



University of Sassari
Ph.D. School in Natural Sciences
Via Muroni 25, I-07100 Sassari, Italy

*Dissertation for the Degree of Doctor of Philosophy in Environmental Biology
presented at Sassari University in 2011
XXIV cycle*

**BIODIVERSITY ASSESSMENT IN MEDITERRANEAN CAVES:
THE CASE OF PORIFERA AS MODEL TAXON**

PH.D. CANDIDATE: *Dr. Barbara Cadeddu*

DIRECTOR OF THE SCHOOL: *Prof. Marco Apollonio*

SUPERVISOR: *Dr Renata Manconi*

Co-SUPERVISOR: *Dr. Fabio Ledda*



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Abstract

The status of marine caves as cryptic, fragmented and vulnerable biotopes require appropriate conservation measures and protection planning. These biotopes represent one of the most important natural heritages in the Mediterranean and recently they have been indeed inserted in the EU Habitat Directive.

These extreme environments are colonized by peculiar faunas composed of species able to tolerate to survive these conditions and characterized by selected adaptive strategies. In addition, the caves, as the islands are isolated and discontinuous habitats that limit or gene flow between populations, making the cave faunas of ideal subjects for speciation analysis.

The aim of present thesis is was to contribute to increase the knowledge, conservation and sustainable use of natural resources represented by the cave fauna of Sardinia, Sicily and the Mediterranean Sea with particular regard to the Porifera.

The collaboration with other research groups allowed to increase the expertise also on other cave-dwelling model taxa rich in endemic species such as the freshwater planarians (Tricladida, Turbellaria), and the Sardinian endemic *Speomolops sardous* (Coleoptera Carabidae).

Riassunto

Le grotte marine sono ambienti criptici, frammentati e vulnerabili che richiedono adeguate misure di conservazione e protezione. Questi biotopi rappresentano uno dei tanti importanti patrimoni naturali del Mediterraneo e recentemente sono stati infatti inseriti nella Direttiva Habitat dell'Unione Europea.

Questi ambienti estremi sono colonizzati da faune peculiari composte da specie capaci di sopravvivere a queste condizioni e caratterizzate da strategie adattative. In più, le grotte, come le isole sono habitat isolati e discontinui che limitano il flusso genico tra le popolazioni, rendendo le faune di grotta soggetti ideali per l'analisi di speciazione.

Lo scopo della presente tesi è quello di contribuire ad accrescere la conoscenza, la conservazione e l'uso sostenibile delle risorse naturali rappresentate dalla fauna di grotta della Sardegna, della Sicilia e del Mar Mediterraneo con particolare riguardo per i Poriferi.

La collaborazione con altri gruppi di ricerca ha permesso di aumentare la competenza su altri taxa modello di grotta ricchi di specie endemiche come le planarie d'acqua dolce (Tricladida, Turbellaria), e l'insetto endemico sardo *Speomolops sardous* (Coleoptera Carabidae).

Intoduction

The caves are precious laboratories for the study of life in atypical conditions. In these environments, in fact, there is a rapid light extinction determining the survival in the inner dark part of only heterotrophic organisms (e.g. bacteria, protozoans, animals). Since the photosynthetic organisms are present only in the entrance of the cave, where there is sufficient light, food chains of semi-dark and dark areas are based on secondary production processes, rather than primary.

These extreme environments are colonized by peculiar faunas composed of species able to tolerate to survive these conditions and characterized by selected adaptive strategies. In addition, the caves, as the islands are isolated and discontinuous habitats that limit or gene flow between populations, making the cave faunas of ideal subjects for speciation analysis.

Marine caves matching the category of midlittoral caves, semi-dark caves and dark caves harbour protected biocoenosis registered as Habitat II.4.3, Habitat IV.3.2 and Habitat V.3.2 (Relini & Giaccone, eds, 2009; Relini & Tunesi, eds, 2009). Appropriate conservation planning of these faunistic elements represents one of the challenges of the western Mediterranean protected areas following European Union directives (Natura 2000, Habitats Directive, Council Directive 92/43/EEC). Caves are one of the listed endangered habitats (code 8330) under the CORINE Biotopes Classification by the European Commission.

15 species of sponges are listed in the appendix II and III of the Barcellona Convention as ‘protected species of the protocol SPA/BIO’: i.e. *Aplysina aerophoba* (Nardo, 1833), *Aplysina cavernicola* (Vacelet, 1959), *Asbestopluma hypogea* Vacelet & Boury-Esnault, 1996, *Axinella cannabina* (Esper, 1794), *Axinella polypoides* Schmidt, 1862, *Geodia cydonium* (Jameson, 1811), *Hippospongia communis* (Lamarck, 1814), *Sarcotragus foetidus* (Schmidt, 1862), *Sarcotragus pipetta* (Schmidt, 1868), *Petrobiona massiliana* Vacelet & Lévi, 1958, *Spongia lamella* (Schulze, 1879), *Spongia mollissima* Schmidt, 1862, *Spongia officinalis* Linnaeus, 1759, *Spongia zimocca* Schmidt, 1862, *Tethya aurantium* (Pallas, 1766), *Thetya citrina* Sarà & Melone, 1965.

More than 3000 terrestrial and marine karstic caves have been recorded from Sardinia (Regional Speleological Register of Sardinia). They harbour a very rich faunal assemblage

characterised by both true cave dwellers and/or a peculiar relict fauna survived to the geological/climatic vicissitudes of the Mediterranean area. The hypogean fauna of Sardinia includes presently more than 500 specific taxa. Moreover the Regione Autonoma Sardegna, supports the UE directives in the field of protection of karstic areas (L.R. n. 4 del 07/08/2007 “Norme per la tutela del patrimonio speleologico delle aree carsiche e per lo sviluppo della speleologia”).

The first step to protect these particular environments is to assess the quantitative and qualitative biodiversity, in other words, the species richness.

The biodiversity measures based only on species richness can sometimes be limiting, since it captures only one biodiversity aspect as the species number and their spatial distribution. However, they are useful in practice because you can quickly identify which are the areas with more species that require protection measures.

The aim of this Ph.D. was to contribute to increase the knowledge, conservation and sustainable use of natural resources represented by the cave fauna of Sardinia, Sicily and, in general, of the Mediterranean Sea. For this project the phylum Porifera is the main model taxon. The collaboration with other research groups allowed to increase the expertise also on other cave-dwelling model taxa rich in endemic species such as the freshwater planarians (Tricladida, Turbellaria), and the Sardinian endemic *Speomolops sardous* (Coleoptera Carabidae).

The aims of this paper were:

- to contribute to the faunistic data collection and to construct databases
- to assess the taxonomic richness and endemism of the populations
- to clarify the status of endemism and vulnerability of some palaeoendemic taxa
- to focus attention on the need to implement appropriate conservation measures to protect rare and little known invertebrate taxa with relict or disjunct geographic distributions.

Reference

RELINI G. & GIACCONE G., 2009. Gli Habitat prioritari del Protocollo SPA/BIO (Convenzione di Barcellona) presenti in Italia. Schede descrittive per l'identificazione. *Biologia marina mediterranea*, **16** (1): 365 pp.

RELINI G. & TUNESI L., 2009. Le specie protette dal protocollo SPA/BIO (Convenzione di Barcellona) presenti in Italia. Schede descrittive per l'identificazione. *Biologia marina mediterranea*, 16 (2): 433 pp.

Abstract I

Porifera checklist and database of Mediterranean marine cave

Manconi R., **Cadeddu B.**, G.A. Stocchino, Pansini M., Pronzato R. & Ledda F.D., 2010

20th International Conference of Subterranean Biology, Postojna, Slovenia, 29 August- 3

September 2010

Poster presentation:

Porifera checklist and database of Mediterranean marine caves

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Conservation measures and protection planning of marine caves are promoted by the EU Habitat Directive. Porifera represent one of the main taxa in cave-dwelling sessile benthic assemblages. In this framework we report on a preliminary biodiversity inventory of sponges from Italian caves, based on the literature review. New data from recent faunistic surveys carried out in some submerged karstic caves of southern Italy (Sardinia and Sicily) are also reported. This contribution is the starting point for the creation of a Porifera database for Mediterranean marine caves. The work was supported by Italian MATTM and PRIN-MIUR, EU project Interreg III Sardinia-Corsica-Tuscany, Fondazione Banco di Sardegna and Regione Autonoma Sardegna.

Abstract II

Biodiversity inventory of Mediterranean marine caves:

Porifera checklist with new records

from Marine Protected Areas of Sardinia and Sicily

Ledda F.D., **Cadeddu B.**, Pansini M., Pronzato R. & Manconi R., 2010

8th World Sponge Conference, Girona, Spain, 20-24 September

Biodiversity inventory of Mediterranean marine caves: Porifera checklist with new records from Marine Protected Areas of Sardinia and Sicily

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Marine caves are fragile, cryptic and fragmented environments that require appropriate conservation measures and protection planning and recently have been inserted in the EU Habitat Directive. In this framework the taxonomic richness assessment, based on the literature, of the Mediterranean caves hosting sponges was performed. This review highlights that most records refer mainly to the Italian, French and Croatian karstic caves. Our new data focus on recent faunistical surveys carried out in four MPAs located in the karst systems of Sardinia and Sicily. Sponge communities have been compared among caves of the different geographic areas. This contribution enlarges the biodiversity inventory of marine caves, where sponge communities represent one of the main structuring components of sessile benthic assemblages, and is a first step for the creation of a Porifera database of the Mediterranean marine caves, currently in progress. Work supported by the Italian MATTM and PRIN-MIUR, the EU project Interreg III Sardinia-Corsica-Tuscany, the Fondazione Banco di Sardegna and Regione Autonoma Sardegna

Paper I

Biodiversity of Sardinian marine caves: sponge fauna

Manconi R., **Cadeddu B.**, Fozzi A., Pansini M., Pronzato R., Ledda F.D., 2011

42° Congresso della Società Italiana di Biologia Marina, Olbia 23-28 maggio 2011

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BIODIVERSITY OF SARDINIAN MARINE CAVES: SPONGE FAUNA

BIODIVERSITÀ DELLE GROTTA MARINE DELLA SARDEGNA: LA FAUNA A PORIFERI

Abstract – This paper focuses on a faunistic study on sponges from three submerged caves of the Marine Protected Area of Capo Caccia-Isola Piana. Results contribute to the assessment of biodiversity of the scarcely known Sardinian Sea.

Key-words: sponges, biodiversity, marine caves, endemism, Mediterranean Sea.

Introduction – The cave-dwelling sponge fauna of Sardinia is scarcely known although the few existing data suggest a notable taxonomic richness for the north-western karstic area of the island. This paper focuses on a faunistic study on W-Sardinian sponges of some submerged marine caves in a Marine Protected Area (MPA) aiming to increase data that can fill gaps in knowledge and to provide an inventory at species level.

Materials and methods – Three submerged caves of the Capo Caccia-Isola Piana MPA, namely Galatea Cave, Falco Cave and Bisbe Cave were surveyed in June 2009. The Galatea cave is located in the Zone A of the MPA, while the other two caves are situated in the Zone B.

Sponge assemblages were sampled within each cave, by scraping from the substratum and photographs, along five sectors identified in relation to the cave morphology. Specimens for sponge identification were photographed *in vivo*, sorted according to the sampling site, preserved (dry and/or ethanol) and registered in a reference collection. A first examination for macroscopic morphology was carried out on each sample by stereomicroscope. Specimens were dissected to obtain representative fragments to be processed for the preparation of skeleton and spicule slides by standard methods. Morphological analysis of diagnostic traits was carried out by optical microscopy with transmitted light on 141 preparations and by image analysis of 182 *in vivo* photographs.

Results – The faunistic census in the three submerged karstic caves revealed the presence of a remarkably diverse sponge assemblage. In total 79 species were censused. The Galatea Cave harbours 47 species belonging to 36 genera and 26 families ascribed to 12 orders. Twelve species are Mediterranean endemics with a value of endemism of 25.5 %. The Falco Cave harbours 37 species belonging to 28 genera and 22 families ascribed to 10 orders. Eleven species are Mediterranean endemics with a value of endemism of 29.7%. The Bisbe Cave harbours 35 species belonging to 26 genera and 19 families ascribed to 11 orders. Eleven species are Mediterranean endemics with a value of endemism of 31.4%.

The sponge fauna survey within the Sardinian marine caves highlighted the presence of four species listed as protected by the SPA/BIO protocol of the Barcelona Convention (Pronzato, 2003; Relini & Tunesi, 2009), namely *Petrobiona massiliana* Vacelet & Lévi, 1958, *Spongia lamella* (Schulze, 1879), *Spongia officinalis* Linnaeus, 1759 and *Spongia zimocca* Schmidt, 1862. In particular all of the four protected sponge species were recorded from the Bisbe Cave.

Conclusions – These data bridge, in part, the gap of knowledge on the sponge fauna of the Sardinian Sea, highlighted by Pansini & Longo (2003). Indeed Porifera from Sardinia have been reported until now, in surveys of cave-dwelling benthos (Bianchi & Morri, 1994) and in descriptions of new and rare species from these cryptic habitats (Manconi *et al.*, 2006, 2009; Manconi & Serusi, 2008). Marine caves are fragile and peculiar environments that have been recently listed in EU Habitat Directive. This work is a contribution to the assessment and inventory of biological diversity of Mediterranean marine caves, an instrument of primary relevance for the correct conservation and management of these habitats and their wildlife.

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Paper II

- Manuscript in preparation -

Taxonomic richness, endemism and rare species of Porifera in karstic
caves of three Marine Protected Areas of Sardinia and Sicily

Manconi R., Cadeddu B., Ledda F.D.

Taxonomic richness, endemism and rare species of Porifera in karstic caves of three Marine Protected Areas of Sardinia and Sicily

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Keywords. *Sponges, submerged caves, faunistic assessment, checklist, hot-spots of biodiversity.*

Abstract. This study deals with the marine sponges from 3 MPAs within karst systems of Sardinia and Sicily. Faunistic surveys were carried out in seven caves in the Sardinia Sea, Ionian Sea and Sicilian Channel focusing on taxonomic richness of cave-dwelling sponges. In total 79 species were found belonging to 47 genera, 33 families and 13 orders of Calcarea and Demospongiae. Comparative analyses of sponge assemblages among the 7 investigated caves indicate that although all three caves have a similar diversity of species, the species composition differs between each region: 4 were found in all caves, 24 were restricted to the Capo Caccia-Isola Piana MPA, 13 were found only in the caves of Plemmirio MPA, 4 were found in the Taccio Vecchio cave. These results show that a) sponge-dwelling marine caves assemblages represent a hot-spot of diversity in Mediterranean karstic areas at the local and regional scale and b) sponges play a key role as main structuring components of sessile benthic assemblages in these cryptic habitats.

Introduction

The status of marine caves as cryptic, fragmented and vulnerable biotopes require appropriate conservation measures and protection planning. These biotopes represent one of the most important natural heritages in the Mediterranean and recently they have been indeed inserted in the EU Habitat Directive (Relini & Giaccone, 2009). Submerged and semi-submerged Mediterranean marine caves host a rich and diversified biota harbouring several faunistic peculiarities among Porifera including bathyal forms (Harmelin, 1997), ‘relict’ species

(Vacelet & Lévi, 1958; Manconi et al., 2006, 2009; Manconi & Serusi, 2008; Pisera & Vacelet, 2010) stygobiont taxa (Lejeune & Chevaldonné 2005) and endemic taxa (Schmidt, 1864; Vacelet & Lévi, 1958; Schulze, 1880; Sarà, 1959, 1961; Schmidt, 1862; Pulitzer-Finali, 1978; Topsent, 1925; Schmidt, 1864; Topsent, 1892; Topsent, 1893; Schmidt, 1864; Pulitzer-Finali & Pronzato, 1980; Schulze, 1879;). Although several studies have addressed the biodiversity in marine caves of the Mediterranean Sea highlighting a high diversity of Porifera (Sarà, 1958, 1961, 1961; Russ & Rutzler, 1959; Labate, 1965; Rutzler, 1966; Boury-Esnault, 1971; Pouliquen, 1972; Pulitzer-Finali & Pronzato, 1976, 1980; Pansini et al., 1977; Pulitzer-Finali, 1977; 1983; Pansini & Pronzato, 1982; Balduzzi et al., 1989; Bibiloni, 1989; Corriero et al., 2000, 2004; Harmelin et al., 2003; Bussotti et al., 2006; Tournon et al., 2009) several karstic areas have been neglected.

To increase basic information on the marine speleofauna of these threatened habitats we recently carried out a faunistic census focusing on the cave-dwelling sponges in some submerged caves of the Sardinian Sea, Ionian Sea and Sicilian Channel in the framework of biodiversity assessment of the Italian Marine Protected areas (MPAs). We report on the taxonomic richness, degree of endemism and presence of rare taxa of Porifera in seven caves.

Materials & Methods

Study areas

The study was carried out within 3 different karstic systems of the Western-Central Mediterranean Sea with different geological and biogeographical histories and characterized by the presence of several submerged and semisubmerged cavities and caves.

Faunistic censuses were performed within 3 caves from the Capo Caccia-Isola Piana Marine Protected Area (Sardinian Sea, North-Western Sardinia), 3 caves from the Plemmirio MPA (Ionian Sea, Eastern Sicily) and 1 caves from the Pelagie MPA (Sicilian Channel, Lampedusa) (Fig. 1).



Fig. 1: Study area in the Mediterranean Sea. Maps of the three Marine Protected Areas with location of the seven investigated caves. **a.** Capo Caccia-Isola Piana MPA (1: Galatea Cave, 2: Falco Cave, 3: Bisbe Cave); **b.** Isole Pelagie MPA (4: Taccio Vecchio Cave); **c.** Plemmirio MPA (5: Mazze Cave, 6: Gymnasium Cave, 7: Gamberi Cave).

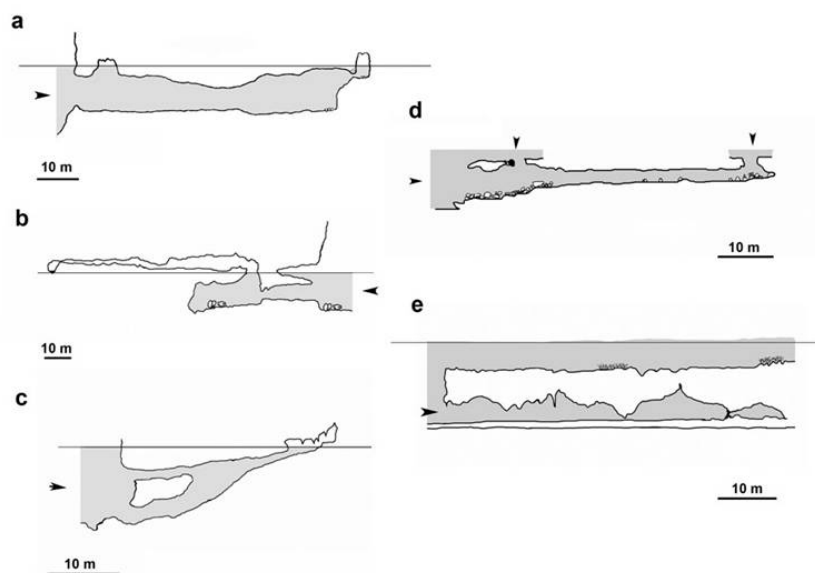


Fig. 2: Profile of some investigated caves: **a.** Galatea Cave; **b.** Falco Cave; **c.** Bisbe Cave; **d.** Taccio Vecchio Cave; **e.** Gymnasium Cave.

Cave	Coordinates	Depth m	Typology (water level, horizontal tunnel, large chambers etc
Galatea	40°34'09" N 8°13'54" E	4 - 9	The Galatea cave is a tunnel shaped long cavern. The entrance with south western exposure is large, the bottom is mostly sandy and after the cave shrank in a narrow rocky passage that divide the cave into two totally dark chambers.
Falco	40°34'09" N 8°13'14" E	5 – 12	The Falco cave is a wide cavern with two entrances exposed to the South. It develops for and the bottom is rocky all along the cave. In the inner part the bottom, in a zone of fresh and saltwater mixing, raise up until an aerial tunnel.
Bisbe	40°34'15" N 8°12'55" E	3 – 10	The Bisbe cave is a wide entrance exposed to the south. The development is of and the bottom is sandy near the entrance while rocky in the inner part. The bottom raise at the end of the cavern and open in a chamber that emerge in a aerial bubble.
Gymnasium	37°00'12" N 15°18'48" E	20	Gymnasium cave has a wide opening and develop like a straight tunnel for in SW – NE direction. The bottom is mainly muddy except in the inner part of the cave where some stalagmites arose from the bottom.
Mazzere	37°00'18" N 15°18'35" E	25	Mazzere cave develop along a SW – NE direction. Is characterized by two branches that divide from the main chamber at the entrance. The left branch go straight for and shrank at the end. The right branch has a narrow initial passage that opens in a wide dark chamber, (long, high). The bottom is characterised by very fine mud.
Gamberi		20	Gamberi cave is so called for the constant presence of the narwal shrimp (<i>Plesionika narval</i>) in the inner part of the cave. Is characterised by a high () and narrow () entrance and develop as a tunnel for .
Taccio Vecchio I	35°31'29" N 12°35'58" E	20–10	Is a long tunnel opening at both extremities. The entrance at depth opens in a wide chamber connecting after at a narrow tunnel that opens at the end in a hole at depth that is the result of the slide of the roof.

The morphology of the selected caves is not homogeneous and enroll different types of caves such as tunnel-like, divided in chambers, with or not the aerial chamber (Fig. 2).

The karstic area of Capo Caccia is well known for hosting more than 200 (estimated) karstic caves of which only 47 are mapped notwithstanding the high interest of speleodivers in this area during the last 50 years (Chessa et al., 1999).

Three caves were selected within the calcareous Punta Giglio promontory, a 80 meters high cliff that decline abruptly in the sea till 30 m depth in front of the coast.

The Plemmirio MPA is localized within the Maddalena Peninsula that is a horst of Pleistocenic formation. The coastline is of calcarenite origin and declines slowly in the sea through shelves alternate by slopes. Data about the morphology and biodiversity of these caves are very scarce (Pitruzzello & Russo, 2008;).

Lampedusa is the biggest island the Pelagie Archipelago; it belongs to the African plateau and is characterized by a series of calcareous beds and promontories of Tortonian and Messinian origin. The north coast is a cliff, 50 to 130 m high, while the South-Eastern side slopes gently into the sea. The monitored cave is located on the Northern side.

Sampling and identification protocols

The 7 caves of the three Marine Protected Areas were surveyed between June and October 2009 by SCUBA diving. For each cave two dives were performed. Each cave was sampled along five sectors identified in relation to the cave morphology. Specimen of sponges from each sector were collected by scraping from the substratum and photographed and subsequently fixed in ethanol or preserved dry on the collected substratum.

For taxonomic identification sponges were dissected under a stereo microscope to observe macro-traits and to obtain representative fragments of the skeleton. Spicules were processed by dissolution of organic matter in boiling 65% nitric acid for demosponges and hypochlorite for calcisponges washed in water, suspended in alcohol and dropped onto slides for Light Microscopy (LM).

Growth form, architecture of the ectosomal and choanosomal skeleton, spicular morphotraits and dimensions, together the topographic localization of spicular types have been considered as diagnostic characters for identification at the species level.

The taxonomic status of cited sponge taxa was validated on the base of the World Porifera Database (van Soest et al., 2011; www.marinespecies.org/porifera/).

As for the concept of endemism, taxa are considered endemic when their geographic range is reported in the literature as restricted to the Mediterranean Sea (Pansini & Longo, 2003,

2008; Voultziadou, 2005). Endemicity values (%) of each cave were estimated on the basis of the number of Mediterranean endemics recorded respect to the total number of species in each cave.

Sampled sponge specimens were registered in an archive, with respective histological slides for light microscopy, to prepare a voucher collection.

Statistical analysis

The checklist produced by the survey of the Sardinian and Sicilian caves was compared with data that show the complete list of Porifera species found in other karstic caves of the Mediterranean Sea, such as the Mitigliano Cave of the Peninsula Sorrentine (Pansini & Pronzato, 1982), the Bue Marino Cave in Tremiti Islands (Corriero et al., 2000), some caves of the Medes Islands (Bibiloni et al., 1984) and three caves in the Gulf of Marseilles (Pouliquen, 1972). Statistical analysis were performed by the software Primer6.

The similarity in sponge composition between samples (caves) was measured with the Bray–Curtis similarity coefficients.

Results

In total 79 sponge species were censused from the 7 investigated karstic submerged caves and a synthesis of data are reported in a checklist (**Appendix**).

The Galatea Cave (Fig. 1-2) harbours 47 species belonging to 36 genera and 26 families ascribed in 12 orders (**Appendix**). Fourteen species are Mediterranean endemics with a value of endemicity of 29.7% (Tab. 1).

The Falco Cave (Fig. 1-2) harbours 37 species belonging to 28 genera and 22 families ascribed in 10 orders (**Appendix**). Twelve species are Mediterranean endemics with a value of endemicity of 32.4% (Tab. 1) .

The Bisbe Cave (Fig. 1-2) harbours 35 species belonging to 26 genera and 19 families ascribed in 10 orders (**Appendix**). Twelve species are Mediterranean endemics with a value of endemicity of 34.2% (Tab. 1).

The Gymnasium Cave (Fig. 1-2) harbours 23 species belonging to 17 genera and 15 families ascribed in 10 orders (**Appendix**). Six species are Mediterranean endemics with a value of endemicity of 26% (Tab. 1).

The Mazzere Cave harbours 26 specie belonging to 21 genera and 17 families ascribed in 12 orders (**Appendix**). Nine species are Mediterranean endemics with a value of endemicity of 34,6 % (Tab. 1).

The Gamberi Cave harbours 22 species belonging to 16 genera and 11 families ascribed in 7 orders (**Appendix**). Five species are Mediterranean endemics with a value of endemicity of 22.7 % (Tab. 1).

The Taccio Vecchio Cave I harbours 33 species belonging to 22 genera and 17 families ascribed in 12 orders (**Appendix**). Eight species are Mediterranean endemics with a value of endemicity of 24 (Tab. 1).

Cave	Species (n)	Endemic species (%)
Galatea	47	29.7
Bisbe	35	34.2
Falco	37	32.4
Mazzere	26	34.6
Gamberi	22	22.7
Gymnasium	23	26
TaccioVecchio	33	24

Tab. 1: Endemicity values of Porifera at the species level as % of Mediterranean endemics recorded in the seven investigated caves.

In the Capo Caccia-Isola Piana MPA (Sardinian Sea) (Fig. 3) the taxonomic richness of sponge fauna is represented by 58 species belonging to 38 genera and 29 families ascribed in 13 orders and 2 classes. Within the Plemmirio MPA (Fig. 3) 44 species were recorded, belonging to 27 genera and 22 families ascribed in 12 orders and 2 classes.

Within the Pelagie MPA (Fig. 3), represented here by a single cave, 33 species belonging to 24 genera and 19 families ascribed in 12 orders and 2 classes were recorded.

Two distinct groups of species are shown by cluster analysis with each having an average similarity higher than 55% (Fig. 4). The first group includes the Alghero caves and the

Marsiglia caves, the second group includes the Maddalena Peninsula caves and the Taccio Vecchio cave.

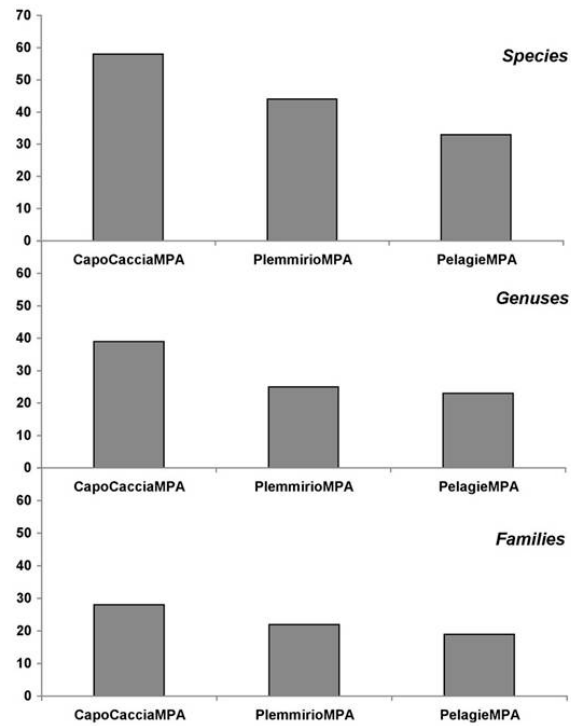


Fig. 3: Taxonomic richness of Porifera at the species, genus and family levels in the three MPAs.

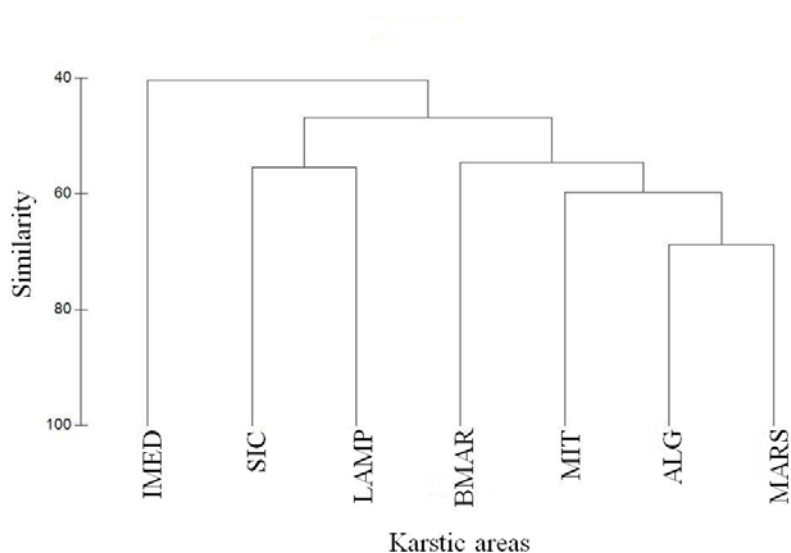


Fig. 4: Cluster analysis among the 3 MPAs and the other karstic areas in the Mediterranean Sea. IMED: Islands Medes caves; SIC: Plemmirio caves; LAMP: Taccio Vecchio cave; BMAR: Bue Marino cave; MIT: Mitigliano cave; ALG: Alghero caves; MARS: Marsiglia caves

Discussion

Taxonomic richness

The investigated karstic areas host a relatively rich sponge fauna. The caves from the three MPAs were characterized by a relatively diverse composition, indeed the faunistic census highlighted species richness values for each cave ranging from 22 to 47 with the highest value in the cave Galatea. Similar species richness values were previously reported from other Mediterranean caves (Sarà, 1961, 1961; Pouliquen, 1972; Pansini & Pronzato, 1982; Balduzzi et al., 1989; Cinelli et al., 1997; Corriero et al., 2000; Corriero et al., 2004; Bibiloni et al., 1989).

Notwithstanding the low sampling effort (1-2 divers of 2 divers per cave in a single season) applied within this faunistic survey, we could record a notably high number of taxa if compared to previous detailed and invasive investigations of marine caves from Marseille, Medes Islands, Sorrento Paeninsula and Tremiti Islands (Pouliquen, 1972; Bibiloni *et al.*, 1984; Pansini & Pronzato, 1982; Corriero *et al.*, 2000).

Interestingly three of the sponge species records from our surveyes, *Timea geministellata* in the Gymnasium cave, *Ircinia retidermata* in the Falco cave, and *Spongia zimocca* in the

Bisbe cave, represent the first discovery in marine caves of these species, previously known only by few works on other rocky systems (Pansini & Longo, 2003, 2008; Voultsiadou, 2005, Coll et al., 2010).

Species richness confirms that caves are an extremely suitable biotope for sponge assemblages probably because these primitive invertebrates are able to adopt successful preadaptive strategies.

The surveys of the 3 MPAs caves give a contribution to the increase of sponge biodiversity knowledge in marine caves. These new data fill in part the scarce information on the diversity of cave-dwelling sponges from Sardinian Sea till now limited to few works (Manconi et al., 2006; Manconi & Serusi, 2008; Manconi et al., 2009), greatly increasing the records of this taxon from this neglected sector of the western Mediterranean, as stressed by previous synopsis (Pansini & Longo, 2003). Moreover the faunistic census in the Taccio Vecchio Cave (Lampedusa Island) increases the existing data of sponge diversity in the Sicily Channel; a comparative analysis performed with the checklist by Pansini e Longo (2003) highlights 9 new species records for this area. The Plemmirio MPA surveys resulted in 5 new sponge species never recorded before from the Ionian sea. Results highlighted that species such *Erylus euastrum*, *Phorbas tenacior*, *Agelas oroides*, *Oscarella lobularis*, *Diplastrella bistellata*, *Terpios fugax*, *Crambe crambe*, *Acanthella acuta*, *Petrosia ficiformis*, *Ircinia variabilis* are the among the commonest species in caves as their well known stygophilic behaviour suggest.

The distribution of *Timea geministellata* until now was known exclusively from the Punta San Lorenzo towards Punta Lagno (Massa Lubrense); the new record from Gymnasium cave enlarges the geographic range of this extremely rare Mediterranean endemism (see also Voultsiadou & Vafidis, 2004). The discovery of *Spongia (Spongia) zimocca*, *Ircinia retidermata* and *Timea geministellata* never recorded before in Mediterranean caves suggests that the biodiversity of these marine caves is underestimated. This status is supported also by the recent description of new species from both the eastern and western Mediterranean basins (Manconi et al. , 2006; Manconi & Serusi, 2008; Perez et al 2004; Vacelet et al 2007).

Biogeographic analysis

The composition of Mediterranean cave-dwelling fauna is characterised by endemic

species and species with a geographic range restricted to Mediterranean and Atlantic-Macaronesian area (van Soest, 1994).

Of 79 identified species, 19 are Mediterranean endemics and some of them were originally described from caves (e.g. *Dercitus (Stoeba) dissimilis*) or were previously recorded almost exclusively from caves (*Petrobiona massiliana*).

Of the whole recorded species from the three MPAs, 26 species are Atlanto-Mediterranean and 8 species are considered cosmopolitan, but this geographic pattern is clearly due to the absence of detailed morphological analyses focusing on key diagnostic traits notwithstanding the extremely conservative and notably low number of target morphological traits of Porifera at the species level. This is the case of the revision of the Plakinidae by Muricy et al., 1998 and of *Oscarella* by Muricy et al., 1996 from Mediterranean caves.

The condition of cosmopolitanism is particularly critical to be sustained for taxa living in an extremely enclosed sea like the Mediterranean where evolutionary forces such as a rhythmic geographic isolation, reproductive isolation bottle-neck.

Analyzing orders, families and genera of sponges recorded from the 7 surveyed caves emerged that orders are mostly of cosmopolitan distribution, except order Agelasida which presents a circum-tropical distribution (van Soest, 1994). Of the 33 families recorded, 26 have a cosmopolitan distribution; two families, Agelasidae and Spongiidae, have a circum-tropical distribution; two families, Timeidae and Rhabderemiidae, have a wide-spread distribution, found in temperate waters and in tropical waters (van Soest, 1994); the monospecific and monogeneric family Petrobionidae is a paleoendemism of the Mediterranean Sea (Manconi et al., 2009).

Of 47 genera recorded, 8 genera have a cosmopolitan distribution; 17 genera have a circum-tropical distribution; 13 genera have a wide-spread distribution and three genera, (*Prosuberites*, *Crella* and *Hymedesmia*) have a typical cold water distribution (van Soest, 1994). Finally, the genus *Hemimycale* is known to have an Atlantic-Mediterranean distribution that reaches the Arctic Sea (Soest et al., 2011; www.marinespecies.org/porifera)

Comparative analysis among the three MPAs

The taxonomic composition of sponge assemblages is very similar in the 3 caves of Punta Giglio. The Falco and Bisbe caves share most species probably because of the very similar

cave typology. On the other hand the Galatea cave that diverges in morphology if compared to the other caves from Capo Caccia MPA, displays a higher taxonomic richness and is characterized by relatively rare sponges belonging to a few species (*i.e. Erylus euastrum* and *Erylus discophorus*). Within the monitored Plemmirio caves no significant difference has been observed in species abundance and composition. Although the effort of sampling in the Taccio Vecchio cave has been very limited, a high number of species have been recorded.

The number of species shared by all investigated caves is very low with only 4 ubiquitous species, namely *Petrobiona massiliana*, *Erylus euastrum*, *Spirastrella cunctatrix*, *Phorbas tenacior*.

This taxonomic heterogeneity seems to be related to the extremely various environmental conditions characterizing also strictly neighbouring caves.

The taxonomic richness analysis highlighted a notably diverging composition between the Ionian Sea, the Sicilian Channel and the Sardinian Sea.

Comparative analyses of sponge assemblages among the 7 investigated caves indicates that although all three localities have a similar diversity of species (22 to 47 species each), the species composition differs between each region: 4 were found in all caves, 25 were restricted to the Punta Giglio Promontory, 10 were found only to the Plemmirio MPA and 4 were found in the Lampedusa.

The caves of Alghero have in common with the caves of Plemmirio, 26 species, 20 genera and 15 orders, while by the Taccio Vecchio cave have in common, 24 species, 19 genera and 16 orders. The caves of Plemmirio have in common with the Taccio Vecchio cave, 23 species, 17 genera and 15 orders.

The caves of Alghero have a richer composition in horny sponges compared to the caves of the other two MPAs. This data may be related with the fact that the Sardinian caves have a structure to large halls (Bisbe and Falco), and then a light intensity entering even the inner zones. While the Sicilian caves of Plemmirio and Lampedusa, are primarily a tunnel structure that relegates the light intensity in the first few meters of the opening cave.

Comparative analysis of the three MPAs against other Mediterranean karstic areas

The comparative analysis showed that the three Alghero caves in the have, in species composition, a greater affinity with the Marseille caves (32 species in common), while the

three Plemmirio caves and the Taccio Vecchio cave have, in species composition, a greater affinity with the Mitigliano cave.

By a biogeographical point of view the similarities between the caves of North-West Sardinia and the caves of Marseille is credible, having regard to the disposition of karstic areas analyzed in the Mediterranean Sea: the Sardinia and the Gulf of Marseilles is located in the western part of the Mediterranean basin.

The affinity between the caves of the Maddalena Peninsula and the Taccio Vecchio cave, with the Mitigliano cave is unclear due to the disposition of these karstic areas in the Mediterranean. Also from a biogeographical point of view, the caves of the Alghero area, would have presented an affinity with the caves of the Medes Islands, given the location of these in the Mediterranean Sea. These dissimilarities may be related to the fact that karstic areas compared, have a different number of species surveyed. Furthermore, consider the different sampling methods used, and the different number of samples for each cave.

The cluster analysis based on the Bray Curtis coefficient with data from 7 caves confirm the result obtained by analyzing the species composition of different areas.

Conclusions

Our new data fill, in part, the gap of knowledge on cave diversity of the Sardinian Sea, the Ionian sea and the Sicilian Channel.

Differences in taxonomic composition seems to be related to both the habitat heterogeneity among caves and the different geological and climatic history of the considered marine sub-basins.

Concerning the analysis of faunal composition, there is a diversity at the species level between the sponge populations to the three MPAs caves considered and other karstic caves of Italian and Mediterranean areas.

The data refer to a small number of measurements performed, only 2 dives for each cave.

This suggests that further sampling may show the presence of a higher number of species

In biogeographical terms we can assume that the differences between the caves of the north-west Sardinia and the caves of Apulia or the Sorrentine peninsula, can be related both to the different geological origin of karstic areas compared to the different evolution of the Sardinia Sea, the Tyrrhenian and southern Adriatic Sea. In fact, the Sardinia

geological origin has much older and remarkably different from the karstic areas of Apulia and Campania.

This hypothesis is confirmed by the similarity between the sponge composition of the North-Western Sardinia caves and the Marseille caves in the Gulf of Lions.

Moreover, the similarities between the sponge composition of the Plemmirio caves and the Lampedusa cave, may be related to the geological origin of the common African Plate.

Conservation

The location of the Pelagie archipelago in the Sicilian Channel makes important faunal data collected, because this area is crucial in biogeographical terms, and understanding the processes of colonization / extinction of fauna in the Mediterranean during its geological history and the relations between the eastern and western basins. Investigated caves matches the category of 'Semi-dark Caves' (Habitats code IV.3.2) and 'dark Caves' (Habitat code V.3.2) (Relini e Giaccone, eds, 2009).

Among the Porifera surveyed, 4 species surveyed are considered in Annexes II and III of the Barcelona Convention as 'Protected species of Protocol SPA / BIO' (vedi Relini e Tunesi, eds, 2009):

Petrobiona massiliana (Vacelet & Lévi, 1958) code species 2566

Spongia (Spongia) lamella (Schulze, 1879) species code 3006.

Spongia (Spongia) officinalis Linnaeus, 1759 species code 3007.

Spongia (Spongia) zimocca Schmidt, 1862, species code 3008.

Overall, the presence of i) high of biological diversity values, ii) a paleoendemic species, rare and protected, and iii) habitats whose conservation is a priority, increase the value of the three MPAs.

These caves also have a high natural value, are extremely rare in the Mediterranean, have high aesthetic - landscape value and substantial economic value as they are in the 'tourist caves' that attract many visitors.

Aknowledgements

This paper is dedicated to the memory of Gaetano Ferruzza and Dario Romano, who tragically passed away on 6 October 2009 in a scuba-diving accident while working in a

marine cave in the Plemmirio MPA, Sicily. The expert speleosub Riccardo Leonardi and Giampiero Mulas of the Società Italiana di Speleologia, are kindly acknowledged for the assistance during cave-diving to R. Manconi and F. Ledda. Research supported by the Italian Ministero dell'Ambiente MATTM ('Studio degli ambienti di grotte marine sommerse (Codice Habitat 8330) nelle Aree Marine Protette di Pelagie, Plemmirio e Capo Caccia'), Ministero dell'Università e della Ricerca Scientifica e Tecnologica (MIUR-PRIN 20085YJMTC 'L'endemismo nella fauna italiana: dalla conoscenza sistematica e biogeografica alla conservazione'), European Project INTERREG Sardinia-Corsica-Tuscany Biodiversity, and Fondazione Banco di Sardegna. F.D. Ledda was supported by a postdoctoral research grant of the Regione Autonoma della Sardegna; B. Cadeddu was supported by a Regione Autonoma della Sardegna.

Appendix

Taxonomic list and distribution of the sponges in 7 Mediterranean caves: GAL: Galatea cave; BIS: Bisbe cave;

FAL: Falco cave; MAZ: Mazzere cave; GAM: Gameri cave; GYM: Gymnasium cave; TVI: Taccio Vecchio

cave.

	GAL	BIS	FAL	MAZ	GAM	GYM	TV
Porifera Grant, 1836							
CALCAREA Bowerbank, 1864							
Calcinea Bidder, 1898							
Clathrinida Hartman, 1958							
Clathrinidae Minchin, 1900							
Clathrina Gray, 1867							
Clathrina rubra (Haeckel, 1872)							+
Clathrina clathrus (Schmidt, 1864)	+	+	+				+
Clathrina contorta Minchin, 1905	+						
Calcaronea Bidder, 1898							
Leucosolenida Hartman, 1958							
Sycettidae Dendy, 1892							
Sycon Risso, 1826							
Sycon elegans (Bowerbank, 1845)				+			
Sycon sp.		+					
Grantiidae Dendy, 1893							
Leucandra Haeckel, 1872							
Leucandra aspera (Schmidt, 1862)						+	
Lithonida Vacelet, 1981							
Petrobionidae Borojevic, 1979							
Petrobiona Vacelet & Lévi, 1958							
Petrobiona massiliiana Vacelet & Lévi, 1958	+	+	+	+	+	+	+
DEMOSPONGIAE Sollas, 1885							
Homoscleromorpha Bergquist, 1978							
Homosclerophorida Dendy, 1905							
Oscarellidae Lendenfeld, 1887							
Oscarella Vosmaer, 1884							
Oscarella lobularis (Schmidt, 1862)	+	+	+	+		+	
Oscarella sp.					+		+
Plakinidae Schulze, 1880							
Corticium Schmidt, 1862							
Corticium candelabrum Schmidt, 1862	+			+	+	+	
Plakina Schulze, 1880							
Plakina trilopha					+		
Plakina sp.				+		+	+
Tetractinomorpha Lévi, 1953							
Astrophorida Sollas, 1888							
Ancorinidae Schmidt, 1870							
Stelletta Schmidt, 1862							
Stelletta lactea Carter, 1871	+		+				
Jaspis Gray, 1867							
Jaspis johnstoni (Schmidt, 1862)	+						
Geodiidae Gray, 1867							
Erylus Gray, 1867							
Erylus discophorus (Schmidt, 1862)	+						
Erylus euastrum (Schmidt, 1868)	+	+	+	+	+	+	+
Pachastrellidae Carter, 1875							
Dercitus Gray, 1867							
Dercitus (Stoeba) Sollas, 1888							
Dercitus (Stoeba) dissimilis (Sarà, 1959)	+						
Hadromerida Topsent, 1894							
Thoosidae (Alectonidae) Rosell, 1996							
Alectona Carter, 1879							
Alectona sp.	+		+				
Clionidae D'Orbigny, 1851							
Cliona Grant, 1826							
Cliona schmidti (Ridley, 1881)	+						
Cliona viridis Schmidt, 1862	+	+	+				
Cliona sp. 1		+			+		
Cliona sp. 2	+						

	GAL	BIS	FAL	MAZ	GAM	GYM	TV
Spirastrellidae Ridley & Dendy, 1886							
<i>Diplastrella</i> Topsent, 1918							
<i>Diplastrella bistellata</i> (Schmidt, 1862)	+	+	+	+	+		+
<i>Diplastrella</i> sp.						+	
<i>Spirastrella</i> Schmidt, 1868							
<i>Spirastrella cunctatrix</i> Schmidt, 1868	+	+	+	+	+	+	+
Suberitidae Schmidt, 1870							
<i>Aptos</i> Gray, 1867							
<i>Aptos aptos</i> (Schmidt, 1864)	+	+	+				
<i>Prosuberites</i> Topsent, 1893							
<i>Prosuberites</i> sp.		+					
<i>Terpios</i> Duchassaing & Michelotti, 1864							
<i>Terpios fugax</i> (Duchassaing & Michelotti, 1864)	+	+	+		+	+	+
Timeidae Topsent, 1928							
<i>Timea</i> Gray, 1867							
<i>Timea geministellata</i> Pulitzer-Finali, 1978						+	
Chondrosida Boury-Esnault & Lopez, 1985							
Chondrillidae Gray, 1872							
<i>Chondrilla</i> Schmidt, 1862							
<i>Chondrilla nucula</i> Schmidt, 1862				+		+	
<i>Chondrosia</i> Nardo, 1847							
<i>Chondrosia reniformis</i> Nardo, 1847	+						+
Ceractinomorpha Lévi, 1953							
Poecilosclerida Topsent, 1928							
Microcionina Hajdu, Van Soest & Hooper, 1994							
Raspailiidae Hentschel, 1923							
Raspailiinae Nardo, 1833							
<i>Eurypon</i> Gray, 1867							
<i>Eurypon major</i> Sarà & Siribelli, 1960				+			
Rhabderemiidae Topsent, 1928							
<i>Rhabderemia</i> Topsent, 1890							
<i>Rhabderemia minutula</i> (Carter, 1876)	+		+				
Myxillina Hajdu, Van Soest & Hooper, 1994							
Chondropsidae Carter, 1886							
<i>Batzella</i> Topsent, 1893							
<i>Batzella inops</i> (Topsent, 1891)	+						
Crambeidae Lévi, 1963 1							
<i>Crambe</i> Vosmaer, 1880							
<i>Crambe crambe</i> (Schmidt, 1862)	+	+	+	+			+
Crellidae Dendy, 1922							
<i>Crella</i> Gray, 1867							
<i>Crella (Crella) elegans</i>							+
<i>Crella</i> sp.	+	+	+				+
Hymedesmiidae Topsent, 1928							
<i>Hemimycale</i> Burton, 1934							
<i>Hemimycale columella</i> (Bowerbank, 1874)	+		+				
<i>Hymedesmia</i> Bowerbank, 1864							
<i>Hymedesmia (Hymedesmia)</i> Bowerbank, 1864							
<i>Hymedesmia (Hymedesmia) baculifera</i>					+		
<i>Hymedesmia (Hymedesmia) paupertas</i> (Bowerbank, 1866)				+			
<i>Hymedesmia (Hymedesmia)</i> sp.	+	+		+		+	+
<i>Phorbas</i> Duchassaing & Michelotti, 1864							
<i>Phorbas dives</i> (Topsent, 1891)			+				+
<i>Phorbas fictitius</i> Bowerbank, 1866	+				+		+
<i>Phorbas tenacior</i> (Topsent, 1925)	+	+	+	+	+	+	+
<i>Phorbas</i> sp.						+	+
Halichondrida Gray, 1867							
Axinellidae Carter, 1875							
<i>Axinella</i> Schmidt, 1862							
<i>Axinella damicornis</i> (Esper, 1794)	+	+	+	+		+	+
<i>Axinella verrucosa</i> (Esper, 1794)	+				+		
Heteroxydidae Dendy, 1905							
<i>Didiscus</i> Dendy, 1922							
<i>Didiscus</i> sp.					+		

	GAL	BIS	FAL	MAZ	GAM	GYM	TV
Dictyonellidae Van Soest, Diaz & Pomponi, 1990							
<i>Acanthella</i> Schmidt, 1862							
<i>Acanthella acuta</i> Schmidt, 1862	+	+	+	+		+	
<i>Dictyonella</i> Schmidt, 1868							
<i>Dictyonella</i> sp.	+	+	+				+
Halichondriidae Gray, 1867							
<i>Halichondria</i> Fleming, 1828							
<i>Halichondria (Halichondria)</i> Fleming, 1828							
<i>Halichondria (Halichondria) genitrix</i> Schmidt, 1870			+				
Agelasida Hartman, 1980							
Agelasidae Verrill, 1907							
<i>Agelas</i> Duchassaing & Michelotti, 1864							
<i>Agelas oroides</i> (Schmidt, 1864)	+	+	+	+		+	+
Haplosclerida Topsent, 1928							
Haplosclerina Topsent, 1928							
Chalinidae Gray, 1867							
<i>Dendroxea</i> Griessinger, 1971							
<i>Dendroxea lenis</i> (Topsent, 1892)	+	+	+				
<i>Haliclona</i> Grant, 1836							
<i>Haliclona (Halicholona)</i> de Laubenfels, 1932							
<i>Haliclona (Halichocona) fulva</i> (Topsent, 1893)		+		+			
<i>Haliclona (Haliclona)</i> Grant, 1836							
<i>Haliclona (Reniera)</i> Schmidt, 1862							
<i>Haliclona (Reniera) cinerea</i> (Grant 1826)					+	+	
<i>Haliclona (Reniera) cratera</i> (Schmidt, 1862)			+	+			
<i>Haliclona (Soestella)</i> De Weerd, 2000							
<i>Haliclona (Soestella) mucosa</i> (Griessinger, 1971)	+	+	+		+		+
<i>Haliclona</i> sp.	+	+	+	+		+	+
Petrosina Boury-Esnault & Van Bevern, 1982							
Phloeodictyidae Carter, 1882							
<i>Oceanapia</i> Norman, 1869							
<i>Oceanapia</i> sp.							+
Petrosiidae Van Soest, 1980							
<i>Petrosia</i> Vosmaer, 1885							
<i>Petrosia (Petrosia)</i> Vosmaer, 1885							
<i>Petrosia (Petrosia) ficiformis</i> (Poiret, 1789)	+	+	+	+	+		
<i>Petrosia (Petrosia)</i> sp.						+	+
Dictyoceratida Minchin, 1900							
Irciniidae Gray, 1867 2							
<i>Ircinia</i> Nardo, 1833							
<i>Ircinia dendroides</i> (Schmidt, 1862)				+	+	+	+
<i>Ircinia oros</i> (Schmidt, 1864)	+	+	+	+			
<i>Ircinia retidermata</i> Pulitzer-Finali & Pronzato, 1980			+				
<i>Ircinia variabilis</i> (Schmidt, 1862)	+	+	+	+		+	+
<i>Ircinia</i> sp.					+		
<i>Sarcotragus</i> Schmidt, 1862							
<i>Sarcotragus foetidus</i> (Schmidt, 1862)							+
Thorectidae Bergquist, 1978							
Thorectinae Bergquist, 1978							
<i>Scalariispongia</i> Cook & Bergquist, 2000							
<i>Scalariispongia proficiens</i> (Pulitzer-Finali & Pronzato, 1980)	+						
<i>Fasciospongia</i> Burton, 1934							
<i>Fasciospongia cavernosa</i> (Schmidt, 1862)	+						
Spongiidae Gray, 1867							
<i>Spongia</i> Linnaeus, 1759							
<i>Spongia (Spongia)</i> Linnaeus, 1759							
<i>Spongia (Spongia) lamella</i> (Schulze, 1879)	+	+	+				
<i>Spongia (Spongia) nitens</i> (Schmidt, 1862)		+	+				
<i>Spongia (Spongia) officinalis</i> Linnaeus, 1759		+	+				
<i>Spongia (Spongia) virgultosa</i> (Schmidt, 1868)	+	+	+				
<i>Spongia (Spongia) zimocca</i> Schmidt, 1862		+					

	GAL	BIS	FAL	MAZ	GAM	GYM	TV
Dysideidae Gray, 1867							
<i>Dysidea</i> Johnston, 1842							
<i>Dysidea avara</i> (Schmidt, 1862)	+	+	+				+
<i>Dysidea fragilis</i> (Montagu, 1818)	+	+	+		+		+
<i>Dysidea incrustans</i> (Schmidt, 1862)	+				+	+	+
<i>Pteraplysilla</i> Topsent, 1905							
<i>Pteraplysilla spinifera</i> (Schulze, 1879)	+	+	+	+	+		+
Dendroceratida Minchin, 1900							
Darwinellidae Merejkowsky, 1879							
<i>Aplysilla</i> Schulze, 1878							
<i>Aplysilla rosea</i> (Barrois, 1876)	+			+			+

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Paper III

- Manuscript in preparation -

Biodiversity assessment and biogeographic traits
of cave-dwelling Porifera (Mediterranean Sea)

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Biodiversity assessment and biogeographic traits of cave-dwelling Porifera (Mediterranean Sea)

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Abstract. The aim of this paper is to assess the taxonomic richness of sponge fauna in Mediterranean marine caves, thus contributing to knowledge of biodiversity in these fragile and vulnerable biotopes. We provide here a checklist as a key tool for the faunistic and the biodiversity management and conservation at the local, regional and global scale. To outline the taxonomic richness and biogeographic patterns of Mediterranean sponge fauna we performed an overview of historical data on sponges from semisubmerged and submerged caves. Additional data have been included after the analysis of the samples from 3 caves of the Capo Caccia-Isola Piana MPA, 3 caves of the Plemmirio MPA, and one cave of the Pelagie MPA.

Introduction

The Mediterranean Sea is a biodiversity hot spot (Coll *et al.*, 2010) harbouring 17, 000 marine species, although the human impact exerted since ancient times by environmental degradation, habitat loss, pollution, and eutrophication are affecting the persistence and survival of a huge number of taxa, together with present climate change and alien species invasions. Despite the interest in natural history investigations dated up to some thousands of years and the key role played by some pioneers marine biology stations (e.g. Stazione Zoologica di Napoli, Banyuls) the assessment of marine taxonomic richness is still incomplete (Pansini & Longo, 2003, 2008; Voultziadou, 2005; Coll *et al.*, 2010). This is particularly true for not easily accessible biotopes such as submerged caves and deep

water where Porifera, mostly skiophilous, are among the dominant taxa in these dark environments.

The aim of this paper is to assess the taxonomic richness of sponge fauna in Mediterranean marine caves, thus contributing to knowledge of biodiversity in these fragile and vulnerable biotopes protected by the Barcelona Convention (Relini e Giaccone, eds, 2009). We provide here a checklist as a key tool for the faunistic and the biodiversity management and conservation at the local, regional and global scale. Checklists are easily accessible instruments for the most different consumers allowing both a continuous updating of knowledge giving detailed information, and the basis for the numerical analyses. We attempt also to test cave-dwelling sponge assemblage to track at the regional level the biogeographical history of Mediterranean fauna. Sponges are key taxa in marine biogeographic analyses for their sessile life style in the adult phase of the life cycle and show low dispersal power propagules i.e. lecithotrophic planktonic larvae (Sarà, 1959, 1961; Pansini & Longo, 2008; Manconi et al., 2009; Manconi & Serusi, 2008).

Materials & Methods

To outline the taxonomic richness and biogeographic patterns of Mediterranean sponge fauna we performed an overview of historical data on sponges from semisubmerged and submerged caves. Our up to date knowledge on the cave-dwelling Mediterranean sponges is included in 72 publications which can be classified in the following groups:

- I. taxonomy on sponges (Sarà, 1959; Pulitzer-Finali, 1969; Voultziadou *et al.*, 1991; Carballo & Garcia-Gomez, 1995; Corriero *et al.*, 1996, 1997; Pansini 1984, 1996; Vacelet & Boury-Esnault, 1996; Bavestrello *et al.*, 1997; Muricy *et al.*, 1998; Pansini & Pesce, 1998; Vacelet & Peres, 1998; Perez *et al.*, 2004; Manconi *et al.*, 2006; Vacelet *et al.*, 2007; Manconi & Serusi, 2008; Pisera & Vacelet, 2011; Manconi, in press);
- II. faunistics on sponges (Sarà, 1958, 1961, 1968; Russ & Rutzler, 1959; Vacelet, 1961, 1969; Rutzler, 1966; Boury-Esnault, 1971; Pouliquen, 1972; Pulitzer-Finali, 1972; Pulitzer-Finali & Pronzato, 1976, 1980; Pansini *et al.*, 1977; Pulitzer-Finali 1977, 1983; Balduzzi *et al.*, 1985; Bibiloni, 1989, 1993; Uriz *et al.*, 1992; Voultziadou & Koukouras, 1993; Mouricy *et al.*, 1996;

Corriero *et al.*, 2000; Voultsiadou & Vafidis, 2004; Voultsiadou, 2005; 2005; Bakran-Petricioli *et al.*, 2007; Manconi *et al.*, 2009);

- III. III. ecology on sponges (Sarà, 1959, 1960, 1964; Labate, 1965; Pansini & Pronzato, 1982); IV. general faunistics and ecology (Laborel & Vacelet, 1958; Cinelli *et al.*, 1977; Balduzzi *et al.*, 1980, 1982, 1989; Bibiloni *et al.*, 1984; Bianchi & Morri, 1994; Morri *et al.*, 1994; Vacelet *et al.*, 1994; Southward *et al.*, 1996; Belmonte *et al.*, 1999; Corriero *et al.*, 1999, 2004; Arko-Pjevac *et al.*, 2001; Harmelin *et al.*, 2003; Belmonte *et al.*, 2006; Bussotti *et al.*, 2006; Moscatello & Belmonte, 2006; Pitruzzello & Russo, 2008; Touron *et al.*, 2009; Denitto *et al.*, 2010).

Additional data have been included after the analysis of the samples from 3 caves of the Capo Caccia-Isola Piana MPA, 3 caves of the Plemmirio MPA, and one cave of the Pelagie MPA.

For the purposes of this study, the Mediterranean Sea was divided into 14 subareas (Fig. 1; Tab. 1), according to the areas investigated in the past by cave sponge workers.

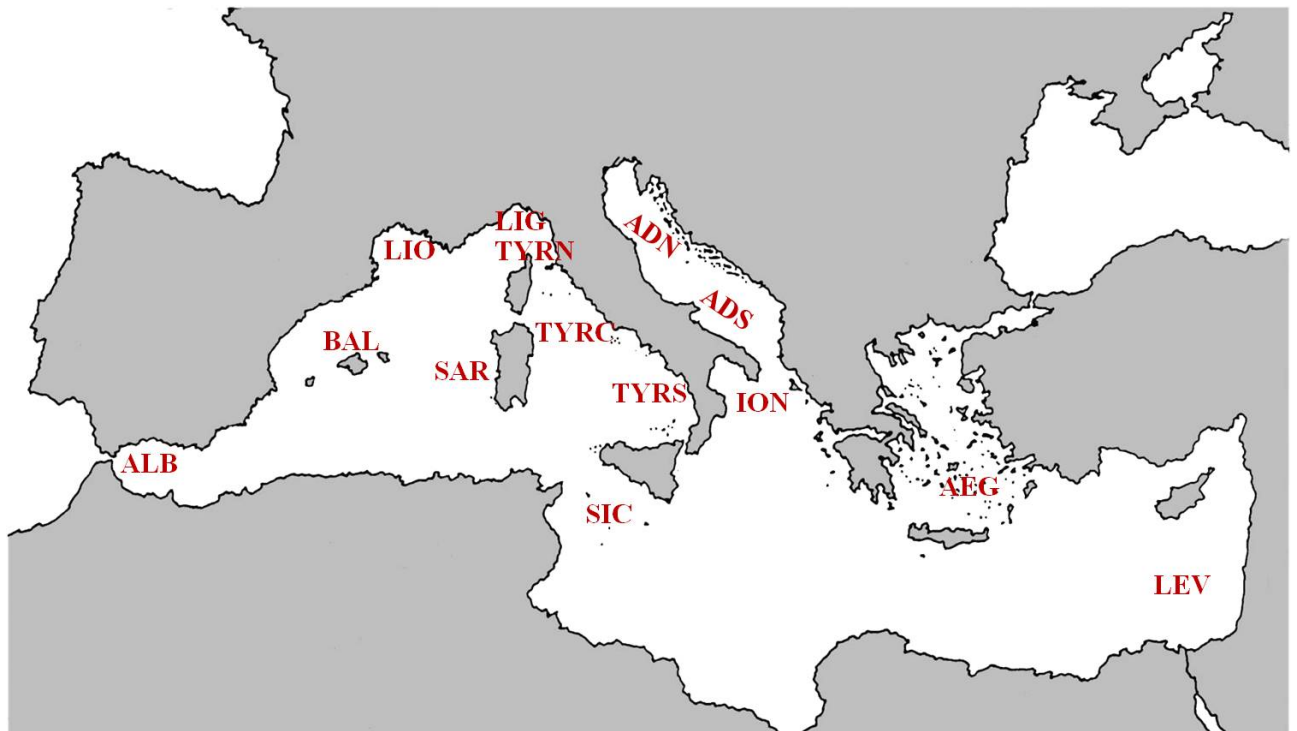


Fig. 1. Maps of the Mediterranean Sea indicated the 14 subareas. ALB: Alboran Sea; BAL: Balearic Sea; SAR: Sardinia Sea; LIO: Golfe du Lion; LIG: Ligurian Sea; TYRN: Northern Tyrrhenian Sea; TYRC: Central Tyrrhenian Sea; TYRS: Southern Tyrrhenian Sea; SIC: Sicily Channel; ION: Ionian Sea; AND: Northern Adriatic Sea; ADS: Southern Adriatic Sea; AEG: Aegean Sea; LEV: Levant Basin.

The sponges recorded in Mediterranean marine caves were assigned to five biogeographical categories according to their distribution as derived from the relevant literature: endemic species (E) distributed exclusively in the Mediterranean, Atlantic–Mediterranean species (AM) distributed in the Mediterranean and the eastern Atlantic, amphi-Atlantic species (AA) distributed in the Mediterranean and both sides of the Atlantic, Indo–Mediterranean species (IM) distributed in the Mediterranean and the Indo–Pacific, and species with a wide distribution or cosmopolitan species (C) distributed at least in the Atlantic and the Indo–Pacific. Some species belonging to these categories are considered potentially Mediterranean endemics.

The faunal composition based on the literature, was assembled in a presence/absence matrix. The resulted checklist was critically reviewed for synonymies following recent trends in Porifera taxonomy Systema Porifera (Hooper & Soest, 2002) and Fauna d’Italia (Manconi, 2012; Pansini, 2012; Pronzato & Manconi, 2012) and after a comparative analyses vs taxonomic database such as the World Porifera Database (Soest et al., 2011; www.marinespecies.org/porifera).

Presence/absence data of cave-dwelling taxa was tested by statistical analysis with Primer6. A comparative analysis was performed on the checklist based on the historical data vs the checklist updated with new data (Manconi *et al.*, in press). Endemicity values and shared species/genera/families/orders were evaluated.

Results

Species number and species presence in the caves of the Mediterranean subareas are shown in **Appendix**.

The sponge fauna of the Mediterranean caves resulted in a total of about 300 species, belonging to 136 genera, 71 families, 18 orders and 3 classes. The class Demospongiae is the most represented with 252 species belonging to 117 genera, 58 families and 12 orders.

The number of relevant publications was positively correlated with the number of species known per area (Fig. 2).

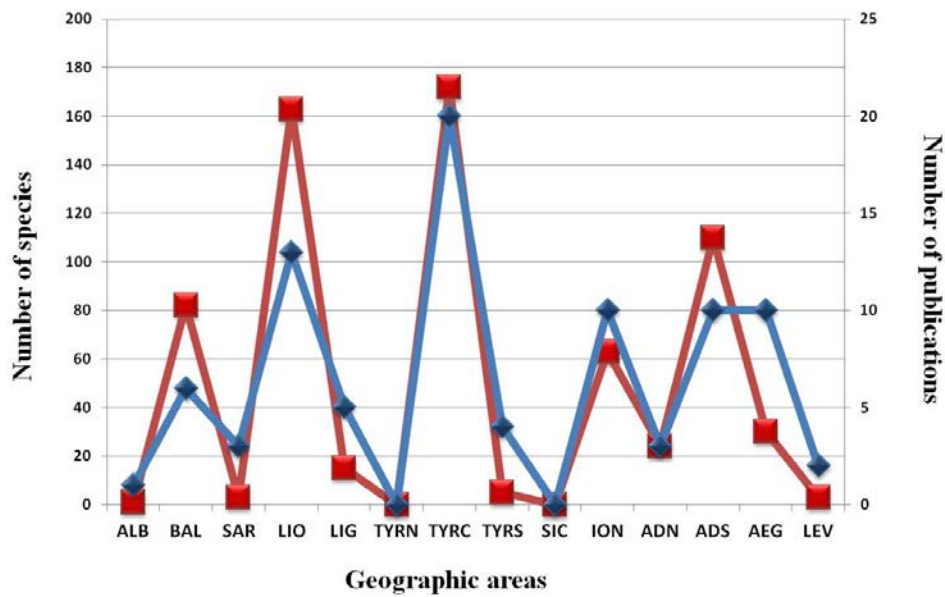


Fig. 2. Relationship between species number (red) and degree of exploration (blue) in the marine caves of the Mediterranean areas

As for historical data for the Mediterranean Sea, the number of papers dealing with cave-dwelling Porifera existing is 72. The most investigated areas are the Central Tyrrhenian Sea, the Golfe du Lion focused in 20 and 13 papers, respectively, and the Ionian Sea, Southern Adriatic Sea and the Aegean Sea 10 papers. Less studied areas are the Balearic Sea, the Ligurian Sea, the Southern Tyrrhenian Sea with 6, 5 and 4 papers, respectively, together with the Sardinia Sea and Northern Adriatic Sea with 3 papers. No data exists for the Northern Tyrrhenian Sea and Sicily Channel (Tab. 1)

As for taxonomic richness the number of recorded porifera in the Mediterranean caves showed that the highest values are in the Central Tyrrhenian Sea with 175 species, Golfe du Lion with 161 species, and Southern Adriatic Sea with 110 species, the Balearic Sea with 80 species, and Ionian Sea with 79 species (Tab. 1).

	TOT	ALB	BAL	SAR	LIO	LIG	TYRN	TYRC	TYRS	SIC	ION	ADN	ADS	AEG	LEV
Publications number	72	1	6	3	13	6	0	20	4	1	11	3	10	10	2
Species	296	1	78	39	161	45	0	175	5	24	79	24	110	39	3
Genera	138	1	54	28	92	34	0	100	5	17	54	20	70	27	3
Families	66	1	33	22	57	27	0	55	5	14	35	19	45	20	3
Orders	17	1	12	10	17	11	0	13	5	7	16	12	14	9	2
AM Species	130	0	38	15	83	20	0	85	2	12	37	10	53	14	0
E Species	114	1	19	11	45	11	0	50	2	4	18	5	28	18	3
C species	38	0	18	12	25	15	0	31	0	7	21	9	23	5	0
AA Species	9	0	1	0	5	0	0	5	1	0	3	0	2	1	0
IM Species	5	0	2	0	4	1	0	3	0	1	0	0	4	1	0

Tab. 1. General faunal and geographical data of the porifera Caves in the Mediterranean areas.

The most common species of the Mediterranean Sea caves (see **Appendix**) are *Clathrina clathrus*, *Petrobiona massiliana*, *Oscarella lobularis*, *Jaspis johnstoni*, *Erylus discophorus*, *Erylus euastrum*, *Cliona viridis*, *Diplastrella bistellata*, *Spirastrella cunctatrix*, *Aaptos aaptos*, *Terpios fugax*, *Chondrosia reniformis*, *Crambe crambe*, *Phorbas tenacior*, *Merlia normani*, *Axinella damicornis*, *Axinella verrucosa*, *Acanthella acuta*, *Agelas oroides*, *Haliclona (Soestella) mucosa*, *Petrosia (Petrosia) ficiformis*, *Ircinia variabilis*, *Sarcotragus fasciculatus*, *Spongia officinalis*, *Spongia virgultosa*, *Dysidea avara*, and *Dysidea fragilis*.

The species exclusively recorded in caves of a single Mediterranean geographical area are as follows: *Axinella estacioi* (1 cave, Strait of Gibraltar); *Stryphnus ponderosus*, *Merlia lipoclavidisca*, *Axinella rugosa*, *Scopalina azurea*, *Spongosorites cavernicola*, *Topsentia garciae*, *Haliclona (Reniera) aquaeductus*, *Hyrtios collectrix* reported in the caves of the Balearic Sea;

Aciculites mediterranea, *Neophrissospongia nana*, *Ircinia retidermata*, *Spongia lamella*, *Spongia zimocca* reported in the caves of the Sardinia Sea;

Clathrina cerebrum, *Clathrina primordialis*, *Clathrina reticulum*, *Guancha lacunosa*, *Leuconia johnstoni*, *Leuconia nivea*, *Sycon sycandra*, *Ute glabra*, *Plectroninia hindei*, *Oscarella viridis*, *Plakina crypta*, *Plakina endoumensis*, *Plakina jani*, *Erylus expletus*, *Geodia gigas*, *Pachastrella monilifera*, *Thrombus abyssi*, *Timea crassa*, *Thymosiopsis cuticulatus*, *Neophrissospongia endoumensis*, *Neophrissospongia nolitangere*, *Neoschrammeniella bowerbanki*, *Clathria (Clathria) compressa*, *Clathria (Clathria) coralloides*, *Clathria (Microcion) spinarcus*, *Clathria (Microcion) strepsitoxa*, *Clathria*

(Thalysias) jolicoeuri, *Eurypon clavatum*, *Rhabderemia gallica*, *Rhabderemia spinosa*, *Rhabderemia topsenti*, *Rhabderemia toxigera*, *Forcepia (Leptolabis) luciensis*, *Crella (Pytheas) rosea*, *Crella (Pytheas) sigmata*, *Hamigera hamigera*, *Myxilla (Myxilla) macrosigma*, *Mycale (Aegogropila) contareni*, *Mycale (Mycale) macilenta*, *Merlia deficiens*, *Axinella vacoleti*, *Axinyssa papillosa*, *Siphonochalina subcornea*, *Haliclona (Gellius) laxa*, *Haliclona (Gellius) microsigma*, *Haliclona (Reniera) citrina*, *Haliclona (Reniera) mediterranea*, *Haliclona aperta*, *Gelliodes luridus*, *Aka labyrinthica*, *Spongionella pulchella* reported in the caves of the Golfe du Lion;

Delectona madreporica, *Hymedesmia (Hymedesmia) castanea*, *Phorbas topsenti*, *Mycale (Mycale) tunicata*, reported in the caves of the Ligurian Sea;

Leucosolenia variabilis, *Sycon ciliatum*, *Sycon quadrangulatum*, *Leucandra crambessa*, *Amphoriscus salfi*, *Placinolopha moncharmonti*, *Tetilla repens*, *Erylus mammilaris*, *Pachymatisma johnstonia*, *Caminus vulcani*, *Poecillastra compressa*, *Thenaea muricata*, *Triptolemma simplex*, *Cliothosa hancocki*, *Spiroxya heteroclyta*, *Paratimea pierantonii*, *Paratimea loricata*, *Polymastia penicillus*, *Rhizaxinella pyrifer*, *Tethya citrina*, *Petromica (Petromica) grimaldi*, *Clathria (Microciona) duplex*, *Clathria (Microciona) gradalis*, *Clathria (Microciona) toximajor*, *Antho (Antho) coriacea*, *Eurypon viride*, *Raspaciona calva*, *Raspaciona robusta*, *Lissodendoryx (Lissodendoryx) basispinosa*, *Crella (Pytheas) nodulosa*, *Crella (Yvesia) topsenti*, *Hymedesmia (Hymedesmia) pansa*, *Hymedesmia (Hymedesmia) zetlandica*, *Hymedesmia (Stylopus) coriacea*, *Hymedesmia (Stylopus) nigrescens*, *Hymedesmia (Stylopus) rectiraphis*, *Mycale (Mycale) dentata*, *Acanthella annulata*, *Tethyspira spinosa*, *Spongisorites intricatus*, *Chalinula limbata*, *Oceanapia decipiens*, *Sarcotragus pipetta*, *Dysidea tupa*, *Spongionella gracilis* reported in the caves of the Central Tyrrhenian Sea;

Didiscus spinoxeatus reported in a single cave of South Tyrrhenian sea;

Cliona carteri, *Pione fastifica*, *Timea geministellata*, *Didiscus stylifer*, *Higginsia ciccaresei* reported in the caves of the Ionian Sea;

Neophrissospongia radjae, reported in a single cave of the island of Korkula (Dalmatian coast);

Cliona vermifera, *Timea stellifasciata*, *Eurypon vesciculare*, *Rhabderemia indica*, *Phorbas plumosus*, *Mycale (Mycale) lingua*, *Dendrectilla tremitensis*, *Haliclona (Halichoclona)*

parietalis, *Haliclona (Gellius) dubia*, *Cacospongia proficiens* reported in the caves of the Southern Adriatic sea;

Plakina weinbergi, *Stryphnus mucronatus*, *Hemiasterella aristoteliana*, *Discodermia polydiscus*, *Petrosia (Strongylophora) vansoesti*, *Ircinia paucifilamentosa*, *Coscinoderma sporadense* reported in the caves of the Aegean Sea;

Microscleroderma lamina, *Euryspongia raouchensis* reported in the caves of the Levantine basin.

The allocation of sponge species occurring in the Mediterranean caves to the different geographical categories (Table 1, **Appendix**) was: 130 Atlantic–Mediterranean species (44%), 114 endemic species (38%), 38 cosmopolitan species (13%), 10 amphi-Atlantic species (3.4%) 5 Indo-Mediterranean species (1.7%).

The allocation of species to the main orders of the 3 classes (Table 2) showed that the richest order, in terms of species number in the Mediterranean caves, was Poecilosclerida (24% of the total species) followed by the Hadromerida and Halichondrida (both 12% of the total species). The other orders with a conspicuous number of species are Haplosclerida (10.7% of the total species) and Dictyoceratida (9.4% of the total species).

Orders of Porifera	ALB	BAL	SAR	LIO	LIG	TYRN	TYRC	TYRS	SIC	ION	ADN	ADS	AEG	LEV
Lyssacinosida	0	0	0	1	0	0	0	0	0	0	1	1	0	0
Clathrinida	0	3	2	9	4	0	6	0	2	3	0	5	0	0
Baerida	0	0	0	2	0	0	0	0	0	0	0	0	0	0
Leucosolenida	0	0	0	3	3	0	8	0	0	2	1	2	0	0
Lithonida	0	0	1	2	1	0	1	1	1	1	0	0	0	0
Homosclerophorida	0	4	2	14	0	0	10	0	0	5	1	5	7	0
Spirophorida	0	0	0	0	0	0	2	0	0	1	0	1	0	0
Astrophorida	0	6	5	15	2	0	17	1	1	6	0	9	6	0
Hadromerida	0	7	6	14	4	0	26	0	3	15	3	18	6	0
Chondrosida	0	2	0	4	1	0	3	0	1	2	1	2	0	0
Lithistida	0	1	2	5	0	0	2	0	0	1	2	0	3	2
Pocilosclerida	0	11	2	42	10	0	40	1	5	12	4	22	2	0
Halichondrida	1	15	2	13	3	0	22	1	1	8	3	14	4	0
Agelasida	0	1	1	1	1	0	1	0	1	1	1	1	1	0
Haplosclerida	0	9	4	17	2	0	14	1	1	7	3	15	4	0
Dictyoceratida	0	15	11	15	10	0	18	0	7	14	2	13	6	1
Dendroceratida	0	2	0	3	2	0	3	0	1	0	0	1	0	0
Verongida	0	2	0	1	1	0	2	2	0	1	2	1	0	0

Tab. 2. Comparison of Mediterranean geographic areas according to their order diversity: species number for every order.

The taxonomic richness of the geographical areas has been compared with the *average taxonomic distinctness* (avTD, $\Delta +$).

As regards the checklist containing historical data the Alboran Sea, the Sardinia Sea, the North Tyrrhenian Sea, the Sicily Channel and the Levantine Basin have a avTD smaller in comparison to other geographical areas, which have values ranging from 95.08 to 96.96.

The taxonomic richness of the geographical areas has been also compared with the *variation in taxonomic distinctness* (varTD, $\lambda +$).

According to this analysis the Alboran Sea, the Sardinia Sea, the North Tyrrhenian Sea, the Sicily Channel and the Levantine Basin have values of varTD different from the other geographical areas ranging from 106.48 to 213.88.

As for the historical checklist updated with the new data (Manconi et al., in press), it has been noticed that the Sardinia Sea and Sicily Channel have values of avTD and values of varTD similar to that of other geographical areas.

Cluster analysis on data of the historical checklist indicated two distinct groups for areas (Fig. 3). The Central Tyrrhenian Sea and the South Adriatic Sea cluster together with 62% of similarity. To this group he associates the Balearic Sea (56% of similarity), the Gulf of Lion (55% of similarity). The second group includes North Adriatic Sea and the Aegean Sea with a 37.5 % of similarity.

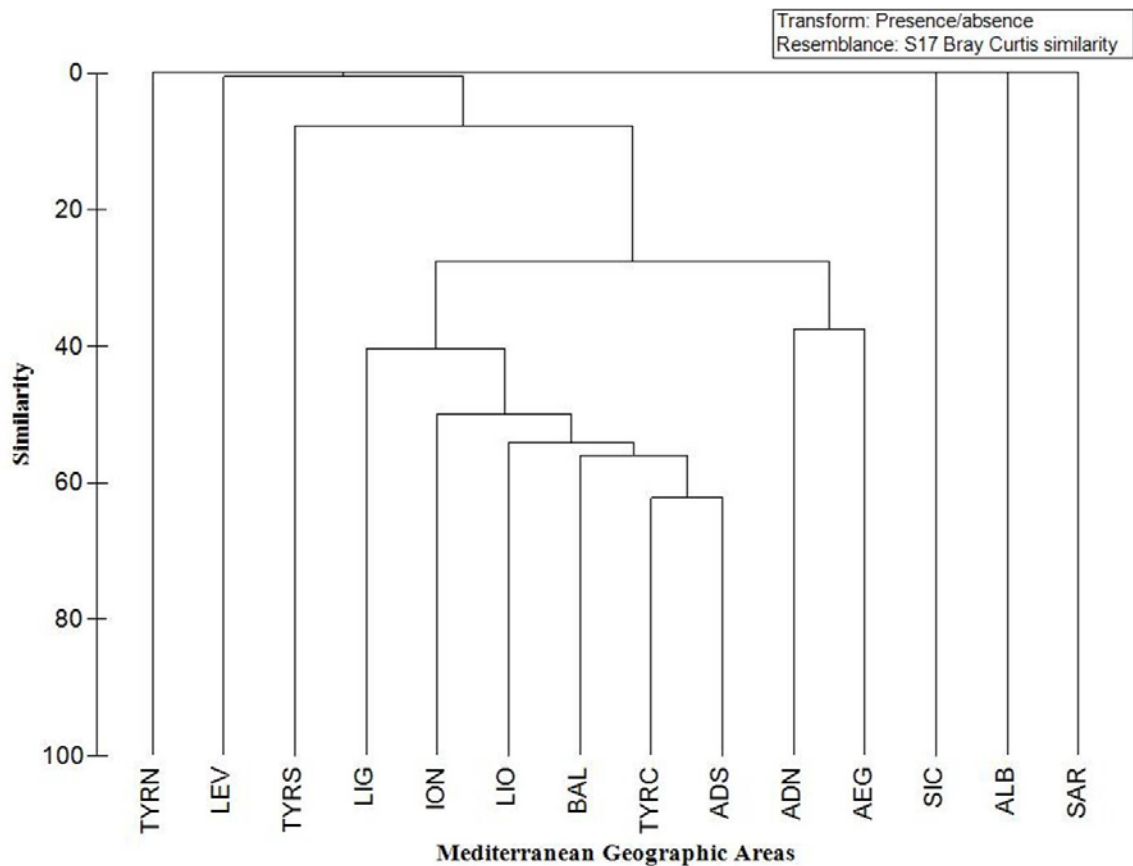


Fig. 3. Cluster analysis among the Mediterranean geographic areas as regards historical data. Cluster analysis of the historical checklist updated with the new data (Manconi *et al.*, in press) indicates two groups (Fig. 4). The first group includes the South Adriatic Sea and the Ionian Sea with 61% of similarity. The Central Tyrrhenian Sea (57% of similarity), the Balearic Sea (52.7% of similarity) and Gulf of Lion (50% of similarity) are associated to the latter group, The second group includes the Sardinia Sea and the Sicily Channel (48% of similarity).

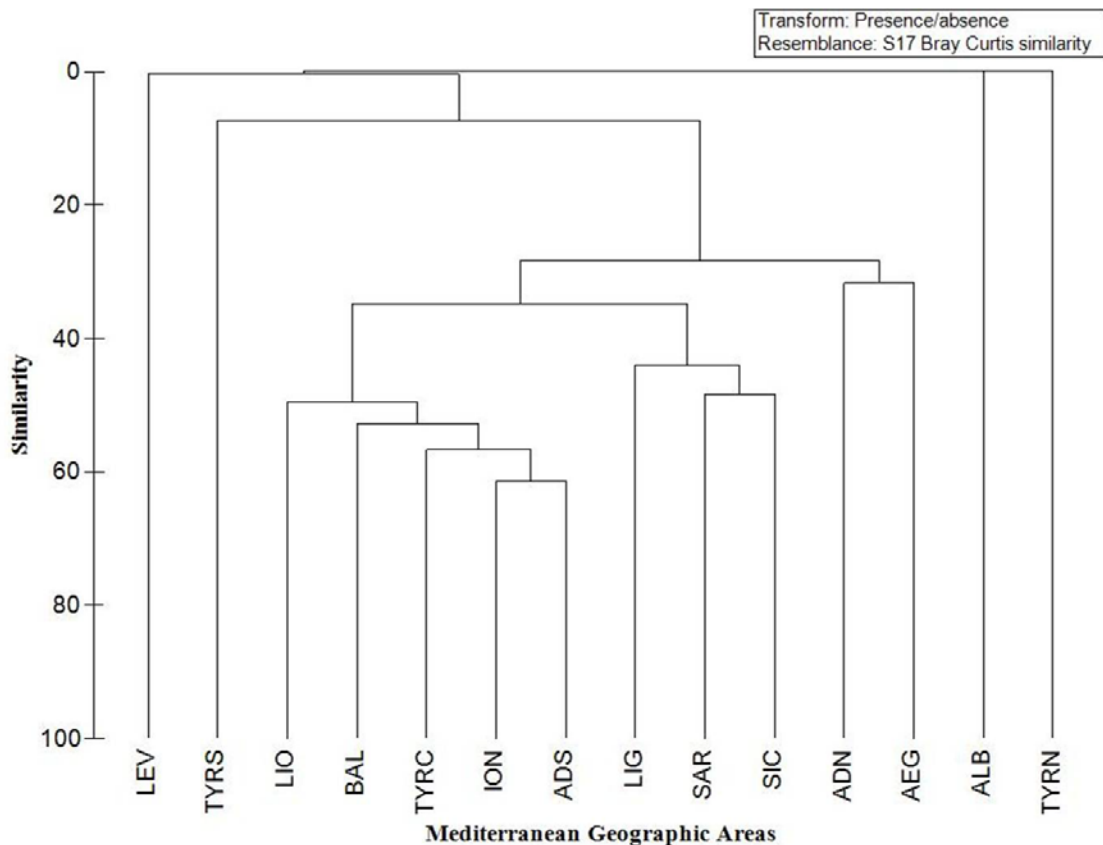


Fig. 3. Cluster analysis among the Mediterranean geographic areas as regards historical data updated with the new data.

Discussion

To date, the sponge fauna of the Mediterranean caves, counting up to 300 species, constitutes 44% of the total Mediterranean sponges (Coll *et al.*, 2010).

Taxonomic analysis indicated a clear separation of the western part from the eastern part of the Mediterranean sea, related to the publications number of the Mediterranean areas.

The sponge distribution between the western and the eastern part of the Mediterranean basin, separated by an ideal border line starting from Tunisia(Cape Bon) and arriving to Greece, encompassing the southern Italian coasts. The western part of the basin is much richer than the eastern part (Pansini & Longo, 2003). This is confirmed by the analysis of our checklist. In the western part of the Mediterranean Sea the highest specific richness (500 species) was recorded along the northern coast of the basin, where the areas mostly represented are the Balearic Sea, the Gulf of Lion and Central Tyrrhenian Sea. Despite the presence of various articles for the Alboran Sea, only one species of cave is signaled

Axinella estacioi, against 184 species censused in this area for other environments (Pansini & Longo, 2003).

In the few published taxonomic papers for the coasts of Tunisia, Malta, southwestern Sicily and some banks of the Sicily Channel, sponges cave are not been signaled. Libyan coasts are unknown. The new records in the caves of Sardinia Sea and Sicily Channel (Manconi et al., in prep) have increased the species number in these areas. This is shown by the assessment of the taxonomic richness, where the Sardinia Sea and the Sicily Channel assume values similar to those of the other geographical zones, and by the cluster analysis where the two areas are together.

In the eastern part of the Mediterranean sea are been reported 279 cave species. On 43 species reported for the Levant basin (Pansini e Longo, 2003), only three are the species cave, *Microscleroderma lamina*, *Gastrophanella phoeniciensis*, *Euryspongia raouchensis*. Another poorly studied area is the aegean sea, where have been reported 39 species cave against 62 species reported in other environments (Pansini e Longo, 2003).

The Ionian sea sponges were carefully studied along the Apulian coasts, where have been reported 79 sponge cave against 181 species reported also in other environmental (Pansini e Longo, 2003).

New data have been reported for this geographical area thanks to the census effected in the Plemmirio MPA (Manconi *et al.*, in prep).

The number species cave reported in the northern and southern part of the Adriatic Sea, doesn't confirm the Pansini & Longo, 2003 data, where the two areas are almost balanced (159 and 157 demosponge species respectively). In fact in the southern Adriatic sea have been reported 110 species cave, while in the northern Adriatic sea have been reported only 24 species cave.

The analysis of the distribution of the geographical categories showed that most of the species occurring in the caves of the Mediterranean sea were Atlanto–Mediterranean, followed by the endemics, the cosmopolitan and the few amphi-Atlantic and Indo-Mediterranean species.

The Atlanto-Mediterranean are the most important group of the sponge-fauna in the Mediterranean cave, due to the intense exchange (in both direction) with the Atlantic. An elevate number of species Atlanto-Mediterranean in the eastern part confirm that the exchange of faunal elements between the Atlantic and the Mediterranean extends to the

eastern part of the latter (Pansini 1992; Koukouras et al. 2001; Voultziadou, 2005). The endemic species are the another important group of the sponge-fauna in the Mediterranean cave, due to the peculiar characteristics of this environment. The western part of the Mediterranean sea has a more elevated number of endemic species in comparison to the eastern part. This confirm the theory of Voultziadou 2005, according to which the endemic species numbers in the Mediterranean follow the general west–east gradient. A another group of species with a cosmopolitan distribution appears very numerous but is certainly over estimated. In effect the accurate study of species regarded as cosmopolitan by electronic microscopy, genetic divergence, cytological criteria and molecular taxonomy often allows to split them into more than one species (Pansini & Longo, 2003).

On 137 genera recorded in the Mediterranean cave, 60 have a cosmopolitan or wide-spread distribution, that is having no distinct centre of diversity and occurring in all three oceans (van Soest, 1994; Hooper & van Soest, 2002). 38 genera have a a distinct tropical-subtropical diversity center, lacking or rare in cold water areas (van Soest, 1994; Hooper & van Soest, 2002).9 genera showing a distinct diversity center in cold water (van Soest, 1994). 30 genera showing a disjunct or restricted distribution, for example North Atlantic-(southern) Indian Ocean (*Guancha*, *Pachymatisma*, *Triptolemma*, *Thrombus*, *Hemiassterella*, *Halisarca*, *Neoschrammeniella*, *Hamigera*, *Monocrepidium*, *Petromica*, *Calyx*); Circum-African (*Crambe*, *Hemimycale*). The genera *Plectroninia*, *Petrobiona*, *Thymosiopsis*, *Raspaciona*, *Dendrectilla* are endemic of the Mediterranean Sea. The genus *Aplysina* is distributed in the east-Pacific and Atlantic Ocean. The genera *Placinolopha*, *Delectona*, *Microscleroderma*, *Spiroxya*, have a distribution in the Indo-Pacific Ocean and Mediterranean (van Soest, 1994; Hooper & van Soest, 2002).

An estimation of sponge diversity at the order level, shows the order mostly represented is Poecilosclerida. The major family of this order such Microcionidae, Hymedesmiidae, Raspailidae, show a worldwid distribution (van Soest, 1994; Hooper & van Soest, 2002). The other order mostly represented are Hadromerida and Halichondrida. The major family of these orders Halicondriidae, Clionidae shows a cosmopolitan distribution.

The data clearly show that the knowledge of the distribution of the sponge fauna in the Mediterranean caves is far from uniform, and is directly proportional to the number of studies performed in each environmental. It is highly probable, therefore, that the actual difference among the sponge-faunas cave of the considered zones are over estimated.

SPECIES	ALB	BAL	SAR	LIO	LIG	TYRN	TYRC	TYRS	SIC	ION	ADN	ADS	AEG	LEV	ZC
<i>Triptolemma</i> de Laubenfels, 1955															
<i>Triptolemma simplex</i> (Sarà, 1959)															E
Thrombidae Sollas, 1888															
<i>Thrombus</i> Sollas, 1886															
<i>Thrombus abyssii</i> (Carter, 1873)				+											AM
Thoosidae Cockerell, 1925															
<i>Alectona</i> Carter, 1879															
<i>Alectona millari</i> Carter, 1879				+			+					+			AM
<i>Delectona</i> de Laubenfels, 1936															
<i>Delectona madreporica</i> Bavestrello, Calcinai, Cerrano, Sarà, 1997					+										AM
<i>Thoosa</i> Hancock, 1849															
<i>Thoosa mollis</i> Volz, 1939				+			+								AM
Hadromerida Topsent, 1894															
Clionaidae d'Orbigny, 1851															
<i>Cliona</i> Grant, 1826															
<i>Cliona carteri</i> (Ridley, 1881)										+					AA
<i>Cliona celata</i> Grant, 1826				+	+		+			+	+	+			C
<i>Cliona rhodensis</i> Rützler & Bromley, 1981										+		+			E
<i>Cliona schmidtii</i> (Ridley, 1881)			+	+			+			+		+	+		C
<i>Cliona vermifera</i> Hancock, 1867												+			AA
<i>Cliona viridis</i> (Schmidt, 1862)			+	+	+		+			+	+	+			C
<i>Cliothosa</i> Topsent, 1905															
<i>Cliothosa hancockii</i> (Topsent, 1888)							+								C
<i>Pione</i> Gray, 1867															
<i>Pione vastifica</i> (Hancock, 1849)										+					C
<i>Spiroxya</i> Topsent, 1896															
<i>Spiroxya heteroclita</i> Topsent, 1896							+								AM
<i>Spiroxya levispira</i> (Topsent, 1898)				+			+								AM
Hemiassterellidae Lendenfeld, 1889															
<i>Hemiassterella</i> Carter, 1879															
<i>Hemiassterella aristoteliana</i> Voultziadou-Koukoura & Van Soest, 1991													+		E
<i>Paratimea</i> Hallmann, 1917															
<i>Paratimea pierantonii</i> (Sarà, 1958)							+								E
<i>Paratimea loricata</i> (Sarà, 1958)							+								E
Placospongiidae Gray, 1867															
<i>Placospongia</i> Gray, 1867															
<i>Placospongia decorticans</i> (Hanitsch, 1895)							+			+		+	+		E
Polymastiidae Gray, 1867															
<i>Polymastia</i> Bowerbank, 1864															
<i>Polymastia penicillus</i> (Montagu, 1818)							+								AM
Spirastrellidae Ridley & Dendy, 1886															
<i>Diplastrella</i> Topsent, 1918															
<i>Diplastrella bistellata</i> (Schmidt, 1862)			+	+	+		+		+	+		+	+		AM
<i>Diplastrella ornata</i> Rützler & Sarà, 1962							+			+		+	+		E

SPECIES	ALB	BAL	SAR	LIO	LIG	TYRN	TYRC	TYRS	SIC	ION	ADN	ADS	AEG	LEV	ZC
<i>Clathria (Microciona) spinarcus</i> (Carter & Hope, 1889)				+											AA
<i>Clathria (Microciona) strepsitoxa</i> (Hope, 1889)				+											AM
<i>Clathria (Microciona) toximajor</i> Topsent, 1925							+								E
<i>Clathria (Microciona) toxitenuis</i> Topsent, 1925				+	+		+					+			AM
<i>Clathria (Thalysias) jolicoeuri</i> (Topsent, 1892)				+											AM
<i>Antho</i> Gray, 1867															
<i>Antho (Antho) involvens</i> (Schmidt, 1864)				+	+		+					+			AM
<i>Antho (Acarinia) coriacea</i> (Bowerbank, 1874)							+								AA
Raspailiidae Nardo, 1833															
<i>Eurypon</i> Gray, 1867															
<i>Eurypon clavatum</i> (Bowerbank, 1866)					+										AM
<i>Eurypon lacazei</i> (Topsent, 1891)		+		+											AM
<i>Eurypon major</i> Sarà & Siribelli, 1960							+			+					AM
<i>Eurypon vesciculare</i> Sarà & Siribelli, 1960												+			E
<i>Eurypon viride</i> (Topsent, 1889)							+								AA
<i>Raspaciona</i> Topsent, 1936															
<i>Raspaciona aculeata</i> (Johnston, 1842)		+		+			+					+			AM
<i>Raspaciona calva</i> Sarà, 1958							+								E
<i>Raspaciona robusta</i> Sarà, 1958							+								E
Rhabderemiidae Topsent, 1928															
<i>Rhabderemia</i> Topsent, 1890															
<i>Rhabderemia gallica</i> van Soest & Hooper, 1993					+										AM
<i>Rhabderemia indica</i> Dendy, 1905												+			IM
<i>Rhabderemia minutula</i> (Carter, 1876)		+		+			+					+			AM
<i>Rhabderemia spinosa</i> Topsent, 1896				+											E
<i>Rhabderemia topsenti</i> van Soest & Hooper, 1993					+										AM
<i>Rhabderemia toxigera</i> Topsent, 1892					+										E
Chondropsidae Carter, 1886															
<i>Batzella</i> Topsent, 1893															
<i>Batzella inops</i> (Topsent, 1891)					+		+					+			AM
Coelosphaeridae Dendy, 1922															
<i>Forcepia</i> Carter, 1874															
<i>Forcepia (Leptolabis) luciensis</i> (Topsent, 1888)					+										AM
<i>Lissodendoryx</i> Topsent, 1892															
<i>Lissodendoryx (Lissodendoryx)</i> <i>basispinosa</i> Sarà, 1958							+								E
Crambeidae Lévi, 1963															
<i>Crambe</i> Vosmaer, 1880															
<i>Crambe crambe</i> (Schmidt, 1862)		+	+	+	+		+		+	+	+	+			AM

SPECIES	ALB	BAL	SAR	LIO	LIG	TYRN	TYRC	TYRS	SIC	ION	ADN	ADS	AEG	LEV	ZC
Agelasidae Verrill, 1907															
<i>Agelas</i> Duchassaing & Michelotti, 1864															
<i>Agelas oroides</i> (Schmidt, 1864)		+	+	+	+		+		+	+	+	+	+		E
Haplosclerida Topsent, 1928															
Calyspongiidae de Laubenfels, 1936															
<i>Siphonochalina</i> Schmidt, 1868															
<i>Siphonochalina subcornea</i> Griessinger, 1971					+										E
Chalinidae Gray, 1867															
<i>Chalinula</i> Schmidt, 1868															
<i>Chalinula limbata</i> (Montagu, 1818)							+								AM
<i>Dendrectilla</i> Pulitzer-Finali, 1983															
<i>Dendrectilla tremitensis</i> Pulitzer-Finali, 1983													+		E
<i>Dendroxea</i> Griessinger, 1971															
<i>Dendroxea lenis</i> (Topsent, 1892)		+	+	+			+			+					AM
<i>Haliclona</i> Grant, 1836															
<i>Haliclona (Haliclona) varia</i> (Sarà, 1958)													+		E
<i>Haliclona (Halichoclona) fulva</i> (Topsent, 1893)		+	+	+			+			+			+		AM
<i>Haliclona (Halichoclona) parietalis</i> (Topsent, 1893)													+		E
<i>Haliclona (Gellius) fibulata</i> (Schmidt, 1862)							+						+		AM
<i>Haliclona (Gellius) dubia</i> (Babic, 1922)													+		E
<i>Haliclona (Gellius) lacazei</i> (Topsent, 1893)		+		+											AM
<i>Haliclona (Gellius) laxa</i> (Topsent, 1892)				+											E
<i>Haliclona (Gellius) microsigma</i> (Babic, 1922)				+											E
<i>Haliclona (Reniera) aquaeductus</i> (Schmidt, 1862)		+													E
<i>Haliclona (Reniera) cinerea</i> (Grant, 1826)							+			+			+		AM
<i>Haliclona (Reniera) citrina</i> (Topsent, 1892)				+											E
<i>Haliclona (Reniera) cratera</i> (Schmidt, 1862)		+					+			+	+	+	+		AM
<i>Haliclona (Reniera) mediterranea</i> Griessinger, 1971				+											E
<i>Haliclona (Rhizoniera) rosea</i> (Bowerbank, 1866)		+									+		+		AM
<i>Haliclona (Rhizoniera) sarai</i> (Pulitzer-Finali, 1969)		+		+			+			+			+		E
<i>Haliclona (Rhizoniera) viscosa</i> (Topsent, 1888)							+						+		AM
<i>Haliclona (Soestella) mucosa</i> (Griessinger, 1971)		+	+	+	+		+		+	+			+		AM
<i>Haliclona (Soestella) valliculata</i> (Griessinger, 1971)				+			+								AM
<i>Haliclona aperta</i> (Sarà, 1960)				+											E
Niphatidae Van Soest, 1980															
<i>Gelliodes</i> Ridley, 1884															
<i>Gelliodes luridus</i> (Lundbeck, 1902)				+											AM
Phloeodictyidae Carter, 1882															

SPECIES	ALB	BAL	SAR	LIO	LIG	TYRN	TYRC	TYRS	SIC	ION	ADN	ADS	AEG	LEV	ZC
<i>Spongia nitens</i> (Schmidt, 1862)			+	+						+					AM
<i>Spongia officinalis</i> Linnaeus, 1759	+	+	+	+			+			+	+	+			C
<i>Spongia virgultosa</i> (Schmidt, 1868)	+	+	+	+			+			+		+			AM
<i>Spongia zimocca</i> Schmidt, 1862			+												E
<i>Hippospongia</i> Schulze, 1879															
<i>Hippospongia communis</i> (Lamarck, 1813)	+			+			+								E
Dysideidae Gray, 1867															
<i>Dysidea</i> Johnston, 1842															
<i>Dysidea avara</i> (Schmidt, 1862)	+	+	+	+					+	+		+			C
<i>Dysidea fragilis</i> (Montagu, 1818)	+	+	+	+			+		+	+		+			C
<i>Dysidea incrustans</i> (Schmidt, 1862)					+		+		+	+					E
<i>Dysidea tupa</i> (Martens, 1824)							+								E
<i>Euryspongia</i> Row, 1911															
<i>Euryspongia raouchensis</i> Vacelet, Bitar, Carteron, Zibrowius & Perez, 2007														+	E
<i>Pleraplysilla</i> Topsent, 1905															
<i>Pleraplysilla minchini</i> Topsent, 1905				+			+								AM
<i>Pleraplysilla spinifera</i> (Schulze, 1879)	+	+	+				+		+	+					AM
Dendroceratida Minchin, 1900															
Darwinellidae Merejkowsky, 1879															
<i>Darwinella</i> Müller, 1865															
<i>Darwinella australiensis</i> Carter, 1885					+		+								C
<i>Aplysilla</i> Schulze, 1878															
<i>Aplysilla rosea</i> (Barrois, 1876)	+		+	+			+		+			+			C
<i>Chelonaplysilla</i> de Laubenfels, 1948															
<i>Chelonaplysilla noevus</i> (Carter, 1876)	+			+											C
Dictyodendrillidae Bergquist, 1980															
<i>Spongionella</i> Bowerbank, 1862															
<i>Spongionella gracilis</i> (Vosmaer, 1883)							+								AM
<i>Spongionella pulchella</i> (Sowerby, 1804)				+											AM
Verongida Bergquist, 1978															
Aplysinidae Carter, 1875															
<i>Aplysina aerophoba</i> Nardo, 1833							+				+				C
<i>Aplysina cavernicola</i> (Vacelet, 1959)	+		+	+			+				+	+			AM
Ianthellidae Hyatt, 1875															
<i>Hexadella</i> Topsent, 1896															
<i>Hexadella racovitzai</i> Topsent, 1896	+									+					AM

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Paper IV

- Manuscript in preparation-

New records of *Petrobiona massiliana* (Porifera: Calcarea) in
karstic caves of Sicily and Sardinia with notes on spicular
morphometry and autofluorescence

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New records of *Petrobiona massiliana* (Porifera: Calcarea) in karstic caves of Sicily and Sardinia with notes on spicular morphometry and autofluorescence

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Abstract

The discovery of unknown populations of the species *Petrobiona massiliana* is reported from shallow water karstic cave of Sicily and Sardinia. The morphological and morphometric comparative analysis between populations confirms the notably conservative spicular morphotraits notwithstanding the notably distant geographical areas. On the contrary the autofluorescence values of calcareous spicules shows a diverging pattern between Sicilian and Sardinian specimens.

Keywords: *biodiversity of Marine Protected Areas, Mediterranean Sea, cave-dwelling sponges, spicular complement analysis*

Introduction

Petrobiona massiliana Vacelet & Lèvi, 1959 (Porifera: Calcarea: Lithonida) is a species endemic to the Mediterranean Sea considered a palaeoendemism and protected by SPA/BIO Protocol of the Barcelona Convention (Manconi *et al.*, 2009). The status of this Mediterranean endemics is particularly problematic because it involves not only the species level, but also the genus *Petrobiona* Vacelet & Lévi, 1958 and the family Petrobionidae Borojevic, 1979, both monotypic.

The present geographical range of *P. massiliana* includes part of the Eastern Mediterranean (Adriatic Sea, Ionian Sea, Crete, Kallithea Greece, Malta, Tunisia) and part of the western basin (Tyrrhenian Sea, Sardinia Sea, Gulf of Lions). The disjunct distribution of cave-dwelling populations of *P. massiliana* in scattered Mediterranean karstic caves, the life cycle characterized by short larval phases and low power of dispersal

of lecithotrophic larvae, together with the status of protected species (included in IUCN Red List since 1979), focus on *P. massiliana* as a potential umbrella species for protection of cave-dwelling biocoenoses and in general of the biotope of Mediterranean marine caves (Manconi *et al.*, 2009).

We report here new records of *P. massiliana* from karstic caves in the Ionian Sea, the Sicilian Channel and north-western Sardinia in the framework a biodiversity assessment in marine caves of three Marine Protected Areas of the Mediterranean Sea. A morphometric analysis of diagnostic characters at the species level (skeletal spicules) and the analysis of calcareous spicules emission spectra was performed in these geographically distant populations. The morphometric data and autofluorescence values of the spicular complement in Sardinia vs. Sicily populations were compared.

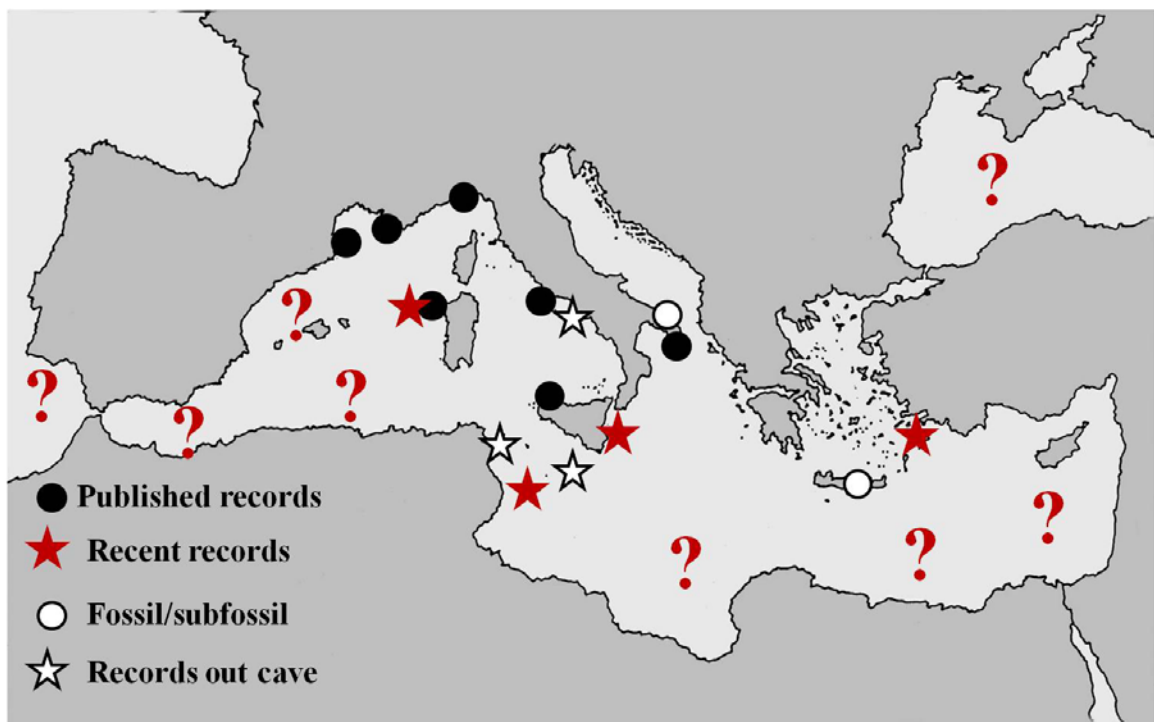


Fig. 1: Disjunct biogeographic patterns on *P. massiliana* in karstic Mediterranean caves

Materials and Methods

The collection of *Petrobiona massiliana* was carried out in the dark to semi-dark zone of 8 submerged shallow water caves during a visual census by SCUBA. The specimens were photographed *in vivo*, collected in small plastic containers, labeled and registered in a

voucher collection. Sponges were preserved in alcohol 70-100 % or dry. Morphological macro-traits such as growth form, size and colour were evaluated by a stereomicroscope, and representative body fragments were dissected to characterize surface morphology and distribution of inhalant and exhalant apertures. Calcareous skeleton fragments and spicules were prepared by digestion in sodium hypochlorite at ambient temperature for a morphometric analysis of all spicular types in light microscopy Ten spicules of all spicular type were analysed by confocal microscope for their 3D reconstruction and the measure of their autofluorescence.

Results

Faunistics

Petrobiona massiliana was discovered in karstic caves of the Sicilian karst (Fig. 1). Conspicuous populations characterize large areas of the dark zone of Gymnasium cave, Mazzere cave and Gamberi cave within the Plemmirio MPA along the eastern coasts of Sicily (Tab. I). Two other populations were recorded in the Taccio Vecchio I cave and Taccio Vecchio II cave of Lampedusa in the Pelagie MPA (Tab. I).

The new faunistic investigations in Sardinia highlighted also the presence of unknown populations in three previously not surveyed caves (Bisbe cave, Falco cave and Archi cave) in the northwestern Sardinian karst within the Capo Caccia-Isola Piana MPA.

All populations were found in the semi-dark to totally dark zones of shallow water caves, typically in facies of variable density.

Morphology and morphometry

Growth form ranged from encrusting to erected, from lobate to digitiform. Body size ranged from thin crusts to 1-2 cm in height and 0.5-1 cm in diameter.

Morphometries of spicules from Sardinia shows the following values (Tab. I). Triactines 32.5-200 μm (in length) x 5-30 μm (in width); diapason triactines: basal actin 30-75 μm (in length) x 5-10 μm (in width), lateral actin 20-57 μm (in length) x 2-8 μm (large); pugiole tetractines: assial actin 50-190 μm (in length) x 5-20 μm (in width), lateral actin 20-170 μm (in length) x 5-20 μm (in width); microdiactines 18-55 μm (in length) x 2-5 μm (in width); macrodiactines 102.5-112.5 x 5 μm .

Morphometries of spicules from Sicily shows the following values (Tab. I).. Triactines 32-210 μm (in length) x 6-27 μm (in width); diapason triactines: basal actin 32-100 μm (in length) x 5-7 μm (in width), lateral actin 15-67 μm (in length) x 2-7 μm (in width); pugiole tetractines: assial actin 65-170 μm (in length) x 2-10 μm (in width), lateral actin 20-170 μm (in length) x 5-20 μm (in width); microdiactines 20-70 μm (in length) x 2-5 μm (in width); macrodiactines 102-235 x 5-17 μm .

The spicular complement of investigated Sardinian and Sicilian populations of *P. massiliana* is characterized by the presence of a rare spicular type, namely macrodiactines.

Locality/cave	Triactines (μm)	Diapason (μm)	Tetractines (μm)	Microdiactines	References
				(μm)	
Marseille type locality/Holotype	50-200 x 20-40*	70-100 total*	300 ax x 80 lat x 8-27 thick*	30-40 x 2*	Vacelet & Levi 1958*
	25-200 x 6-40	30-70 x 5-8.5 bas 20-50 x 4-7 lat	40-130 x 22-28 lat 16-40 x 5.5 8-100 x 10-28 ax 30-70 x 5.5-8.5 ax 200 max ax x 100 max ax x 15 max thick	30-60 x 2-3 °	Vacelet et al. 2002°
Apulia Teatrino Cave	150 x 20-30 30-50 x 5-15	70-75 x 5 total	130-150 ax x 80-135 lat X 7.5-18 thick	30-40 x 2-3	Sarà. 1963
Sorrento Peninsula Tuffo-Tuffo cave	45-300 x 15-48	75-130 x 6-8 total	100-160 ax x 60-105 lat x 5-10 thick	30-65 x 2-4 23 x 7.5-10	Rutzler. 1996
Punta Giglio Terrazze cave	45-170 x 10-25	35-60 x 3-8 bas 23-48 x 3-5 lat	80-155 ax x 50-95 lat x 5-19 thick	25-40 x 3	Manconi et al. 2009
Punta Giglio Galatea cave	55-175 x 10-25	45-63 x 5-8 bas 20-55 x 4-5 lat	70-190 ax x 45-100 lat x 5-15 thick	25-38 x 2-3	Manconi et al. 2009
Punta Giglio Fantasmi cave	50-200 x 10-30	35-63 x 3-8 bas 20-55 x 3-8 lat	50-180 ax x 35-170 lat x 4-20 thick	28-55 x 2-4	Manconi et al. 2009
Capo Caccia Nereo cave	35-200 x 5-30	40-73 x 5-10 bas 20-50 x 4-8 lat	65-147.5 ax x 20-40 lat x 5-10 thick	18-43 x 3	Manconi et al. 2009
Punta Giglio Falco cave	32.5-142.5 x 7.5-22.5	30-75 x 5-7.5 bas 22.5-57.5 x 2-7.5 lat	66-182.5 ax x 45-87 lat x 5-10 thick	25-5 x 2.5	Present paper
Punta Giglio Bisbe cave	63-145 x 7-25	32.5-55 x 5-7.5 bas 25-45 x 2-5 lat	65-165 ax x 15-40 lat x 2.5-7.5 thick	25-43 x 2.5	Present paper
Lampedusa Island Taccio Vecclio cave	37.5-187.5 x 7.5-25	37.5-60 x 2.5-5 bas 15-65 x 2.5-7.5 lat	100-170 ax x 30-50 lat x 7.5-10 thick	25-47.5 x 2.5	Present paper
Maddalena Peninsula Mazzere cave	37.5-170 x 5-25	32.5-155 x 5-7.5 bas 30-52.5 x 2.5-7.5 lat	110-175 ax x 25-50 lat x 5-12.5 thick	32.5-87.5 x 2.5	Present paper
Maddalena Peninsula Gynnasium cave	65-210 x 12.5-27.5	37.5-60 x 2.5-5 bas 20-75 x 2.5-5 lat	80-135 ax x 25-35 lat x 2.5-10 thick	25-70 x 2.5	Present paper
Maddalena Peninsula Gamberi cave	32.5-130 x 6-17.5	37.5-90 x 5-7.5 bas 32.5-67.5 x 2.5-7.5		29-53 x 2.5	Present paper

Table I. Spicular traits of *Petrobia massiliana* from Sardinian and Sicilian caves to other Mediterranean records. * Original description; ° Systema Porifera; total: total length; bas: basal; lat: lateral; ax: axial; thick: thickness.

Autofluorescence analysis of calcareous spicules

The analysis of autofluorescence in calcareous spicules of *Petrobiona massiliana* by confocal microscopy highlighted peculiar patterns.

For autofluorescence values at an excitation of 488 nm all the *P. massiliana* populations examined have an emission spectrum with a peak at 550 nm (Fig. 2).

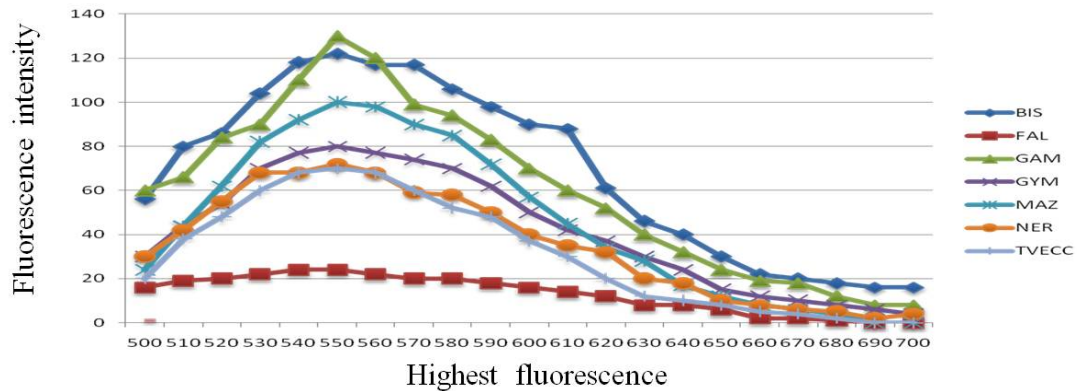


Fig. 2: *Petrobiona massiliana*. Emission spectrum at a wavelength of 488 nm. BIS-Bisbe cave. FAL-Falco cave. NER-Nereo cave. GAM-Gamberi cave. GYM-Gymnasium cave. MAZ-Mazzere cave. TVECC-Taccio Vecchio cave

With an excitation of 496 nm and 514 nm, only the populations from eastern Sicily (Mazzere cave, Gymnasium cave and Gamberi cave) show increasing values of emission up to 560-580 and then decreased. At this wavelength the other populations from Sardinia (n=3) and Lampedusa (n=1) do not emit or emit a very weak fluorescence (Figs 3, 4).

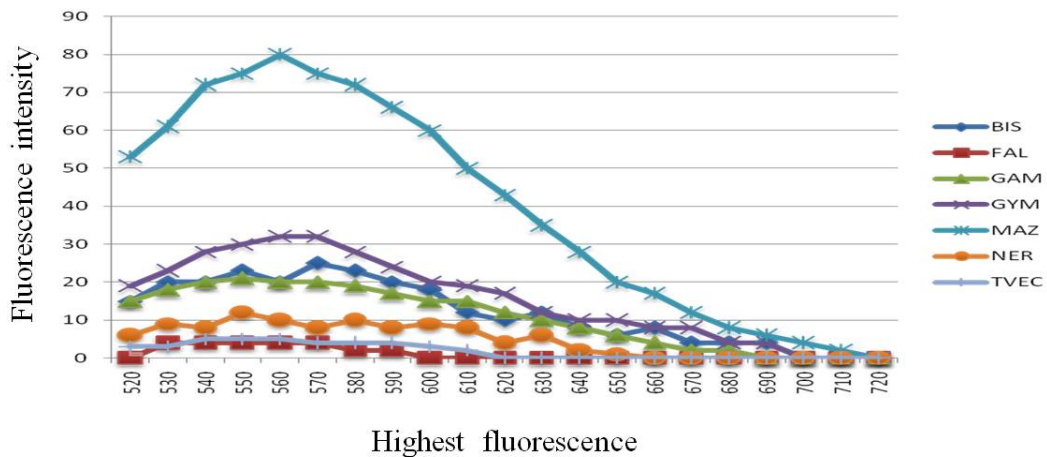


Fig. 3: *Petrobiona massiliana*. Emission spectrum at a wavelength of 496 nm. BIS-Bisbe cave. FAL-Falco cave. NER-Nereo cave. GAM-Gamberi cave. GYM-Gymnasium cave. MAZ-Mazzere cave. TVECC-Taccio Vecchio cave

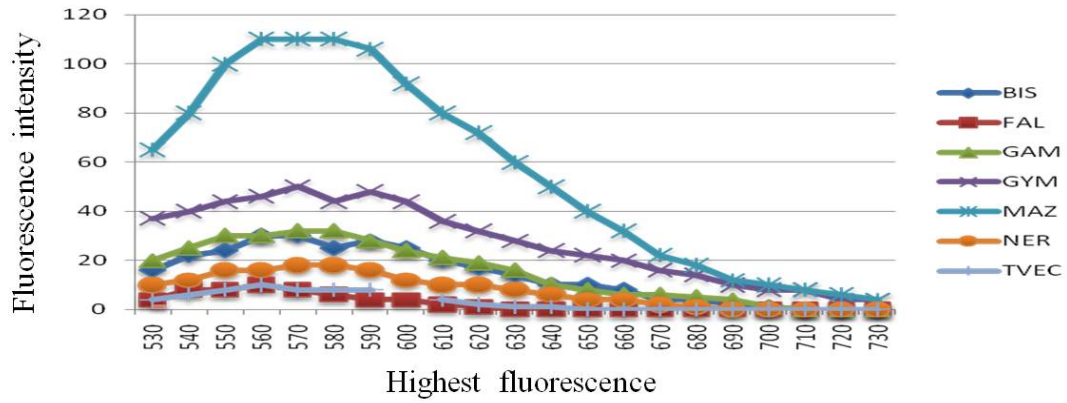


Fig. 4: *Petrobiona massiliana*. Emission spectrum at a wavelength of 514 nm. BIS-Bisbe cave. FAL-Falco cave. NER-Nereo cave. GAM-Gamberi cave. GYM-Gymnasium cave. MAZ-Mazzere cave. TVECC-Taccio Vecchio cave

At a wavelength of 543 nm, the population with the highest values of fluorescence emission is the Mazzere cave (Fig. 5).

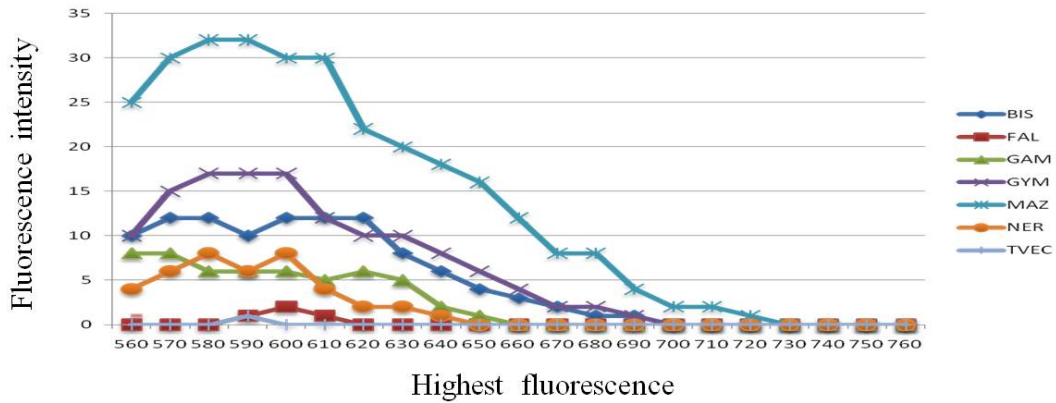


Fig. 5: *Petrobiona massiliana*. Emission spectrum at a wavelength of 543 nm. BIS-Bisbe cave. FAL-Falco cave. NER-Nereo cave. GAM-Gamberi cave. GYM-Gymnasium cave. MAZ-Mazzere cave. TVECC-Taccio Vecchio cave

The highest fluorescence emission at an excitation of 633 nm was observed in populations from Gamberi and Mazzere caves. All the others cave-dwelling populations do not emit or emit a very small fluorescence at this wavelength (Fig. 6)

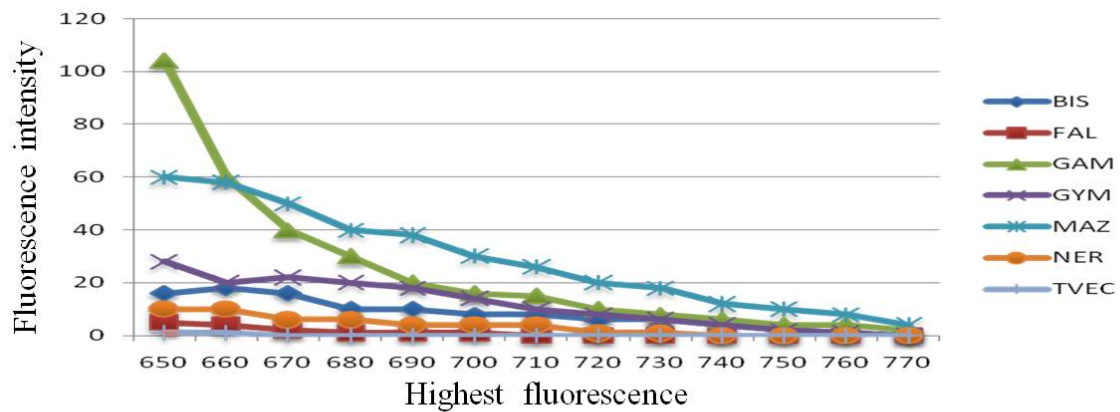


Fig. 6: *Petrobiona massiliana*. Emission spectrum at a wavelength of 633 nm. BIS-Bisbe cave. FAL-Falco cave. NER-Nereo cave. GAM-Gamberi cave. GYM-Gymnasium cave. MAZ-Mazzere cave. TVECC-Taccio Vecchio cave

Discussion

Faunistics

The discovery of *Petrobiona massiliana* in Sicily and Lampedusa represents the first record for the Ionian Sea (Fig. 1). As for the Sicilian Channel, where the species was previously found only in the Zembra MPA (Tunisia) this is the first record for the Archipelago of Pelagie (Fig. 1).

As for Sardinia *P. massiliana* was found in three more than previous four caves recorded by Manconi *et al.* (2009) (Fig. 1). Although focused researches pointed out on the potential enlargement of the geographic range to the caves of Mediterranean Iberian coast and of the Balearic Archipelago the species has not been reported until now in the in the westernmost basin of the Mediterranean (Vacelet, pers. com.). The presence of this species in the Sardinia Sea contrasts with the absence in the neighbouring Balearic Sea over the imaginary border line Marseille-Tunisia, notwithstanding the Sardinia Sea and the Balearic Sea share a common geological origin during the shifting towards east of the Sardinia-Corsica microplate dated to the Miocene.

Morphometric analysis of spicular complement

A preliminary morphometric analysis performed on ten spicules of all spicular type showed no differences between the Sardinian and Sicilian caves populations (Tab. I).

It has been observed, however, the presence in both the Sardinian and Sicilian populations of a rare spicular type, the macrodiactin, reported only once by Vacelet (1964) for some Marseilles cave-dwelling populations and considered rare or absent in most of the specimens studied by the same author in other Mediterranean caves. The morphometries of macrodiactines reported by Vacelet (1964) are in the range 270-480 x 11-20 μm . The new data show that the range of macrodiactine measurements from Sardinian populations is 102.5-112.5 x 5 μm , while the macrodiactine of Sicilian populations are characterized by a range of 102-235 x 5-17 μm .

These data suggest that further morphometrics studies may give more information on this spicular type, their function and position in the skeleton and, in general, on the variability of diagnostic skeletal characters in populations geographically distant.

The emission spectra analysis show interesting differences between the Sicilian and Sardinian populations.

The populations of the Maddalena Peninsula caves have peak emission at different wavelengths in contrast to other populations (Sardinia and Lampedusa) that have shown only one emission spectrum at a wavelength of 488 nm. The most diverging population showing emission at all tested wavelengths is from the Mazzere cave .

The analysis was done on only one type spicules for each population, so it is necessary to extend and develop the study to confirm these first results, and to investigate in depth the causes of the emission spectra divergence of calcareous spicules. Moreover the autofluorescence emission by a organism or a structure can be used to understand the molecular composition of organic and inorganic materials in relation to environmental conditions in different geographic areas and/or the genetic distance between populations.

Hypothetically the species *P. massiliana* can be used as an indicator of climate changes As suggested also by Berry et al., 2005, Gilis et al., 2011 a and b. In fact, changes in environmental conditions such as temperature, salinity and bio-availability nutrients, have an impact on the composition of the calcareous skeletons of many marine species that can be used as natural recorders .

From a faunistic point of, the new records confirm the hypothesis by Manconi *et al.* (2009) that *P. massiliana* is not so rare in the Mediterranean basin but it is represented by several cryptic populations highly fragmented in space mostly restricted to the cave biotope.

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Abstract III

Subterranean Planarians (Platyhelminthes, Tricladida) from Sardinia

Stocchino G.A., Sluys R., Manconi R., Marcia P., **Cadeddu B.**, Corso G. & Pala M.

11th International Symposium on Flatworm Biology, Belgium, July 26-30, 2009

SUBTERRANEAN PLANARIANS (PLATYHELMINTHES, TRICLADIDA) FROM SARDINIA

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In this contribution, we report on records of species of Dugesidae, Dendrocoelidae, Planariidae and Rhynchodemidae from groundwaters in Sardinian karstic caves and springs. The fissiparous specimens of Dugesidae from a NW-Sardinian cave were ascribed to the genus *Dugesia*. Previous reports of this genus from subterranean waters refer only to a few caves along the Italian peninsula. As to Dendrocoelidae, specimens belonging to the genus *Dendrocoelum* were found in two caves (NW Sardinia) and in a spring of Asinara islet. In another spring from the latter islet, very small white specimens of *Phagocata* s.l. (Planariidae) were found. In a cave of SW-Sardinia some depigmented anophtalmous specimens belonging to *Atrioplanaria* sp. (Planariidae) were collected, confirming the distribution of this genus throughout Sardinia, which was previously recorded only from two northern and south-western subterranean waters of the island. Although only an allochthonous species of terrestrial planarian (*Bipalium kewense*) was reported until now from Sardinia, land planarians belonging to an autochthonous species of the Rhynchodemidae were collected from a cave in the north of the island.

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Abstract IV

Tricladids from Italian groundwaters (Platyhelminthes, Tricladida)

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Cadeddu B., Corso G & Pala M.

20th International Conference of Subterranean Biology, Postojna, Slovenia, 29 August- 3 September 2010.

Poster presentation:

Triclad from Italian groundwaters (Platyhelminthes, Tricladida)

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Subterranean triclad from Italy include both stygobiotic and stygophilous species. The first record on triclad from Italian groundwaters is dated as far back as 1890 when Garbini reported the stygophilous species *Polycelis nigra* from two wells near Verona (north-eastern Italy). Up to now, among stygobiotic species, the genera *Dendrocoelum* (Dendrocoelidae), *Atrioplanaria*, *Phagocata* s.l. and *Polycelis* (Planariidae) are reported. Stygophilous species belong to the genera *Dugesia* (Dugesiidae), *Dendrocoelum* (Dendrocoelidae), *Polycelis* and *Crenobia* (Planariidae). Data on both stygobiotic and stygophilous species are mainly restricted to central-northern Italy and Sardinia. This contribution provides an account on the subterranean triclad from Sardinia with new records and a taxonomic synopsis on Italian taxa. Funds were provided by PRIN-MIUR and the EU project Interreg III Sardinia-Corsica-Tuscany. G. Stocchino acknowledges financial support from SYNTHESYS, a programme of the European Commission under the 6th Research and Technological Development Framework Programme “Structuring the European Research Area”, which enabled GS to work at the Zoological Museum Amsterdam in November and December 2008 (grant number: NL-TAF 4717).

Groundwater distribution of aquatic Copepoda in caves of Slovakia: central Europe
 Miroslav Štáhl, Zuzana Váňová