig. 2, Tabs. 1, 2). More details of pools environmental features in Cottarelli et al. (2012).

Molara Cave (38°08'47.30"N, 13°18'17.57"E, cadastral number Si PA 53) is karstic, formed in coral limestone of the Jura-Triassic (Mesozoic Era); it opens at an altitude of about 90 m asl on a large outcropping of about 500 m2, the cave develops predominantly horizontally with one large room about a hundred meters long, which is divided into a sequence of smaller rooms from large breakdown deposits on a muddy clay-rich soil. The epikarst rock layer is about 15-20 m thick. As for Conza cave, due to the karstic conditions, there is no running water in the area, and water input to the cave is exclusively due to percolation of rainfall, which drips extensively from the ceiling to saturate the clay soil, and all the pools of the cave remain wet throughout the year. Samples were collected from three pools and three drips (Fig. 2, Tabs. 1, 2).

Entella Cave (37°46'45.15"N, 13°06'46.93"E, cadastral number Si Pa 310) is one of the largest caves in Sicily, is located on an outcrop, the "Rocca di Entella", formed by gypsum evaporitic layers 2-3 m thick dating back to the Messinian age about 6 MYA (Catalano 1986). The cave originated from tectonic shifts and resulting breakdowns, and karstic dissolution. The cave opens at the bottom of the western side of the outcrop, at 388 m asl, and develops linearly along a main fracture for about 600 m; it is characterized by narrow passages, deep shafts, collapse rooms partially filled by breakdown deposits, and carbonate speleothems in the distal part of the cave.

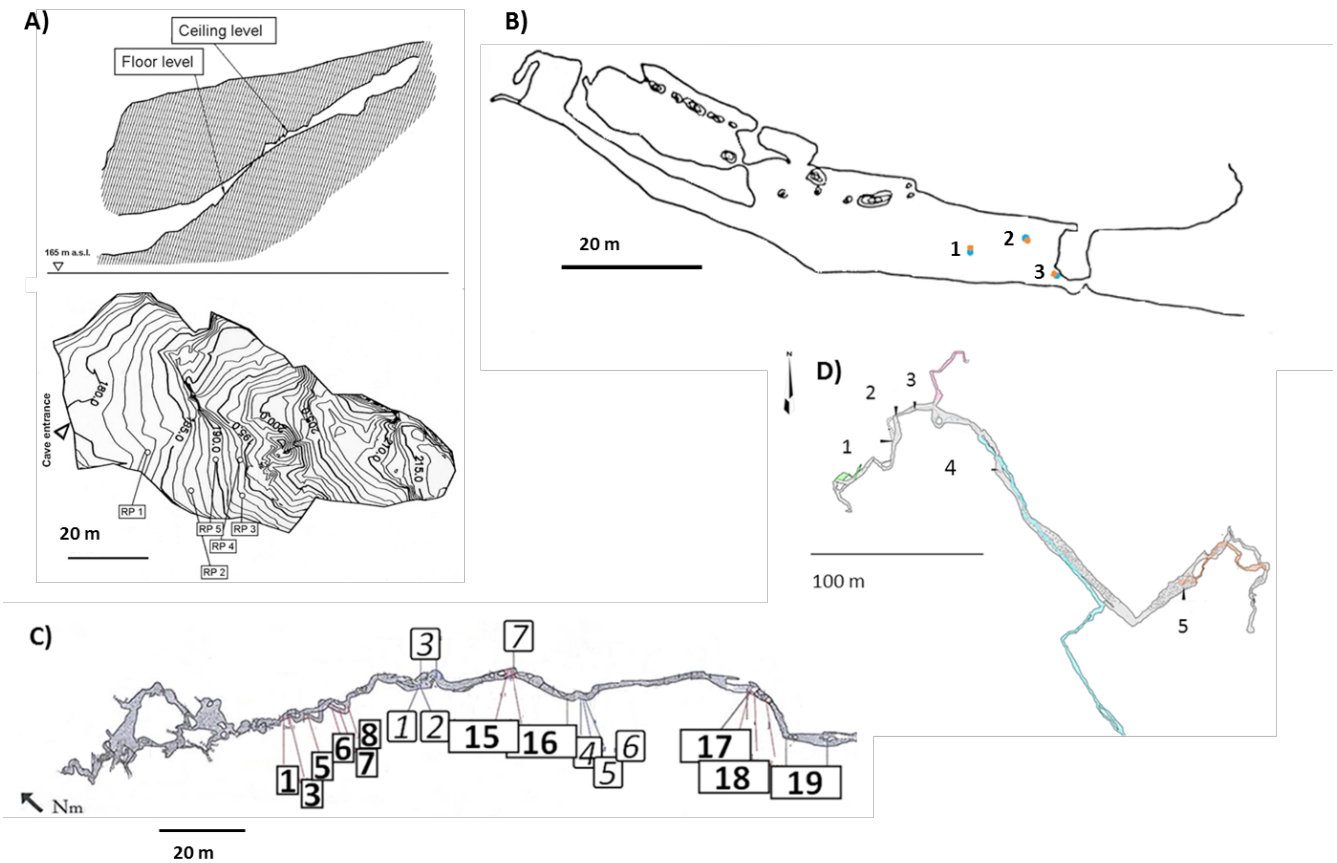

Figure 2. Sicilian caves: A) Conza Cave. Top: longitudinal section along the cave main axis; bottom: horizontal section and, topographical relief and position of the rimstone pools (from: Cottarelli et al. 2012); B) Molara Cave. Longitudinal section along the cave main axis; blue: drip pools; orange: drip funnels; C) Entella Cave. Longitudinal section along the cave main axis; bold: drip pools; italics: drip funnels; D) Zubbia del Cavallo. Longitudinal section along the cave main axis; numbers: drip pools.

Table 1. Details of the sampling stations (pools and /or drips), collecting dates (for pool samples) or filtering periods (for drip samples) and hydrological conditions, for each investigated cave.

	Collecting stations	Collecting dates/periods	Hydrological conditions
Conza Cave	Five rimstone pools distributed over a maximum range of 26 m (Fig. 2A, Tab. 2)	Monthly, from October to December 2009 to April 2011, and once in each pools after it refilled (Supplementary Table)	By December 2009 pool 5 was dry; the remaining pools dried later on, were dry by August 2010, and remained dry until October 2010 (Cottarelli et al. 2012)
Molara Cave	Three pools fed by dripping water and water collecting on the impermeable clay soil. Three drips, one near each pool (Fig. 2B). Pools and drips at a maximum distance of 12 m (Fig. 2B, Tab. 2). Pools 2 and 3 located in areas with more intense drip, which often overflew from the collecting buckets to feed the pools	Monthly from April 2013 to April 2014 (i.e., approximately 30 days filtration for each drip sample)	Pools 1 and 2 dry respectively from end of July to November 2013, and from end of September to November 2013, but water was always dripping and filling the buckets
Entella Cave	Eleven pools and seven drips distributed over a length of about 160 m (Fig. 2C, Tab. 2). The pools are filled with water throughout the year through drip and water surfacing from the floor	Monthly from October 2012 to April 2014 (i.e., approximately 30 days filtration for each drip sample) from seven pools and seven funnels; from November 2013 four more pools were added	Collections in the upper branch of the cave, where drip is constant throughout the year
Zubbia del Cavallo	Five drip pools, distributed over about 250 m distance (Fig. 2D, Tab. 2)	06 July 2014 and 31 August 2014	
A Ponte di Veja Cave	Five drips (Fig. 3)	8 filtering periods: 05.X.2007–04.XI.2007; 04.XI.2007–15.I.2008; 15.I.2008–27.III.2008; 27.III.2008–04.IV.2008; 04.V.2008–10.VI 2008; 10.VI.2008–22.VII.2008; 22.VII.2008–27.VIII.2008; 27.VIII.2008–01.X.2008. 323 days	27/08/2008: one funnel dry
Covolo della Croce Cave	Four drips (Fig. 3)	4 filtering periods: 22.IV.2008–10.VI.2008; 10.VI.2008–22.VII.2008; 22.VII.2008–27.VIII.2008; 27.VIII.2008–01.X.2008. 164 days.	27/08/2008 and 01/10/2008: one funnel dry
Roverè Mille Cave	Four drips (Fig. 3)	4 filtering periods: 09.IV.2008–09.V.2008; 09.V.2008–03.VII.2008; 03.VII.2008–04.VIII.2008; 04.VIII.2008–01.X.2008. 176 days	01/10/2008: one funnel dry
Buso della Rana	One drip (Fig. 3)	1 filtering period: 04.V.2008–10.V 2008. 36 days	

Table 2. Distance (m) between pairs of rimstone pools, funnels, pools and funnels, Sicilian caves.

Entella Cave		Conza Cave		Molara Cave		Zubbia del Cavallo	
Pools 1-3	3	Pools 1-3	26	site 1-2	8	Pools 1-1 bis	7
Pools 3-5	6.5	Pools 1-4	23	site 1-3	12	Pools 1bis-2	10
Pools 5-6	12	Pools 1-5	17	Site 2-3	6	Pools 2-3	12
Pools 6-7	2.5	Pools 3-4	8.5			Pools 3-4	70
Pools 7-8	5	Pools 3-5	11			Pools 4-5	155
Pools 8-15	50	Pools 4-5	7				
Pools 15-16	1.5						
Pools 16-17	70						
Pools 17-18	2						
Pools 18-19	6						
Pool 1-funnel 1	47						
Funnels 1-2	3						
Funnels 2-3	8						
Funnels 3-4	38						
Funnels 4-5	4						
Funnels 5-6	2						
Funnels 6-7	22						

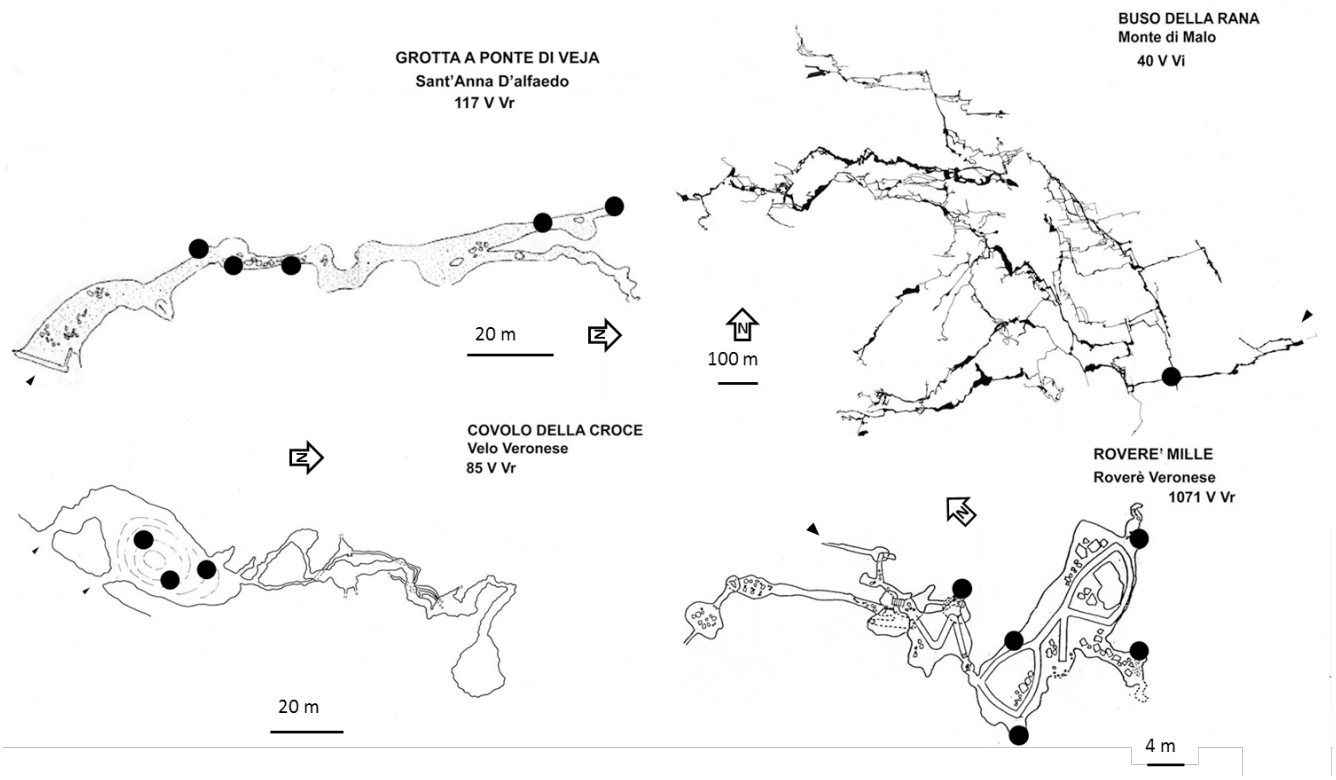

Figure 3. Location of the four caves, Lessinian Massif: longitudinal sections along the caves main axis; dots: location of the drip funnels.

The epikarst layer is about 200 m thick. The cave is fossil, it is never flooded, and the water input to the cave is due to rainfall expressed as temporary drips. Water also surfaces into the cave floor from a set of faults and collects in pools. Hence, pools can be fed only from surfacing water, only from drip, or from a mix of the two. Samples were collected from a total of eleven pools and seven drips (Fig. 2, Tabs. 1, 2).

Zubbia del Cavallo Cave (37°28'59.38"N, 13°25'58.25"E, cadastral number Si AG 2087) opens in the largest Messinian evaporite outcrop of Sicily. Evaporites are composed of a succession of evaporitic limestone, gypsum, and salt, with many intercalations of clays, marls, and carbonates (Catalano 1986). The cave opens at 200 m asl, and develops linearly for about 150 meters before forking in two branches: a lower active branch, and an upper fossil branch; both branches are characterized by abundant drip (much more intense than in Entella cave) and wet floor. The epikarst layer is about 40 m thick. Samples were collected from 5 drips (Fig. 2, Tabs. 1, 2).

Water samples were collected in one occasion in Conza, Entella and Molara caves from some of the pools and funnels, and analyzed for hardness, main cations and anions concentration.

Four caves were investigated in the Lessinian Massif in the Venetian pre-Alps of northern Italy (Fig. 1). Three of them (Covolo della Croce, Roverè Mille, Ponte Veja) are in a distance range of 5-9 km from each other, while Buso della Rana is 23-31 km from the previous cluster. All caves are in the same main watershed (Adige River), but are in valleys of different subcatchments: Vajo della Marciora (Ponte di Veja), Valle del Covolo (Covolo della Croce), Rana Stream (Buso della Rana, from which the stream originates). Roverè Mille is not in a valley or near a river, it opens on Mount Capriolo. The geological structure of the massif is dominated by carbonate rocks of Cretaceous and Jurassic ages in the northern part, with a basal 1000-m deep dolomite stratum, covered by several limestone banks of different origin, reaching a thickness of about 550 m. Galassi et al. (2009) reported how the hydrological regime of the Lessinian Massif is mostly governed by meteorological events, with infiltrating rainfall flowing "vertically through the vadose zone of the karstic massif and reaches the deep saturated aquifers in a few hours to a maximum of a few days through an extended network of large and small fractures in the limestone strata". The cumulative monthly rainfall for the sampled years recorded by Meteotrentino (www.meteotrentino.it), from Ala meteorological station, show abundant rainfall throughout the year, with peaks in the late fall and spring. In all caves, epikarst fauna was collected only with drip funnels, with schedule/number of funnels differing among caves (Tabs. 1, 2), approximately monthly from May to October 2008, except in Ponte di Veja where sampling started on October 2007.

Covolo della Croce cave (45° 36' 36.2"N 11° 07' 11.5"E, cadastral number 85 V Vr) opens on the left side of Valle del Covolo (Covolo Valley), at 875 m asl, it is 456 m long, downsloping with a height range of -9 m. The cave develops into grey limestone formations, three funnels were placed in the entrance room (Fig. 3, Tabs. 1, 2). The epikarst layer above the cave is very thin, and roots of the surface vegetation are visible on the ceiling. An hypogean stream flows in part of the cave.

Ponte di Veja Cave (45° 36' 29.5"N 10° 58' 12.3"E, cadastral number 117 V Vr) is one of the five galleries branching from an ancient cave, whose ceiling collapsed due to karstic erosion. The cave is sub-horizontal, 180 m long, it opens on the north-eastern side of the Ponte di Veja arch at about 600 m asl. Pleistocene deposits fill the entrance gallery floor, with a thickness of 6 m at the entrance and 3 m near the lower passage. The cave entrance is developed into the San Vigilio oolitic limestone formation, overlaid by a layer of Rosso Ammonitico Veronese, and of Biancone limestone near the surface. A small stream, dry in summer months, runs through the cave. Five drip funnels were placed along the main gallery and in one of the terminal branches (Fig. 3, Tabs. 1, 2) where the epikarst layer is about 50 m thick.

Roverè Mille Cave (45° 35' 31.1"N 11° 04' 446"E, cadastral number 1071 V Vr) opens at 1005 m asl, on a small cliff on Mt. Capriolo, in a highly karstified area. The cave develops into San Vigilio oolitic limestone, for about 300 m. The entrance is a 7 m deep wall, leading to a downsloping large room, and to side wells and a main room, about 30 m long and characterized by abundant speleothemes; four drip funnels were placed there and in the first room (Fig. 3, Tabs. 1, 2). The cave is fossil, all the water originates from drips, the epikarst layer above the sampling sites is about 10 m thick.

Buso della Rana Cave (45°39′00.32″N 11°21′46.79″E, cadastral number 40 V Vi) is the

largest in the region, developing sub-horizontally for 26 km, opening at 340 m asl with a large room at the base of a cliff. The cave entrance is the main spring of the karst system forming the Rana Creek. The main branch, 1800 m long, leads to a subterranean lake and a first branch, where we placed the funnel. The cave continues in a network of galleries (Fig. 3). The cave lies on an impermeable basalt layer, it opens into Oligocene Castelgomberto calcarenite, which has a maximum thickness of 200 m and are formed by massive beds rich in foraminifera and nummulites. The only drip funnel was placed in the cave (Fig. 3, Tabs. 1, 2) in an area where the ceiling is about 150 m thick.

In all caves, at each sampling occasion, all the water filling the pools was collected with a syringe. Dripping water was collected with a funnel and a bucket device (herein: drip funnel), similar to the one described by Pipan and Brancelj (2001, 20013) and Brancelj (2004). All samples were sorted under a stereomicroscope and preserved in 70 % ethanol or molecular biology grade absolute ethanol. Crustaceans (Copepoda and Bathynellacea) were studied at the Department for Innovation in Biological, Agro-food and Forest systems, Tuscia University, Viterbo, and at the Edmund Mach Foundation, Research and Innovation Centre, S. Michele all'Adige. Taxonomic and phylogenetic results have been published elsewhere (Cottarelli et al. 2012; Bruno et al. 2017).

In the Sicilian caves, the volume of each funnel/pool was measured at each occasion, allowing the expression of taxa abundance in N. ind. L-1. For the Conza and Zubbia del Cavallo caves, the similarity between pairs of sampling sites (drips and/or pools) were calculated with the Bray-Curtis similarity Index over mean values per pool/drip; for the remaining two caves only one copepod species was collected in all samples, and no similarity could be computed. For Zubbia del Cavallo, data were used to run a cluster analysis based on Jaccard Similarity Index and group averaging. In the Lessinian caves, volumes were not constantly measured, and the number and length of collections differed among caves, therefore, the abundance was expressed as N. ind/funnel/day. The high number of taxa collected allowed comparing the epikarst communities composition among caves (i.e., β diversity), by calculating a similarity matrix between caves based on the Jaccard similarity Index and use it to run a cluster analysis.

RESULTS

Sicilian caves: diversity spatial patterns within caves

The Molara-Conza caves - The copepod fauna of Conza cave (Tab. 3) was represented by the stygobiotic cyclopoid *Speocyclops italicus* Kiefer 1938, and the stygobiotic harpacticoids *Stammericaris diversitatis* (Cottarelli and Bruno 2012) and *Bryocamptus (Rheocamptus) stillae* Cottarelli and Bruno 2012. *Speocyclops italicus* is endemic to Italy; this species was first described by Kiefer (1938) from Castelcivita Cave (Salerno Province, southern Italy) but it is widely distributed in central and southern Italy in several groundwater habitats, although in caves, according to Stoch (2005), *S. italicus* is mainly present in percolating water. *Stammericaris diversitatis* and *Bryocamptus (Rheocamptus) stillae* are so far endemic to Sicily and to this cave (Cottarelli et al. 2012). The results of three years of sampling (although with qualitative and temporally uneven methods) showed that the copepod fauna composition in the rimstone pools remained relatively constant: *B. stillae* was collected only in rimstone pools 1 and 3 (total abundance: 1469 ind L-1), *S. italicus* in pool 1 (total abundance: 133 ind L-1), *S. diversitatis* in pools 4 and 5 (total abundance: 58 ind L-1) (Fig. 4A). The Bray-Curtis similarity index calculated between pairs of pools measured zero except for pools 1-3 (value: 6) and pools 4-5 (value: 60). The approximate distance between pools is listed in table 2. The most similar pools (pools 4 and 5), characterized by the presence of *S. diversitatis*, are about 7 m apart, and are at about 30 m distance from the second group (pools 1 and 3), which are 26 m apart. In Molara Cave, collections were conducted on a monthly basis for one year: even if this cave is close to the previous one (about 5 km of linear distance) and with a comparable epikarst thickness (15-20 m), the direct collection of drips (besides the more traditional collection of pools) resulted in a high number of individuals belonging to *Stammericaris destillans* Bruno and Cottarelli 2017 (425 individuals in pools, 318 in drips, i.e., 2302 and 32919 ind L-1, respectively) a species recently described and endemic to the cave (Bruno et al. 2017). Specimens were collected with much higher abundances in drips than in the corresponding adjacent pool (Fig. 4B). Pool 3, which never dried, had highest total abundances. Site 1 and 3 are about 12 m apart, site 1 and 2 about

8 m, site 2 and 3 about 6 m (Tab. 2), but the density of dripping copepods was very different: drip 1 had the lowest densities, which were 12-times higher in drip 2 and 21 times in drip 3.

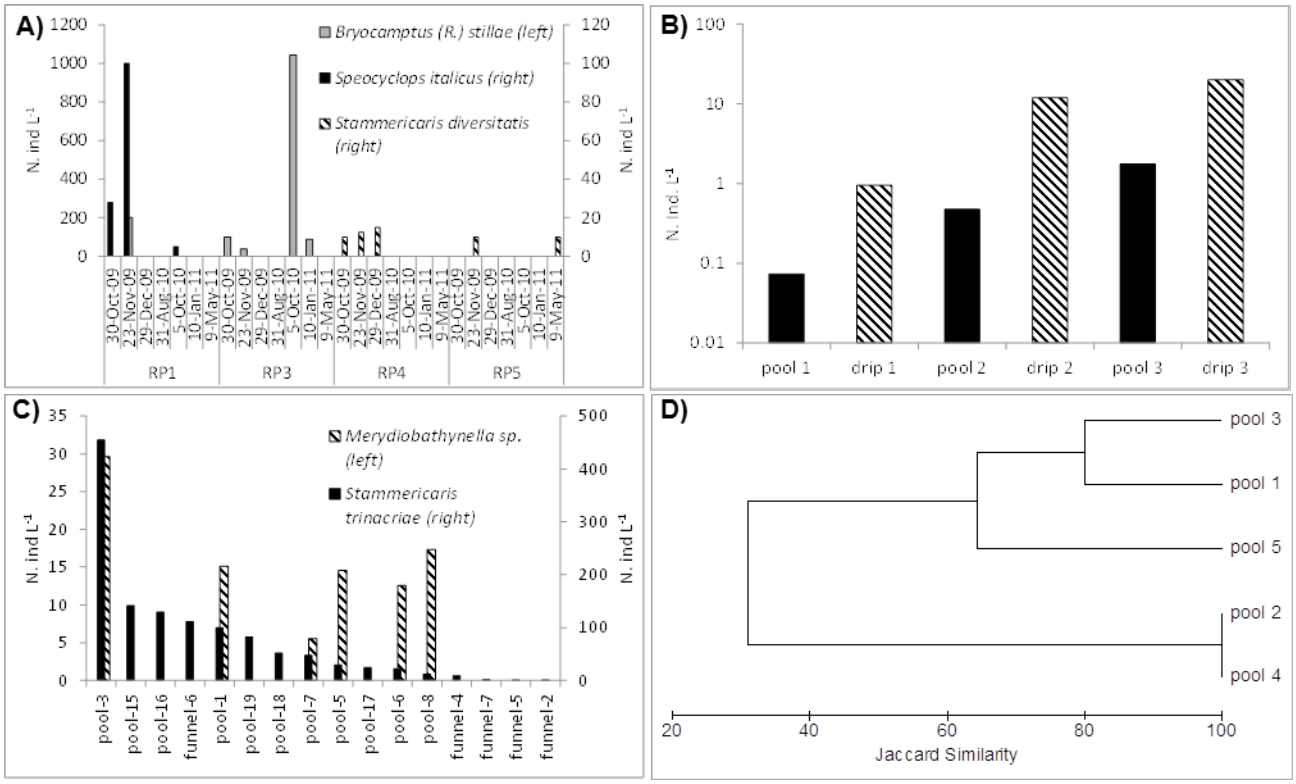

Figure 4. A) Conza cave: Density of collected copepods for each rimstone pool and sampling date; B) Molara cave: total density of *Stammericaris destillans* for each drip funnel and pool, calculated over all sampling dates; C) Entella Cave. Mean density of the two stygobiotic taxa, for each drip funnel and pool, calculated over all sampling dates; D) Zubbia del Cavallo. Cluster analysis of pool copepods. Collecting stations as in Fig. 2.

The Entella cave - The water dripping from the fractured upper layers of the Entella Cave hosts an unexpectedly abundant population of *Stammericars trinacriae* (Pesce, Galassi & Cottarelli, 1988), with a total of 8362 adults and larval stages (=17359 ind L-1), and 124 nauplii (384 ind L-1); 795 individuals (1706 ind L-1) of *Meridiobathynella* sp. were collected as well (Tab. 3). *Stammericaris trinacriae* is endemic to Sicily (Pesce and Galassi 1987; Pesce et al. 1988). Samples were collected monthly for 1.5 years in the upper branch of the cave, where drip is constant throughout the year, from the drips or from the pools, which are filled with water throughout the year through drip and water surfacing from the floor, or from a mix of the two. The two taxa had a clear distribution pattern over the small spatial cave scale (distance from the first to the last pool: 159 m, Tab. 2), related to their habitat. Collecting sites can be divided in three groups, based on the origin of their water. The funnels by design collect water only from the drips directly above them. *Stammericaris trinacriae* was collected in all these pools, whereas *Merydiobathynella* sp. was always absent (Fig. 4C). Pools 1-3-5-6-7-8 are fed by a mixture of drip, and water surfacing from cracks and faults, and flowing either on the floor or on the walls, and collecting in the pools. *Merydiobathynella* sp. was only collected in these pools; *Stammericaris trinacriae* was collected as well in all pools, with particularly high densities in pool 3. Pools 15-19 (Fig. 4C) are in a section of the cave developed in fractured limestone, where water flows quickly from the epikarst; all of them are fed only by water dripping directly on each one of them. Only *S. trinacriae* was collected there. Such different distribution over small spatial scale strongly suggests that the only habitat of *S. trinacriae* is the epikarst, whereas *Meridiobathynella* lives in the phreatic system. A

second harpacticoid species, the stygoxenes *Phyllognathopus viguieri* (Maupas 1892), was collected only in one sampling occasion in pool 3. This species is cosmopolite, with a wide ecological tolerance, collected from surface and groundwater, bromeliads and leaf litter. Given the one sporadic record, it is impossible to determine if this species, collected in a pool, originates from the epikarst, from the phreatic flow, or from the hygropetric habitat.

Table 3. List of taxa and number of individuals collected, their chorotype and endemism (following Stoch, 2005), and specialization to life in groundwater (following Gibert et al. 1994). Number of individuals for each cave, taxa recorded for the first time in each cave marked with asterisk. PV = Ponte Veja; CC = Covolo della Croce; ZC = Zubbia del Cavallo; PV = Ponte Veja; BR = Buso della Rana; EC = Entella Cave; MC = Molara Cave; CoC = Conza Cave. E = European; E-E = E-European; H = Holarctic; AL = Alpine; AP = Apenninic; AL-D = Alpine-Dinaric; AS-E = Asiatic-European; C = Cosmopolitan; STX = Stygoxene; STP = Stygophyle; STB = Stygobite. See Supplementary Table for more details.

Family	Species	Cave (n.ind.)	Chorotype	Endemism	Specialization
Canthocamptidae	*Bryocamptus (Rheocamptus) tatrensis* Minkiewicz 1916	PV (36), CC (1)	E-E		STP
Canthocamptidae	*Bryocamptus (Rheocamptus) zschokkei* (Schmeil 1893)	ZC (75)	H		STX
Canthocamptidae	*Bryocamptus (Rheocamptus) stillae* Cottarelli and Bruno 2012	CoC (360*)		Conza cave	STB
Canthocamptidae	*Lessinocamptus insoletus* (Chappuis 1928)	PV (25*), CC (1*)	AL	Lessinia	STB
Canthocamptidae	*Elaphoidella pseudophreatica* Sterba 1956	BR (3)	AL	Italy	STB
Canthocamptidae	*Elaphoidella* cf. *phreatica* (Chappuis 1925)	ZC (185)	AP		STB
Canthocamptidae	*Elaphoidella* sp. A (sp. A in Ruffo and Stoch 2006)	BR (8)	AL	Italy	STB
Canthocamptidae	*Ceuthonectes serbicus* Chappuis 1924	BR (1*)	AL-D		STB
Canthocamptidae	*Maraenobiotus brucei* (Richard 1898)	PV (44*)	E-E		STB
Canthocamptidae	*Moraria (Moraria) poppei* (Mràzek 1983)	PV (1*)	E		STX
Canthocamptidae	*Moraria (Moraria) stankovitchi* Chappuis 1924	PV (1*)	AL-D		STB
Canthocamptidae	*Moraria sp. A (*sp.A in Ruffo and Stoch 2006)	PV (1*), CC (3*), RM (10*)	AL	Italy	STB
Phyllognathopodidae	*Phyllognathopus viguieri* (Maupas 1892)	EC (22*)	C		STX
Canthocamptidae	*Attheyella crassa* (G.O. Sars 1862)	ZC (6*)	AS-E		STX
Ameiridae	*Nitocrella* cf. *stammeri*	ZC (87)			STX
Parastenocarididae	*Stammericaris diversitatis* (Cottarelli and Bruno 2012)	CoC (18*)		Conza cave	STB
Parastenocarididae	*Stammericars trinacriae* (Pesce, Galassi and Cottarelli 1988)	EC (8362*)		Entella cave	STB
Parastenocarididae	*Stammericaris destillans* Bruno and Cottarelli 2017	MC (743*)		Molara cave	STB
Cyclopidae	*Paracyclops imminutus* Kiefer 1929	PV (13)	E		STP
Cyclopidae	*Speocyclops italicus* Kiefer 1938	CoC (36*)	AP	Italy	STB
Cyclopidae	*Speocyclops infernus* (Kiefer 1930)	PV (7 *), CC (4)	AL		STB
Cyclopidae	*Speocyclops* sp.	ZC (61)			STB

Table 4. Results of chemical analyses, Sicilian caves.

Cave		K^+	Ca^{2+}	Mg^{2+}	Na^+	Hardness °F	Cl^-	F^-	$SO4^{2-}$
Conza (10 April 2008)	mean pool	12.1	66.0	62.6	55.2	42.2	92.4	0.2	45.0
Entella (5 April 2014)	mean pool	145.0	579.8	125.3	206.9	196.4	287.5	3.5	1921.7
	mean funnel	154.4	592.5	119.9	208.2	197.3	352.4	3.3	1865.0
Molara (5 April 2014)	mean pool	0.1	58.1	2.3	9.3	15.4	15.9	0.5	5.7
	mean funnel		63.8	2.4	9.3		14.6	0.3	10.8

The Zubbia del Cavallo - The pools of the caves hosted the most diverse copepod fauna of all the Sicilian caves investigated so far (Tab. 3), with three stygobiotic taxa: *Elaphoidella cf. phreatica* (185 individuals in total, 325 ind L-1), *Nitocrella stammeri* Chappuis, 1938 (87 ind, 150 ind L-1) *Speocyclops* sp. (61 ind, 103 ind L-1), and two stygoxene species: *Attheyella crassa* (Sars G.O., 1863) (6 ind, 12 ind L-1) rare in the cave having been collected once, in one pool, and *Bryocamptus (Rheocamptus) zschokkei* (Schmeil, 1893) (75 ind, 138 ind. L-1). Although a similarity between pools can be detected (Fig. 4D), with pool 4 and 2, which are 80 m distant from each other, characterized by the presence of *Elaphoidella cf. phreatica* only, and pool 5 by the highest diversity (all taxa collected there, which is the only collection site for *Attheyella crassa*), striking differences on short distance as in the previous caves were not detected.

The physical and chemical characteristics of the water differed among the three caves (Tab. 4). Water of Entella Cave was characterized by high concentrations of cations (K+, Na+, Ca2+, Mg2+), anions (Cl-, F-, SO42-), with sulfates typically about 3 orders of magnitude higher than in the two carbonatic caves.

The Lessinian caves: diversity spatial patterns among caves

A total of eleven species (one cyclopoid and ten harpacticoids species, eight of which known to Science) were collected in the Lessinian caves; most of these species are stygobiotic and one species (*Lessinocamptus insoletus*) is endemic of the Buso della Rana (Tab. 3) (Stoch, 1997). Ponte di Veja, due to the higher number of drip funnels and longer collecting period, yielded the highest diversity (8 taxa), and Covolo della Croce the lowest (one species). Buso della Rana, even if sampled only in one occasion, and with one funnel, had a relatively high diversity, including the one above-mentioned endemic species. Most of the collected species were new records for each cave: six (out of eight total) species in Ponte di Veja, two species (out of three) in Covolo della Croce, one species in Roverè Mille (copepods had never been collected there before), one species (out of three) in Buso della Rana (Tab. 3). The Jaccard similarity index calculated among caves (and expressed as percentage similarity), was related to distance: Roverè 1000 and Covolo della Croce (at 3.8 km distance) were the most similar (Jaccard β: 25), sharing the presence of *Moraria* sp. A.; Ponte di Veja was very different from the previous two caves (Jaccard β: 12.5 with Roverè 1000, distance 8.7 km, and 50 with Covolo della Croce, distance 11.7 km), being characterized by the presence of *Maraenobiotus brucei*, *Paracyclops imminutus*, *Moraria poppei* and *Moraria stankovitchi*, not collected in any other cave. Buso della Rana was 100 % dissimilar from the other caves (distance: 31 km from Ponte Veja, 19.5 km from Covolo della Croce and 23 km from Roverè Mille), and characterized by the presence of the two *Elaphoidella* (*Elaphoidella* sp. A and *E. pseudophreatica*) and *Ceuthonectes serbicus*, exclusive to this cave. The pattern of similarity is detectable by the cluster analysis based on Jaccard similarity among caves (Fig. 5A). The limestone thickness, i.e., a proxy for the extent of the epikarst system and hence of the available habitat for copepods, was related to species number and copepod abundance, with the highest number of species (8) collected at Ponte Veja where the thickness is around 50 m, and the highest number of funnels/dates were sampled, and lower numbers at Covolo della Croce (4 species) and Roverè 1000 (1 species) where the limestone ceiling is about 1-3 m thick. Buso della Rana, even if sampled in one

occasion, with one trap, yielded 3 taxa, but with the highest number of individuals/funnel/day (Fig. 5B).

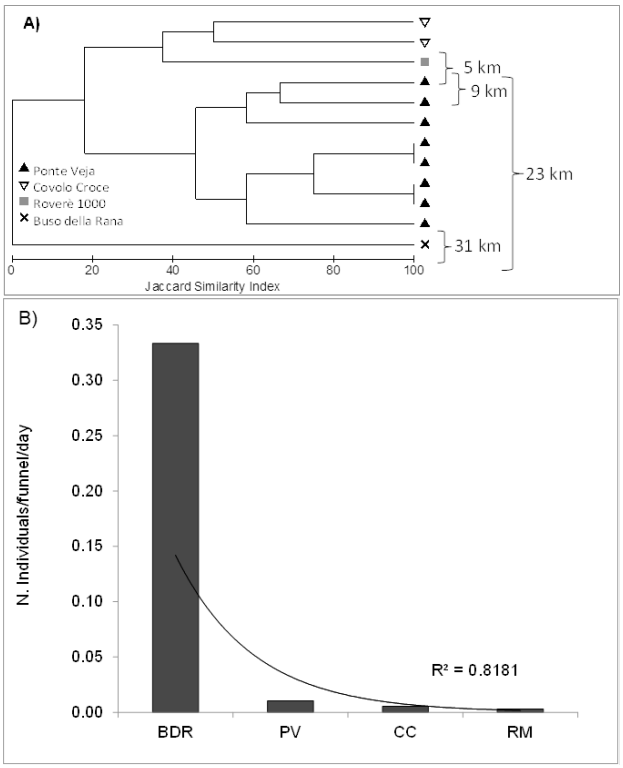

Figure 5. Lessinian caves: A) Cluster analysis of similarity among caves; B) Number of individuals collected per funnel, each day, and exponential trendline.

DISCUSSION

The number of pools/drips and the number of samples (i.e., temporal scale) of the different caves analyzed here are very different, as this paper compares records from different projects and different researchers. However, Pipan and Culver (2007a) showed that, at least for caves in the Dinaric Mountains of Slovenia, 3–4 months of continuous sampling were sufficient to find 90% of the species in a drip, that five drips were sufficient to find 90% of the species in a cave, and that five caves were sufficient to find 90% of the species in a region. Hence, the data discussed here can provide a good description of the epikarst copepod fauna of the investigated caves/areas.

Diversity and endemisms

The crustacean fauna found in the epikarst drip of each of the studied caves is characterized by interesting endemic harpacticoid copepods and cyclopoids, and bathynellacean syncarids, often collected in only one cave. Higher diversity was recorded for the Lessinian caves (12 species of copepods in the Lessinian, and 10 species of copepods and one bathynellacean in Sicily) with four species endemic to northern Italy (*L. insoletus*, *E. pseudophreatica*, *Elaphoidella* sp. A, *Moraria* sp. A); most of the taxa collected in Sicily are endemic to a single cave (*B. stillae*, *S. diversitatis*, *S. destillans*) or to Sicily (*S. trinacriae*).

In the Sicilian carbonatic caves, the number of stygobiotic taxa collected is lower than those recorded for much larger caves with more favorable hydrology and climate (for instance, caves in the Slovenian karst), or for caves of comparable extent, hydrology, and structure. For instance, in the Slovenian epikarst system, Brancelj (2002) recorded one cyclopoid and eleven harpacticoid taxa (including one Parastenocarididae) from 19 puddles and basins on eight sampling dates in Velika Pasjica, a 75 m long cave with ceiling thickness ranging from 2 to 10 m; Moldovan et al. (2012) recorded 4 species of harpacticoids (including one Parastenocarididae) and one cyclopoid from seven drips in Ciur Izbuc Cave in Transylvania (Romania), in the 425 m long active branch of the cave; Pipan et al. (2007b) recorded seven species of harpacticoids (including two Parastenocarididae) and three of cyclopoids from 13 ceiling drips in Organ Cave, West Virginia, a very large cave, but in a section with a ceiling thickness of 25 m.

Conza Cave hosts an interesting specialized fauna, with two taxa described from the cave (the stygobiotic harpacticoids *Stammericaris diversitatis* and *Bryocamptus (Rheocamptus) stillae* Cottarelli and Bruno 2012) and so far endemic to this cave, and a third species, *Speocyclops italicus* Kiefer 1938, endemic to Italy, but widely distributed in central and southern Italy in several groundwater habitats, although in caves according to Stoch (2005), *S. italicus* is mainly present in percolating water. Although in Conza cave we sampled only the rimstone pools, the faunistic results suggest that the copepods collected in the pools were dislodged from the epikarst and were carried by dripping water into the pools. In fact, considering that the rimstone pools of Conza Cave are only filled by percolating water and they are dry for long periods (see table 1 in Cottarelli et al. 2012), and because the existence of resting stages in the collected species has never been documented (thus excluding their survival in dry pools), the presence of these

copepods in pools immediately after they refill with dripping water would prove that the pools were colonized only by organisms dripping from the epikarst. Similarly, *Stammericaris destillans*, the only species collected in Molara cave, was collected with much higher abundances in drips than in the adjacent pool.

Entella cave was the first gypsum cave of Sicily investigated for groundwater copepod fauna; data from caves of this typology are scarce for the rest of Italy as well. Stygobiotic fauna was represented by *Stammericaris trinacriae*, a rare copepod, endemic to Sicily, originally described based on few specimens collected in 1986 and 1987 wells in Trapani, and a Syncarida, *Meridiobathynella* sp., a species probably new to science. The different distribution (drip vs pools) over small spatial scale recorded in the cave strongly suggests that the only habitat of this population of *S. trinacriae* is the epikarst, whereas *Meridiobathynella* lives in the phreatic system. The third microcrustacean collected in the cave, the stygoxene *Phyllognathopus viguieri* (Maupas 1892), is species with a wide ecological tolerance, collected worldwide from surface and groundwater, bromeliads and leaf litter, collected in Entella cave only in one occasion, it is impossible to determine if the presence in the pool originates from the epikarst, from the phreatic flow, or from the hygropetric habitat.

Zubbia del Cavallo is the second gypsum cave of Sicily; copepod biocoenosys are richer and more equally distributed in the pools and hence, in the epikarst system than in the other Sicilian caves, and particularly comparing this cave with Entella, the other gypsum cave, where only one species was collected. However, the very dry conditions occurring in Entella for part of the year probably act as a strong filter, selecting for fauna such as *Stammericaris trinacriae*, which, due to the very small size, can penetrate in the thinner crevices where they can find a suitable habitat, thanks to capillary action of the water percolating in the fractures of small diameter. Moldovan at al. (2011) showed (in karstic caves) that smaller and connected voids are typically inhabited by *Parastenocaris* sp. (a genus morphologically very similar to *Stammericaris*) and that such system allows for a fast transfer of water and animals from the surface or between levels of the subsurface zones, which might also explain the high abundance of *Stammericaris trinacriae* in the drip of this cave. The recorded species richness in Zubbia del Cavallo is comparable to that of other epikarst systems in carbonatic systems of Romania: for instance the estimated species richness including observed and unobserved species in five Romanian caves ranged from 3 to 6 (Meleg et al. 2011), and the number of stygobiotic copepods ranged from 2 in a Slovenian cave to 33 in a Romanian cave (Pipan and Culver 2013). In Sicily, although data are limited and more caves should be investigated, caves in evaporitic gypsum appear to host more diverse stygobiotic copepod assemblages than the carbonatic ones, as already observed by Stoch et al. (2009) for the groundwater fauna of upper Secchia Valley, in the Reggiano Apennines.

The collection of two new and one endemic species of *Stammericaris* in the Sicilian caves is particularly interesting, because the genus belongs to Parastenocarididae, a family highly specialized for life in groundwater, where these harpacticoids are often one of the dominant families. The comparison of distribution, abundance and habitat of the three species of Parastenocarididae indicates that on a short distance range, the heterogeneity in epikarst morphology and hydrology results in different propensities of copepods to drift away from the microfractures. The density of *Stammericaris* collected in the pools is much higher in Molara than in Conza cave (totalling 2302 and 58 ind L-1, respectively) notwithstanding the similar habitus of the two species, which, due to their reduced size and peculiar body shape can probably penetrate in very small fractures of the rock, not accessible to the other copepods. This suggests that *S. destillans* has a higher tendency to drift than *S. diversitatis*. The latter species in fact, was rare in the rimstone pools compared to *B. (R.) stillae* and *S. italicus* (which are larger, longer, stouter and therefore less flexible organisms), possibly due to the capillary action of the water percolating in the fractures of small diameter which helps the small Parastenocarididae to resist the traction force of water and avoid drifting, thus ensuring the survival in the epikarst (Cottarelli et al. 2012). *Stammericaris trinacriae*, on the other hand, was collected with higher densities than the former two congeners in Entella cave, showing an even higher tendency to drift than *S. destillans*. *Stammericaris trinacriae* was originally described from a few specimens collected from two wells in Trapani (Sicily, Italy) at a depth of 10.5 and 4.5 m, respectively, about 30 and 50 km west of the Entella

Cave (Pesce and Galassi 1987; Pesce et al. 1987, 1988) in a carbonate platform area; this species therefore has a quite wide distribution and the epikarst might not be its primary habitat. The high densities of this species which must be occurring in the epikarst of Entella, and in the underlying vadose zone, which is indeed very wide (the rock layer above the cave is about 200 m thick, with dripping water throughout the year) compared to the rarity of this species in the phreatic system of the type locality, could be related to the physical and chemical parameters of Entella Cave, which is characterized, among others, by high concentrations of K+, Na+, Ca2+, Mg2+. A positive correlation between distribution of epikarst specie, including an epikarst Parastenocarididae (*Parastenocaris cf. andreji*), and sodium concentration was reported by Pipan and Culver (2013) for the drip of a Slovenia cave.

The records of the epikarst species widen the knowledge on the distribution and geonemy of several taxa. The genus *Speocyclops* was unknown for Sicily until *S. italicus* was collected in Conza Cave; the collection of a second species of the genus in Zubbia del Cavallo and of a third species (Cottarelli, unpubl) in the hyporheic habitat of the Alcantara River widens considerably the distribution of this genus in Sicily. *Nitocrella stammeri* Chappuis, 1938 has a Mediterranean distribution, widely distributed in coastal subterranean habitats (anchialine caves, wells) of the Mediterranean (Italy, Spain, Greece, Turkey); this species was already recorded from several phreatic (wells) and hyporheic sites in Sicily (Cottarelli and Fasano 1979, Pesce and Galassi 1987), Zubbia del Cavallo is the first record in an inland cave. The morphological peculiarity of the specimens collected in this cave confirm what already mentioned by Pesce and Galassi (1987) for other Sicilian populations, i.e., that these might represent a different taxon, or a group of cryptic species. *Elaphoidella* is one of the largest freshwater genera, with about 200 species and subspecies presently known (Mori and Brancelj 2008), and more than 140 stygobiotic species living exclusively in the subsurface (Galassi 2001; http://www.marinespecies.org). This is the first record of *E. cf. phreatica* in Sicily; the nominal species is known from both phreatic and hyporheic habitats in central and northern Italy, and it is common in southern and central Europe, inhabiting predominantly porous aquifers (Mori and Brancelj 2008). Two other species of the genus have been reported from Sicily, but exclusively from phreatic waters (wells): *E. elaphoides* and *E. plutonis* (Pesce and Galassi 1987, Pesce et al. 1987).

The vadose zone of the Lessinian Massif is known to harbor high diversity of microcrustaceans, including many endemic species due to the ancient geological age of the aquifers, high habitat fragmentation, and isolation of microhabitats, factors, which concurred to promote speciation by vicariance (Galassi et al. 2009). The effects of Quaternary glaciations shaped stygobiotic species distribution, as the Massif was only marginally covered by ice (Latella and Sauro 2007), and the extensive networks of fractures of the karstic system represented a refuge for stygobionts, boosting isolation and speciation. A large number of taxa are known for the Lessinian groundwater habitats in general (Caoduro et al. 1995; Ruffo and Stoch 2006) and from the epikarst (Galassi et al. 2009). Nonetheless, our epikarst drip collections harbored species which had not yet been recorded for the caves and/or for the entire Lessinian massif. Specifically, in Ponte di Veja, where five species of copepods were already recorded, the new records are represented by *Paracyclops imminutus*, *Lessinocamoptus insoletus*, *Moraria poppei*, *Moraria stankovitchi*, *Moraria* sp. A. and *Maraenobiotus brucei* (a new record for the Italian fauna). All these species are STP/stygoxene except *L. insoletus*, belonging to a genus known only from the vadose zone of the Lessinian caves (Ruffo and Stoch 2006), with three knows species. *Lessinocamptus insoletus* was so far known only for Buso della Rana. The same species was a new record for Ponte di Veja where five taxa are known from literature, together with *Moraria* sp. A. This species, known from Buso della Rana (Ruffo and Stoch, 2006), was also the only copepod collected in Roverè Mille (where there are no previous records of copepods). Finally, in Buso della Rana, where 18 species were already known, we collected one additional specie, the stygobiotic *Ceuthonectes serbicus*, species recorded from caves in areas adjacent to the Lessinian (Stoch 2000-2004).

The lower diversity recorded in the Sicilian compared to the Lessinian caves is probably driven by ecological factors rather than biogeographical ones: environmental conditions such as high surface temperatures, high ionic concentration of the Sicilian caves might act as a filter for stygoxene taxa, and also for the colonization of these system

by stygobiotic taxa. Indeed, Moldovan et al. (2012) plead against the theory of accidental downward drift of individuals when voids are flooded and more for an active drift when water floods the voids; hence, water intermittency might act as the main driver of epikarst drip diversity.

Spatial scales: within caves

Many epikarst copepods are known to have a linear distribution within the epikarst zone of only a few hundred meters (Pipan et al. 2006a, Moldovan et al. 2011), and the epikarst fauna is not uniformly distributed but rather divided in "blocks" probably characterized by different environmental conditions and, as a consequence, by different faunal assemblages (Brancelj 2002, Meleg et al. 2011). Faunistic differences in groups of pools which reveal two spatially distinct groups of water bodies within a cave over a spatial distance of about 70 m is reported by Brancelj (2002) for Velika Pasjica cave in Slovenia, and for a distance of about 300 m by Moldovan et al. (2011) in Ciur Izbuc Cave in Romania. In Conza Cave, it is very likely that the epikarst is represented by two separated percolation units which feed the two groups of rimstone pools, and that these two "islands" could even be characterized by microfractures of different size, given the smaller size of *S. diversitatis* when compared with *B. stillae* and *S. italicus*. For Molara and Conza cave, a distance of 5 km is sufficient to separate two isolated epikarst systems. In Entella cave, differences were recorded as well over a linear distance of about 160 m, in Conza cave, over 30 m, in Zubbia del Cavallo over 80 m (even if the differences were not as striking as for the previous two caves).

Spatial scales: among caves

The analysis of the Lessinian cave copepods shows a distinct species composition for each of the caves, with Buso della Rana completely different from the other caves. Such differences are related to the geographical distance among caves: Buso della Rana is the most distant cave (23-31 km from the remaining caves), it develops in the Western section of the Lessinian massif (Monti Lessini Vicentini) while the remaining caves are in the Eastern section (Monti Lessini Veronesi). The three active caves (BR, PV, CC) are located in different subcatchments. Buso della Rana is therefore well-isolated from the other caves, there is no faunistic exchange with the other caves. In fact, lateral mobility of epikarst copepods is very low, often less than 100 m (Pipan and Culver 2007a), and their ability to actively colonize caves which are far apart is very poor. Lateral passive transport is not likely to occur on such large spatial scales, as Pipan and Culver (2013) reported that although drips within several hundred meters of each other tend to have similar composition, there is a decline in similarity with distance, and that the relationship between distance and dissimilarity breaks down when drips greater than 1 km apart are considered, and communities are substituted by new ones.

Other factors might be important in determining species distribution (as demonstrated from studies in Romania and Slovenia), including ceiling thickness, habitat connectivity and habitat size (Pipan and Culver 2013), and ecological conditions. In fact, drip water conductivity and pH differed among caves, and the only non-stygobiotic species collected (i.e., *B. tatrensis*, *M. brucei*, and *P. imminutus*) were collected in high conductivity and low pH drip water, i.e., with conditions closer to those of surface water. The highest abundance of copepods recorded in Ponte di Veja and Buso della Rana might be related to habitat connectivity and size: a recent study of Arena Cave (Latella and Sauro 2007), a cave developing in the Verona area of the Lessinian Massif, about 10 km north of the investigated caves and in the same rock outcrops of Ponte di Veja e al Buso della Rana reports high discontinuities among the rock layers forming the outcrops. Moreover, the upper layers of Ponte di Veja are partly built of Rosso Ammonitico, a highly soluble type of limestone, characterized by a dense network of rectangular fractures (Sauro 1973). The resulting voids can store water and, possibly, stygofauna.

CONCLUSIONS

Overall, our results underline how the drip funnels allow measuring accurately the diversity of the vadose zone, extremely relevant for the total estimate of aquatic hypogean fauna diversity, as already reported by several authors (Pipan 2005, Pipan & Culver, 2005, 2007a). The epikarst copepod fauna can contribute about 20% of the cave fauna diversity at the regional level, with copepods representing up to 50% of total stygofauna (Pipan and Culver 2007a). Epikarst copepod species richness is estimated to almost double that of the saturated zone of karstic aquifers (Pipan and Culver

2007a). Meleg et al. (2011) investigated five caves within two hydrographic basins in Romania and reported a different distribution inside caves for all recorded major taxonomic groups and species, some of which were found at only one sampling site, and crustacean community composition varying within caves and over a larger scale (i.e., between watersheds). These data are in accordance with our analysis of small and large scale faunistic assemblages of epikarst copepods.

The crustacean fauna found in the epikarst drip of each of the studied caves is characterized by high level of heterogeneity and local endemism in stygobiotic harpacticoid copepods and cyclopoids, and bathynellacean syncarids, often collected in only one cave. The spatial analysis showed very different distributions over short spatial scales (tens of kilometers) and, within each cave, the distribution also varied over distances of a few meters. Our data correspond with other studies: the epikarst fauna is not uniformly distributed but rather divided in "blocks" probably characterized by different environmental conditions and, as a consequence, by different taxocoenoses. The data highlight the epikarst as a source of "hidden" biodiversity, and the importance of management protection plans which include not only the caves, but also the epikarst overlying layer and the water sources that feed it.

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