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VIII WORLD SPONGE CONFERENCE 2010

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ancient animals new challenges

BOOK OF ABSTRACTS

VIII WORLD
SPONGE
CONFERENCE

BOOK OF ABSTRACTS

GIRONA, 20-24 SEPTEMBER 2010

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PREFACE

This book of abstracts contains contributions by all participants of the VIII Word Sponge Conference, convened September 2010 in Girona. The primary goal of the VIII Conference has been to continue the traditions of research excellence, international collaboration and knowledge transfer, and sponge-centric enthusiasm that have characterized the Sponge Symposia series since its inception in 1968. Organized by William Fry on behalf of the Zoological Society of London, the 1968 Symposium hosted 40 participants. In the four decades since its origin, the number of Symposia participants has been steadily increasing, as scientists from disparate disciplines unite under the common theme of Poriferan research. In this VIII Conference edition, the Symposia consists of 265 delegates with a mean of 1,3 contributions per delegate.

The areas of research covered by the 354 contributions are largely reflective of the current trends in the Marine Life Sciences. Notably, some of the hot topics in previous Symposia, such as Natural Products and Chemical Ecology, which were born and flourished in Amsterdam (1993), Brisbane (1998) and Rapallo (2002), are poorly represented in the Girona convention. In contrast, the genetic/genomic disciplines have experienced a sudden increase, in particular within the framework of sponge Phylogenetics and Evolution. A total of 154 contributions are attributed to the field of Ecology, 60 are related to cell and organism studies (including gene expression), and 48 focus on Phylogeny and Evolution. Only 28 contributions address the secondary chemistry and the sponge applications. Finally, contrary to the expectations from general scientific trends, the field of taxonomy has survived Conference after Conference and remains a prominent discipline of sponge research, by-passing the changing scientific paradigm.

We are excited about this new Conference and hope you will enjoy the talks, posters, round tables, informal discussions, meeting friends ... and also the country. We have done our best to achieve this goal in the context of a world crisis. But we realize that if the Conference is successful, the merit belongs to you, the attendants. Thanks for your enthusiastic participation and welcome to Girona!

The Organizing Committee

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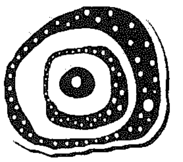

OPENING CONFERENCE

Sponges on reefs: 35 years of collaborative research on the Mesoamerican Barrier Reef

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After more than three decades of research at one coral-reef location, multidisciplinary teams operating under a Smithsonian-sponsored program have generated groundbreaking data and ideas about reef sponges that would be impossible for lone investigators or short- term expeditions to match. Launched by a few biologists and geologists at the National Museum of Natural History, Washington, in the early 1970s, this field research program evolved into the Caribbean Coral Reef Ecosystems (CCRE) endeavor dedicated to the long-term interdisciplinary study of a section of the Mesoamerican Barrier Reef (MBR), the Atlantic's largest reef complex. The MBR extends 250 km southward from Yucatan, Mexico, into the Gulf of Honduras, most of it located off the coast of Belize. It lies 20–40 km from the mainland, separated by a lagoon averaging about 15 m in depth (3 m in the north, 24 m in the south, and 2–4 m on the outer reef platform). Because of the reef's location, continental influences such as excessive nutrient runoff remain minimal, while calm, productive, lagoon waters inshore and low-nutrient blue-water environments offshore are within a few hundred meters. Subsystems include the actual reef (composed of isolated patch reefs, and the barrier back reef, crest, fore reef, and fore-reef slope along the margin of the continental shelf), seagrass meadows, and mangrove islands. Among the varied macrobenthos of this relatively unspoiled reef ecosystem, sponges stand out for their ubiquity, range of color, rich species and biomass, and ecological influence; they populate the rocky hard substrata, some sandy bottoms, and, because the mean tidal range in the area is only 20 cm, the subtidal stilt roots and peat banks of the mangroves as well. In 1972, the CCRE established a field station on Carrie Bow Cay, a tiny sand islet off southern Belize formed by reef-crest debris, to provide year-round support for research by varied experts concerned with investigating biodiversity in the broadest sense, developmental biology, species interaction, oceanographic and carbonate-geological processes, community development over time starting in the Pleistocene, and distributional, physiological, and chemical ecology. Early on, program participants consisted of staff of the National Museum of Natural History, but eventually, despite financial constraints, collaborators were brought in from other academic institutions worldwide. Numerous studies examined the biological and geological role of Porifera in the reef communities. At last count, 113 researchers focused on sponges of the Carrie Bow area, with 88 (78%) conducting fieldwork and the remainder coauthoring publications. Of the fieldworkers, 63 (72%) studied sponges directly, while the rest (25, or 22%) dealt with sponge associates. To date, 117 scientific papers have been published on the results of this research, while many more are in progress.

The earliest comprehensive study of Caribbean sponges, published in 1864 by Duchassaing & Michelotti, dealt exclusively with collections from the Lesser Antilles.

Subsequent work by Schmidt, Carter, Dendy, and Topsent between 1870 and 1890 covered mainly the Gulf of Mexico and the West Indies. The first sponge known from Belize (then British Honduras) was a tiny (5x12-mm) *Polymastia biclavata* (now genus *Coelosphaera)*, sent to England by a local collector and described by B. W. Priest before the Quekett Microscopical Club of London in 1881. This remained the only record from Belize for the next 56 years, until the British Rosaura Expedition of 1937/38 collected five species from Belize City harbor and Turneffe Island atoll; even those specimens were not described until Burton's treatise in 1954.

When our CCRE researchers arrived in Belize in the 1970s, studies centered on systematics and faunistics, including the quantitative distribution of benthic organisms among the various shallow-water habitats. Over the next 30 years or so, taxonomy was approached by methods ranging from basic morphology to fine structure, DNA barcoding, and ecological manipulations. One highlight of these years was a workshop for six experts on Caribbean Porifera held at Carrie Bow Cay. CCRE studies have identified 30 new species, one of them first "recognized" by a predatory starfish, and many more are still under study. Several taxonomic groups were revised locally (among them Clionaidae, Mycalidae, and Chalinidae) or Caribbean-wide (as in the case of *Lissodendoryx* [Coelosphaeridae], *Iotrochota* [Iotrochotidae], and Axinellidae). Researchers not only documented species by photography and drawings , but also analyzed and scientifically illustrated many of the communities dominated by sponges. Several spawning events were recorded, and the development of selected species was studied by electron microscopy. Larvae were sampled from the wild and settlement preferences and development investigated in the laboratory.

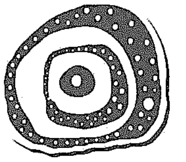
Initially, ecological work focused on the distribution of common species and their role in reef structure and zonation, on the abundance of species with photosynthetic symbionts, and on bioerosion, the latter in collaboration with carbonate geologists during habitat and reef-zonation surveys. Research teams placed limestone plates in all reef sections to measure benthos settlement and growth and excavation rates; explored reef caves and cavities, including the interior of conch shells discarded by local fishermen; and experimented with instruments recording in-situ sponge pumping activity and day-versus-night respiration rates of species hosting cyanobacterial symbionts. Exploration quickly expanded to the nearby Twin Cays mangrove, where stilt-roots and peat banks along its tidal channels were discovered loaded with sponges. When numerous plankton tows over the reef failed to collect sponge larvae, innovative design produced a self-contained, flow-generating sampler that could be deployed inside the reef framework to capture near-bottom plankton.

The range of subsequent research included the cycling of silica and nutrients, symbiosis, disease, mutualism, space competition, and predation. Silicon turned out to be quite rare in this calcium-carbonate-dominated environment, as sponge spicules and a few other siliceous skeletons were rapidly dissolved and reused by new sponge generations. CCRE workers showed that sponge-generated silicon far exceeded the productivity of diatoms off Belize, and, equally important, that unexpected nutrient contributions from sponges are related to nitrogen fixation by bacterial symbionts. With the aid of electron microscopy, morphotypes of the latter from various hosts were studied and their molecular characterizations is currently under way. Particularly significant, some of these symbionts, or certain relatives, are causing sponge disease or contributing to it. Suspected mutualistic relationships relate mainly to interaction between sponges and their diverse invertebrate associates, particularly parazoanthid epizoans and ophiuroid and crustacean endofauna. Interestingly, the relationship with *Parazoanthus* cover appears to be parasitism, for instead of protecting sponges from fish predation, this organism depresses their pumping activity. Meanwhile, brittlestars receive predator protection from their sponge hosts and repay them by cleaning their surfaces of fine sediments.

One of the most striking discoveries on the reefs off Carrie Bow Cay was that alpheid (or snapping) shrimps form complex social organizations inside sponges, the first description of eusociality in the sea, and closely comparable to that in termite nests of the nearby mangrove. Another important finding was that patterns of sponge distribution are governed substantially by competition for space and predation, with ensuing success for encrusting species depending on fast growth rates, the effects of allelochemicals, and the timing of settlement on vacant substrate. Transplant experiments switching reef and mangrove species clearly show that a surprising diversity of spongivores influence distribution.

Environmental factors such as light, temperature, salinity, desiccation, substrate, and sediments were also studied for their effects on sponge individuals and populations. Sponge competitors, for example, were shown to have thrived on shallow-water corals suffering from stress and bleaching (and possibly disease) related to water-warming events and enhanced ultraviolet radiation during some hurricanes. In the Belizean mangroves, temperature and salinity extremes in the inshore lakes and extensive tidal channels affect the structure of the root-fouling community, which in turn benefits the mangrove trees. The composition of healthy sponge populations in a given area provides clues to changes caused by subtle environmental degradation, such as the introduction of pollutants. Some sponges were found to be quite resistant to desiccation during extreme, long-lasting low tides and to have cellular osmoregulation capacity. Although sponges are only modest calcareous skeleton contributors in modern seas, they play a major role in producing limestone mud through bioerosion. At the same time, they incorporate various kinds and sizes of calcareous particles or adopt morphological and physiological strategies for living buried in sand.

Through its contributions to a wide range of publications over the years, CCRE research has strongly demonstrated the importance of sponges in tropical shallow-water habitats and documented effective techniques of preparation, study, and monitoring. Strikingly, sponges in general have fared better than reef corals—vital contributors to the substrate and geomorphology of the reef ecosystem—following the recent degradation of the once-pristine reef tract near Carrie Bow Cay by violent hurricanes, earthquakes, excessive nutrient run-off from the mainland, and water-warming episodes. However, there is hope that conservation measures will be accelerated and strictly enforced to reverse this decline. Clearly, even the most robust sponges will not survive without the reef and mangrove structures that provide them—not to mention the entire reef community—with a home.



KEYNOTES

A framework for a phylogenetic classification of Porifera following the PhyloCode

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Recent advances in molecular and computational biology have led to a burst of new sponge phylogenies. Unfortunately, many of these phylogenies are not translated into classifications thus accentuating the phylogeny / classification gap (Franz 2005) and the creation of 'phantom taxonomies' (Padial & de la Riva 2007). In a group in which the standard ranks are already in use, naming a newly discovered clade requires either the use of an unconventional intermediate rank (e.g., supersubfamily) or the shifting of less or more inclusive clades to lower or higher ranks, thus causing a cascade of name changes. This situation discourages systematists from naming clades until an entire classification is developed. Meanwhile, well-supported clades are left unnamed, and taxonomy falls progressively farther behind knowledge of phylogeny (Cantino & de Queiroz, 2010 http://www.phylocode.org). To avoid this, a phylogenetic classification using phylogenetic definitions, following the principle and rules of the PhyloCode v.4c, is the best solution to keep sponge phylogeny up-to-date and incorporated into sponge taxonomy. For the time being only three works have given phylogenetic definitions of sponge clades: • Manuel et al. (2003): 5 names (all nomen cladi conversum) • Borchiellini et al. (2004): 5 names (all nomen cladi conversum) • Cárdenas et al. (2010): 7 names (6 nomen cladi conversum and 1 nomen cladi novum). However these names are seldom used and the PhyloCode is most often ignored. In an effort to acquaint sponge phylogeneticists and taxonomists with phylogenetic classifications, we here propose to include all PhyloCode names in the World Porifera Database (www.marinespecies.org/porifera/). The WPD has an expert team of editors who could guarantee that the clade names proposed 1) follow the PhyloCode rules and principles and 2) are linked to a publication and to a phylogenetic tree. The purpose of the PhyloCode is not to replace existing names but to provide an alternative system for governing the application of both existing and newly proposed names. We propose that only well supported poriferan clades should be named following Article 9.4. When possible, pre-existing names will be used to avoid the proliferation of names ('conversion' in the sense of the PhyloCode recommendation, 10.1). Furthermore, names will be employed in a sense close to their classical sense in the Linnaean system (recommendation 10A). The species name under the PhyloCode is a binomen (Article 21): the first part of a species binomen is a 'prenomen' and the second part is a specifier or an epithet. The author(s) of a PhyloCode name is (are) the person(s) who establish that name, including publication of a phylogenetic definition for it (Article 19). When cited, these author(s) appear in square brackets, after the person(s) who first published the name (Article 20.2): Calcinea Bidder, 1898 [Manuel et al., 2003], Geodia Lamarck, 1815 [Cárdenas et al., 2010].

Integrating new knowledge of molecules into the physiology and ecology of sponges

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The recent availability of the first sponge genome (Amphimedon queenslandica) and of the transcriptome, and soon the metabolomes, of other sponges gives us a new way of understanding sponges: by their molecular components. This wealth of information confirms some long-held ideas about sponge form and function, but also poses new puzzles. In a way it is the absence of particular genes or gene families from the genome of Amphimedon that is most interesting and which generate questions that highlight the need for an organismal approach. To understand how molecules work in the physiology and organization of the sponge we therefore need so study the cell biology, physiology and ecology of sponges, in the lab and in situ. For example, the genome tells us that sponges have genes for post-synaptic signaling, as well as a complement of potassium and calcium channels. And although we cannot find clear synaptic-like structures between sponge cells, expression work of sponge channels in frog oocytes suggests that some K-channels from Amphimedon are able to re-set the membrane potential quickly, hinting at the ability of this demosponge to be able to send electrical signals. We also now know that sponges have genes for tissue polarity and adhesion, but not for occlusion, yet new physiological studies from our group show that sponge epithelia have high electrical resistance, and those with high resistance occlude the passage of small molecules as do other animal epithelia. And we know that sponges have receptors for GABA and for metabotropic glutamate signaling, but apparently no ionotropic receptors. Our in vitro studies confirm that sponges, like other animals, use glutamate to stimulate and GABA to inhibit contractions of their tissues, and interestingly the rate of signal transmission is slow as would be expected if they use metabatropic rather than ionotropic receptors. Sponges also possess a repertoire of genes involved in control of cell proliferation, in regulation of development, and in self-nonself recognition. In vitro expression work with genes involved in stem cell maintenance confirms that archaeocytes are the main stem cell population and are able to differentiate into many cell types in the sponge. Cell biological studies confirm that choanocytes are pluripotent, differentiating into more choanocytes and into gametes. But field and lab studies also show that in some sponges the turnover rate of choanocytes is greater than that of the most prolific epithelium of a vertebrate gut, and they are shed daily. Therefore the diverse roles of choanocytes in pumping, feeding, and forming gametes implies differential gene expression possibly within a sponge ontogenetically, and in different sponges; but what triggers cells to enter one pathway and not another is as yet unknown. Molecular data is a powerful new tool for interpreting sponge form and function. However given the ancient evolutionary lineage of sponges, and their specialization as suspension feeders, which makes them extremely dependent on the chemistry of their environment, we need a sound understanding of the structure and function of the whole animal in its environment to integrate molecular and functional data – a true organismal approach.

Researching the "Redwood of the Reef": Growth, age, demographics and bleaching of the Caribbean giant barrel sponge, *Xestospongia muta*

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Considering their importance in most benthic marine communities, we understand less about many basic aspects of the biology of sponges than most other animal groups. How old are they? How fast do they grow? Are their populations increasing? What is their recruitment rate? Since 1997, my students and I have studied the demographics of over 600 giant barrel sponges, *Xestospongia muta*, in 12 permanent plots on the coral reefs off Key Largo, Florida, USA. Over a 4.5-year interval, we measured the volume of 104 tagged sponges using digital images to determine growth rates of *X. muta*. Five models were fit to the cubed root of initial and final volume estimates to determine which best described growth. The mean specific growth rate was 0.52 ± 0.65 per year, but sponges grew as fast or slow as 404 or 2% per year. Growth of *X. muta* was best described by the general von Bertalanffy and Tanaka growth curves. The largest sponge within our transects was estimated to be 127 years old, and age extrapolations for photographs of the largest *X. muta* are in excess of 2,300 years, placing this species among the longest-lived animals on earth. From 2000 to 2006, population densities of *X. muta* significantly increased at sites on Conch Reef by a mean of 46% and on Pickles Reef by a mean of 33%. In 2006, densities of *X. muta* on Conch Reef ranged from 0.134 to 0.277 sponges/m2, with the largest size class of sponges constituting 75% of the total sponge volume. Increased population density resulted from a significant increase in the number of sponges in the smallest size class. Recruit survival did not significantly change through time; however, a significant interaction between season and year on recruitment suggests large recruitment pulses are driving population increases. Using a stage-based matrix model, projections indicate that populations of *X. muta* will continue to increase under present conditions, but population growth may be negatively affected by continued mortality of the largest individuals from "sponge orange band" (SOB), a pathogenic syndrome. Like other sponge species, surface tissues of *X. muta* contain cyanobacterial symbionts that give the sponge its brownish coloration. Beginning in the 1990s, two types of bleaching of *X. muta* was observed on Caribbean reefs: non-fatal cyclic bleaching, and fatal bleaching, synonymous with SOB. To determine if sponge bleaching is caused by stress, as in reef-building corals, we quantified expression of the stress indicator gene hsp70 in healthy and bleached sponge tissue and in sponges subjected to temperature and salinity extremes. Heat-shock protein expression was higher for sponges with SOB, but not those undergoing cyclic bleaching. This and other studies suggest that the cyanobacterial symbionts of *X. muta* are commensals, rather than mutualists on which the sponge is dependent.

Sponge-microbe symbioses: recent advances and new directions

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Many sponges are now recognized to host abundant and diverse communities of symbiotic microorganisms. Over the past 4 years, rapid advances have been made by numerous studies that examine the diversity of sponge-microbe symbioses. Initial expectations of a consistent, vertically transmitted microbial community in sponges have been challenged by recent documentation of highly variable, horizontally transmitted communities, as well as vertically transmitted communities that appear unique to specific sponge species. This talk will synthesize recent advances in the ecology of sponge-microbe symbioses, focusing on (1) the diversity of these associations; (2) the specificity (or lack of specificity) of these associations over host phylogeny, geography, and time; (3) horizontal and vertical modes of symbiont transmission; and (4) the positive and negative impacts of microbial symbionts on their host sponges. In particular, experimental data will be reviewed that support a mixed model of symbiont transmission, with portions of a sponge's microbial community consisting of horizontally acquired, potentially transient microbes, and other portions consisting of vertically transmitted, resident microbes. Recent advances that examine the impacts of microbial symbionts on their hosts include approaches that use stable isotope ratios to elucidate the contribution of bacteria to sponge carbon and nitrogen metabolism, in addition to studies of sponge diseases. New avenues of research in sponge microbiology include the use of metagenomic sequencing to determine hypothetical roles of microbial symbionts, coupled with experimental cultivation of the symbionts and verification of metabolic contributions to host sponges. Since many of the most intriguing symbionts of sponges have yet to be cultured, further research in this area is clearly needed.

Application of sponges for production of new medicines

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Aquaculture is thus far the best method to produce bioactive compounds from sponges, although also this technique is not fully developed. To gain more insight in the nutritional needs for growth, we studied the growth rate of Haliclona oculata in its natural environment, Oosterschelde, the Netherlands, and monitored environmental parameters in parallel. A stereo photogrammetry approach was used for measuring growth rates. Stereo pictures were taken and used to measure volumetric changes. To obtain more knowledge about the carbon requirements for growth by sponges, respiration rate and clearance rate was measured in situ in Haliclona oculata and compared to the earlier measured growth rate. We found that only 34% of the particulate carbon pumped through the sponge was used for both respiration and growth. About 10% of the total used carbon was fixed in biomass and over 90% was used for generating energy for growth, maintenance, reproduction and pumping. To improve growth, basic knowledge about how food sources are used by the sponge is needed. To find the exact relation between food retained and food converted to sponge biomass we need to be able to distinguish between feed components and sponge biomass, which means we need biomarkers. The fatty acid (FA) composition of organisms is specific and can therefore be used as biomarkers. We identified and compared fatty acid profiles of sponges in their habitats and the corresponding FAs in the suspended particulate matter (SPM) in the surrounding water. Using a 13C pulse-chase approach metabolic rate can be studied inside organisms. The sponges were fed 13C labeled diatom (Skeletonema costatum) for 8 hours in a closed system and they took up between 75 and 85 % of the diatoms added. At different times whole sponges were sampled for total 13C enrichment, and fatty acid composition and 13C enrichment. The algae fed to the sponges were taken up by the sponges within 8 hrs, first conversion started during the first day and total conversion lasted until at least 3 weeks after feeding. In different studies it was shown that sponges grow slow, but are able to regenerate damaged tissue fast, moreover, it has been found that damaged tissue coincides with higher secondary metabolite production. We have examined the change of carbon metabolic rate of fatty acid synthesis due to mechanical damage of sponge tissue. Filtration and respiration rate in both sponges responded quickly to damage. The rate of respiration was reduced immediately after damage. 6 Hours after damage the filtration rate increased to a level that was higher than the starting value, while the respiration rate returned to the initial value before damage.

Evolution and Phylogeny of Sponges and other Non-Bilaterian Animals

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Deep level metazoan relationships have long been controversial issues. Especially a well resolved and supported phylogeny of non-bilaterian animals is needed to provide a robust framework for reconstructing early metazoan evolution. Expanding molecular datasets and continuing advances in phylogenomic methods are increasingly being used, aiming to unravel these relationships. However, important nodes remain notoriously difficult to resolve. For example, some recent large-scale metazoan phylogenomic analyses – contrary to classical conceptions – found ctenophores to be the earliest branching metazoan taxon and favoured a sister-group relationship between sponges and cnidarians, while other analyses suggest that the Placozoa diverged first or that sponges are a paraphyletic assemblage that share a grade of construction rather than common ancestry. Several new large phylogenomic analyses, based on datasets with different gene- and increased taxon-sampling of non-bilaterians, will be presented. Monophyletic Porifera is always recovered, regardless of outgroup choice and evolutionary model. Most analyses favour a sister-group relationship of Ctenophora+Cnidaria, reviving the 'Coelenterata' concept. However, Placozoa relationships remain unstable as they are highly dependent on taxon sampling, evolutionary model selection and outgroup choice. Additional data from non-bilaterians, among them from deeply diverging lineages within Placozoa, and more critical data analyses will be required to unequivocally resolve the branching order of all non-bilaterian groups. However, results so far allow insights into the evolution of some key traits of the Metazoa, including those of sponges.

Ecological interactions of sponge species

Wulff J

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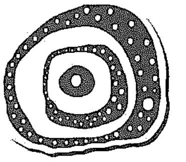
A surge of studies of the ecological interactions of sponges with other organisms and their abiotic environments has bolstered confidence in our general understanding of how sponges fit into their ecosystems. They consume smaller sizes of particulate organic material and, in collaboration with symbiotic microbes, dissolved organics; and they are in turn fed upon by a small number of charismatic animals such as angelfishes, nudibranchs, and hawksbill turtles. Sponges are relatively successful in competition for space, and at asexual propagation and regeneration after partial mortality. Many of their interactions are moderated by chemistry, and they can have profound effects, both positive and negative, on substratum stability and suitability for other organisms. Sponges are especially adept at striking up collaborative associations with organisms of all types. Changing conditions in coastal marine ecosystems are generating questions that reveal some uncertainties about how to predict specific results of ecological interactions of sponges. Concerns have been expressed about both decreases and increases in sponges. For example, if sponge abundance diminished dramatically, would the water column become murky? Could bacteria in sewage and mariculture effluents be controlled by increased sponge abundance? If plankton production rates increase in response to increased water column nutrients will sponges be clogged or grow faster? Will damaged reefs be unable to recover if sponge populations diminish? Will spongivores die, or feed on ascidians or algae, if sponges decrease? If sponge abundance increases, will it be at the expense of other sessile organisms, such as corals; or will it improve water quality and substratum stability? Can sponges perform homeostatic miracles, or will they finally be defeated and vanish, taking with them the enormous number of species with which they have established mutually beneficial associations? Lurking within each of these questions is the challenging additional question: Is sponge diversity as important as sponge abundance, i.e., how much does it matter, and under what circumstances does it matter, exactly which sponge species are involved?



ORAL PRESENTATIONS

Sponge islands in an algal sea: associated fauna of two Mediterranean sponges

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Marine biodiversity in the Mediterranean has received a fraction of the attention from the scientific world compared to the terrestrial habitats of the Mediterranean. With the Mediterranean Sea undergoing rapid alteration due to human mediated influences, it is vital to improve our understanding of this biodiversity to ensure its conservation and management. Here we examined the "hidden" biodiversity provided by two sponges (*Aplysina aerophoba* and *Ircinia variabilis*) to that of the dominant algal community at Cadaqués, Spain. It was hypothesized that *A. aerophoba* and *I. variabilis* would provide a significant island refuge within algal dominated community to numerous associated fauna. A significantly different ($P<0.05$) associated fauna was found within the algal community and the two sponge species. The algal community had a great number of individuals and greater density of associated fauna than both sponge species (Algal community > *I. variabilis* > *A. areophoba*). However, *I. variabilis* had the greatest species richness (*I. variabilis* > Algal community > *A. areophoba*) and diversity of associated fauna (*I. variabilis* > Algal community > *A. areophoba*). It is clear that the habitat provided by sponges and the "hidden diversity" they contain, is a critical and often overlooked component of the marine environment. This "hidden biodiversity" needs to be taken into account when examining the biodiversity of an area.

Freshwater sponges have sealed epithelia which control ion transport

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Sponges are the only metazoans in which the presence of 'true' epithelia is doubted. This is because of a perceived absence of cell junctions, polarity and a basement membrane. Epithelia form protective barriers to regulate secretion and absorption between the mesenchyme and environment. These properties have not been tested in sponges. Here we show sponges have epithelia with high resistance and which occlude the passage of small molecules. Sponges are capable of coordinated behaviour, have glutaminergic signaling, and their pinacocytes are stable over time (1,2). They are said to lack basal lamina but homoscleromorphs have a distinct basement membrane with type IV collagen (3) and demosponges possess spongin short chain collagen (4). Transmission electron microscopy has revealed areas similar to septate junctions or desmosomes between various sponge cells (5,6,7). Furthermore, cell junction genes have been found in sponges including neurexin (*Oscarella carmela* (8)), integrins (*Ophlitaspongia tenuis* (9)), tetraspanin and MAGI (*Suberities domuncula* (10)). These observations suggest that adhesion and sealing may occur between sponge cells. However, the genome of *Amphimedon queenlandica* does not contain occludin or claudin genes, classical proteins associated with sealing tight junctions in vertebrates, and it lacks many septate junction genes (11). To measure the passage of ions across sponge tissue, we grew aggregates and gemmules of *Spongilla lacustris* on permeable Millicell tissue culture wells. Tissue formed a confluent layer with the attributes of a typical sponge (including canals and an osculum) over the permeable membranes in 3-5 days. Resistance was recorded using an electron voltometer (Evom) to impart voltage across the cultures. The recorded transepithelial resistance of *S. lacustris* epithelia was higher or equal to many vertebrate epithelia including proximal kidney tubules. We found that cultures with a high resistance were able to completely occlude small tracer molecules such as PEG-4000, 3H-Inulin, and Ruthenium Red. Cultures had a negative transepithelial potential (TEP) in normal recording media, positive TEP in chloride-free media, and slightly negative TEP in sodium-free media. These results demonstrate that sponge tissues occlude solutes and control ion transport, the physiological attributes of true epithelia. In the absences of classical occluding proteins this implies that genes for solute occlusion developed before the currently known occluding genes. These unknown sponge proteins may also underlie the physiology of all other animals. Finally, these results suggest that sponges have tissue-level rather than cell-level organization which challenges the definition of Eumetazoa.

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Sycon ciliatum as a model system for evolutionary developmental biology studies

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Sponges have long been recognized as a key group for understanding evolution of animal development. Traditionally, study of sponge development have been limited to descriptive embryology studies, which demonstrated amazing diversity in types of cleavage, morphogenetic movements and timing of cell differentiation during development among different sponge groups. Recently, thanks to the sequencing of the first sponge genome, the one of *Amphimedon queenslandica*, and development of protocols to study gene expression during its embryonic development, we began to gain understanding of genes involved in development of this demosponge. In addition, transcriptome resources and molecular methods, mainly aimed at study of adult morphogenesis, have been developed for Homoscleromorph species, allowing us to gain insight into developmental toolkit of this group. However, except for identification of a handful of developmentally important genes, no data on molecular aspects of development are available for the third important group of sponges, the Calcarea. This is somehow of a paradox, given the fact that it was the observation of development of the calcareous sponges that led Ernst Haeckel to propose his famous "gastrea theory" and the term "gastrulation". We would like to propose *Sycon ciliatum* as a developmental biology model system that will fill this important gap among the emerging sponge models. *Sycon ciliatum* has broad distribution through North-East Atlantic, growing in large numbers at shallow depths allowing for easy sampling. It has a long reproductive season - in our area (southern Norway) the larvae are released from June until at least November, and is characterized by a short life cycle (no more than a year). Large specimens can spawn thousands of transparent, simple larvae, which readily metamorphose and develop into juveniles in laboratory conditions. We have developed transcriptome resources and molecular methods allowing to study gene expression during all developmental stages at cell-level resolution, and are now employing these methods to investigate conserved metazoan developmental regulatory genes in *Sycon*. We find that these genes are expressed in patterns suggesting they play similar roles in development of *Sycon* as they do in higher metazoans: A eumetazoan germ line marker, Piwi, is expressed during gametogenesis. Brachyury, which is involved in gastrulation of animals ranging from Cnidarians to vertebrates is expressed during gastrulation-like process occurring in metamorphosis of larvae to juveniles. Transcription factors from the Sox family display dynamic spatio-temporal expression patterns in embryonic and postembryonic development. We are currently developing methods to study gene function, as well as protocols for complete life cycle culture, which will allow to investigate molecular mechanisms governing development in this important group of sponges.

The Halichondrida of northern Australia and phylogenetic implications for the sponge classification

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A taxonomic revision of the Order Halichondrida (Demospongiae) of northern Australia and adjacent areas was undertaken using recent and extensive museum collections that consisted of at least 1280 specimens. Most specimens were collected by SCUBA diving during different biodiversity and bioprospecting surveys over the last three decades. Specimens were initially identified into Operational Taxonomic Units (OTU's) and fully documented with field data, in situ photographs and description of the main taxonomic characteristics. Detailed examination of specimens included in each OTU revealed approximately 142 taxa representing four families (i.e Axinellidae, Dictyonellidae, Halichondriidae, and Heteroxyidae) and 22 genera. Approximately 74 OTUs could not be assigned unambiguously to previously published species and are currently in progress of being described as new species. The number of OTUs initially identified was significantly higher than the number of species represented in the area. The discrepancy was attributed to the high degree of morphological polymorphism observed in many species such as those included in the genera *Axinella*, *Acanthella* and *Stylissa*. High levels of intra-individual polymorphism at the genomic level were also detected in Axinella aruensis suggesting the possibility of species hybridisation within Halichondrida. These extreme levels of both morphological and genetic polymorphism observed within some halichondrid species seriously affects species delimitation and estimates of diversity within the group and complicates further the study of their phylogenetic relationships. It remains to be tested whether these highly polymorphic species include complex of species (either cryptic or hybrids) only detectable using genetic methods. Study of the distribution patterns of the revised species is currently in progress; preliminary results indicate that the group includes a relatively high percentage of 'apparent endemics' (short-range endemism), and that biogeographic patterns are concordant with the bioregions proposed for northern Australia by the Integrated Marine and Coastal Regionalisation of Australia and with the provinces and ecoregions defined by Spalding et al. (2007). Phylogenetic studies based on molecular data from other authors indicate disparate phylogenetic relationships within Halichondrida (e.g. *Cymbastela* and *Acanthella spp*), and between Halichondrida and taxa from other orders (e.g *Agelas*, *Axinella* and *Stylissa spp; Reniochalina*, *Ptilocaulis* and *Axechina spp*), and suggest that the order is non-monophyletic. Morphological results derived from this study are largely not congruent with those enigmatic relationships consistently reproduced through those molecular studies. The monophyly of Halichondrida remains therefore inconclusive and the value of both molecular and morphological characters to resolve phylogenetic relationships continues to be debatable.

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The pathology of Sponge Orange Band disease affecting the Caribbean barrel sponge *Xestospongia muta*

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While beneficial sponge-microbe associations have received much attention in recent years, less effort has been undertaken to investigate the interactions of sponges with potentially pathogenic microorganisms. The aim of this study was to examine a disease condition, termed "Sponge Orange Band", affecting the prominent Caribbean sponge *Xestospongia muta*. Sponge Orange Band disease is characterized by gradual tissue discolouration ranging from healthy brown to bleached white and is sometimes accompanied by an orange band-like transition zone. The disease leads to visual degradation and erosion and can result in the collapse of the entire sponge. Tissue samples were collected from healthy and diseased individuals, the latter of which were distinguished according to surface colouration (brown, orange, white). Scanning and transmission electron microscopy revealed that the loss of colour during disease is accompanied by massive destruction of the pinacoderm leaving only spicules and detritus behind. Chlorophyll a content determined by spectrophotometry and the characteristic *X. muta* secondary metabolites as measured by HPLC were significantly lower in bleached than in healthy tissues. Denaturing gradient gel electrophoresis (DGGE) using cyanobacteria-specific 16S rDNA primers revealed a distinct shift in that the *Synechococcus/Prochlorococcus* clade of sponge symbionts was absent in the bleached tissues while several clades of unspecific cyanobacteria, including lineages associated with coral disease (i.e. *Leptolyngbya*), had invaded. DGGE using eubacteria-specific 16S rDNA primers revealed no evidence for the appearance of what might be considered a bacterial pathogen. Moreover, the eubacterial community profiles of diseased individuals were highly variable unlike those of healthy sponges. Additionally, underwater infection experiments were conducted by transplanting bleached cores into healthy individuals which were then monitored regularly by Scuba diving for up to eleven days. No visual signs of disease developed, rather the surfaces around the tissue implants started to heal up. In conclusion, we have documented that Sponge Orange Band disease in *X. muta* is accompanied by massive tissue destruction as well as loss of the characteristic microbial and chemical signatures. Even though new bacteria were shown to colonize the bleached areas, there is no indication for the involvement of a microbial pathogen as an etiologic agent of disease leaving the cause of Sponge Orange Band disease in *X. muta* to be identified.

Population structure of selected sponge species (Porifera: Demospongiae) from the Indonesian Archipelago

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Indonesia is an archipelago located in the Indo-West Pacific (IWP) centre of marine biodiversity. Its highly diverse coral reefs provide various habitats for marine invertebrates, including sponges. However, the Indonesian sponge fauna is insufficiently studied, which is a major disadvantage for marine area management. Studies on population connectivity from different habitats in the archipelago could provide important information for management efforts, e.g. by identifying genetically isolated areas that warrant special attention. The oceanography of the Indonesian archipelago is dominated by the Indonesian throughflow (ITF) currents, which influence marine larval dispersal among reefs. The current moves up to millions of cubic-meters of water per second from the Pacific to the Indian Ocean. Nevertheless, most sponges are supposed to have restricted larval dispersal, frequently resulting in limited gene flow. We examined genetic structuring between populations of selected sponges species in different habitats using mitochondrial and nuclear DNA-sequence markers. On a small spatial scale we focused on population-variation in the Spermonde Archipelago (SW Sulawesi) and on a large spatial scale we investigated populations from different sites throughout the Indonesian Archipelago. We also compared the genetic results with phenotypic differences, as populations in different habitats might have variations in phenotypes as well as genotypes. Studies of the phenotype included spicule morphometrics and skeletal architecture. Our aim was to contribute to the understanding of dispersal patterns and genetic connectivity among Indo-Pacific sponges.

Prevalence of asexual reproduction in sponges on reef flat and shelf habitats – implications for ecosystem stability in a changing climate

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Sponges characterise substantial proportions of subtidal marine environments, from the tropics to the poles. Even in coral reef ecosystems, sponges dominate habitats below 15 metres depth. These organisms have a wide repertoire of reproductive strategies, with diverse modes of both sexual and asexual activity. Asexual modes of reproduction are common in many benthic invertebrates and appear to be well suited to conditions of extreme (but predictable) habitat disturbance. The large size of propagules confers advantage during settlement and recruitment in disturbed or changing microhabitats, whereas sexually produced progeny are much smaller and therefore vulnerable to predation, overgrowth or smothering by sediments. Within population ranges of sponges, centrally located adults appear to reproduce sexually, possibly due to habitat predictability. At population edges, physical conditions are more unpredictable; asexual reproduction occurs regularly and these propagules fare better. It could therefore be argued that individuals at the edges of populations may have 'older', less diverse genomes than those in central parts of populations in marine benthic ecosystems. This presents a conundrum in a Darwinian sense, as when marine benthic conditions change, such as altered sedimentation regimes, the arguably genetically more diverse central regions of the population may well be less equipped to continue recruiting than asexually produced genomes at population edges. A further element connected to reproductive strategy and evolutionary process in these organisms, is the role of microbial symbionts. The heightened degree of physiological/biochemical flexibility added to the host phenotype by microbial symbionts is considered highly relevant. Asexual propagules will benefit immediately by symbiont inclusion, whereas vertical transmission or acquisition from the rare seawater biosphere is required with sexual reproduction. Selective process is argued therefore, to act on the wider, complete, long term strategy of the metazoan/symbiont collective and the shared genome. A number of climate change response scenarios are discussed to explore the implications of reproductive strategy on the stability of sponge characterised reef communities.

How might climate change and anthropogenic disturbance influence tropical sponge assemblages?

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Coral reef ecosystems across the world are threatened by a range of human-mediated disturbances and the future effects of climate change. Although changes in some of the dominant reef fauna have already been well-documented in response to these threats, particularly for corals and fish, variability in other groups remain poorly described and understood. Given that sponges fulfil a number of important functional roles on coral reefs, changes in their abundance may have significant ecosystem functioning effects. Our work is focused on elucidating the relative roles of biological and physical factors in driving sponge temporal and spatial variability in order to develop a predictive model to understand the future impacts of climate change and anthropogenic disturbance on sponge assemblages. Here we discuss our work on the temporal variability in the sponge fauna of the Wakatobi National Marine Park, SE Sulawesi, Indonesia. We examined changes in sponge assemblages at three different sites over a 6-year period. We found significant changes in the abundance of sponges, and considerable changes in assemblage composition over time, demonstrating that some components of these sponge assemblages are highly dynamic. These changes correlated with declines in fish populations, but not with coral cover, which has remained constant over the study period. It is clear from our research so far, and perhaps not unexpectedly, that the factors driving sponge assemblages are complex and interacting. We discuss our progress towards predicting how sponges may respond to future threats of climate change and human disturbances.

Photosynthetic symbionts of the Mediterranean sponge *Petrosia ficiformis*: characterization and implication in the prevention of fouling

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Marine biofouling is a complex colonisation process of natural or artificial submerged surfaces. The first step corresponds to the biofilm formation (bacteria, microalgae...) then followed by macroalgae and/or invertebrates (barnacles ...) settlement. Marine invertebrates have developed physical and/or chemical defences to keep a low level of settlement on their surface. Some sponge metabolites have been shown to be involved in this natural antifouling process [1] and in some particular cases, active molecules could be produced by heterotrophic (bacteria) or autotrophic (microalgae and cyanobacteria) endosymbionts. The present research is focused on a common Mediterranean sponge (*Petrosia ficiformis*) which is known to host cyanobacteria from a sponge-specific clade, *Candidatus Synechococcus spongiarum*. The sponge surface covering and cyanobacteria density vary with environmental conditions and particularly light intensity. Isolation attempts and characterisation of cyanobacterial symbionts will be first described. In order to assess the implication of these symbionts in the fouling prevention, antiadhesion and antibacterial potential of sponge, with and without cyanobacteria, have been investigated. Freeze-dried extracts have been analyzed by LC/ESI-MSn and fractionated by column chromatography on silica gel. Then, the bioguided isolation of active secondary metabolites has been performed using an antiadhesion assay with marine pioneer bacteria (*Pseudoalteromonas spp.*), 96-multiwell plates and Sytored61 staining. The chemical characterization of active fractions by 1H and 13C NMR spectroscopies revealed the presence of three chemical types: polyacetylenes, steroids and fatty-acids. Four abundant sponge compounds have been purified; three polyacetylenes and a sterol (petrosterol). Effective concentration 50 (EC50) was determined from a sigmoid dose-response curve and showed values in the range between 0.03 and 0.11mg.mL-1 for sponge extracts. Depending on the bacterial strain, purified molecules exhibited high to moderate activities: 9.57 to 191.86μM for polyacetylenes (purified from sponge without cyanobacteria) and 6.50 to 35.15μM for petrosterol (purified from sponge with cyanobacteria). In addition, the sensitivity of the bacterial strains tested varied with the molecules. According to these results, major sterol in *P. ficiformis* with cyanobacteria, petrosterol showed a significant antiadhesion activity. The polyacetylenes, known as sponge metabolites, also exhibited antiadhesion activities. Then, both the sponge and its symbionts could be involved in the fouling chemical defence. A complementary study of molecules extracted from isolated cyanobacteria should confirm these results.

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The effect of a cold water shock on the coral reef sponge fauna of the Florida Keys, USA

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On the 1st of January, 2010, water temperatures along the Florida reef tract began to plummet, reaching a low of 12° C on the 13th of January (8° C below the mean winter lower limit of 20° C) and remaining below 20° C until the 19th or 20th of the month. Florida's reefs lie farther North than most, and though seasonal water temperature change can be considerable (summer mean of 30° C vs. winter mean of 22° C), not since 1977 has an event of this kind disturbed these reefs. Rates of physiological processes are tightly linked to temperature, such that slight fluctuations (e.g., 1° C) can have large effects on the rates of reactions; large fluctuations, like those in January, can rapidly lead to death, as evidenced by the losses of corals, fishes, and marine mammals all along the Florida Keys reef tract during the cold shock. Though the loss of life resulting from this disturbance is concerning and lamentable, such "natural experiments" provide the opportunity to collect a wealth of information which would otherwise be impossible to obtain for both logistical and ethical reasons. Decreased water temperatures are known to negatively influence both adult sponges (e.g., body size, ability to form skeletons, and water pumping activity) as well as sponge larvae (e.g., swimming speed, settle and recruitment ability), but while cold water episodes are known to occur in tropical locales, for instance during periodic cold water upwelling, the effects of dramatically decreased water temperatures have not been reported for sponges for which prior census data are available. Seven months prior to the cold shock, in June 2009, the sponge community within the Looe Key Existing Management Area (Looe EMA), a typical low relief, rocky reef in the middle of Hawks Channel, Florida Keys, was censused using 1 m^2 quadrats (N=17 quadrats). All non-cryptic sponges found within each quadrat were enumerated, mapped, measured (volume), and identified. Within the 17 m^2 area, a total of 1, 215 individuals representing 68 species were indentified, accounting for a 99,808.1 cm^3 of sponge biomass. Species accumulation curves confirmed the community had been adequately sampled. On February 3rd, 2010, roughly two weeks after the end of the cold shock, the sponges at the Looe EMA site were recensused; roving diver surveys at this site, along with visits to sites closer to shore, were also performed at this time. The number of individuals in the post shock census had dropped to 827, a loss of 31%, while total sponge biomass had fallen by 26%, to 73, 410 cm^3. Combined information from the census and the roving diver survey suggest injury and mortality were species specific (e.g., some species suffered losses as high as 90% while others appeared unaffected), and were associated with particular growth forms (e.g., erect branching and tube shaped sponges suffered greater injury and mortality). Sponge mortality and injury were elevated at sites closer to shore, which are buffered less by warm water flowing into the Keys off the Gulf Stream.

The helpfulness of homoscleromorph sponge *Oscarella lobularis* to better understand early evolution of animal development

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The early evolution of animals, that predated the emergence of Bilaterians complexity, is still contentious. This is mainly due to both still debated relationships between non bilaterian animals (Porifera, Placozoa, Cnidaria and Ctenophora) and important lack of genomic and functional data concerning these taxa. Sponges (Porifera) were often shown to branch basally in the metazoan phylogenetic tree. There position and peculiar bodyplan features make their comparison to other animals a promising approach to provide insights into the early evolution of molecular mechanisms controlling morphogenetic processes and its link with the diversification of animal forms. While no obvious consensus was found concerning the monophyly vs paraphyly of Porifera and their exact branching relatively to other non-bilaterian, it is now clear that Homoscleromorpha, previously defined as part of Demospongiae, are a fourth high level sponge taxon, alongside the three classically recognized (Calcarea, Demospongiae and Hexactinellida). Thus they constitute a valuable comparison with the distantly-related demosponge, *Amphimedon queenslandica*, the only sponge which genome is presently sequenced. Interestingly, Homoscleromorpha own characteristics reminiscent to eumetazoan ones, absent from other sponges, noteworthy true epithelial level of tissue organization. This peculiarity may have important consequences concerning cell communication and morphogenetic mechanisms. We thus chose to focus our studies on the evolution of molecular mechanism involved either in the differentiation and patterning of neuro-sensory cells or in epithelial patterning and morphogenesis. In these two cases, two signalling pathways, Notch and Wnt are known to be implicated in Eumetazoa. From a genomic point of view, our studies have shown that these pathways are highly conserved in Demospongiae and Homoscleromorpha sponges but that the gene repertoire of homoscleromorphs is often more diversified. Concerning possible functions in which these molecular tools may be implicated, we suggest that Wnt signalling is a common and perhaps ancient feature of metazoan epithelial patterning and morphogenesis, in addition to a previously proposed role in body axis patterning. And our results can provide new arguments in favor of a potential neurosensory like role of choanocytes.

Molecular taxonomy and phylogeny of the Astrophorida (Porifera, Demospongiae) – an unexpected high level of spicule homoplasy

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The Astrophorida (Demospongiae) encompasses more than 660 species, geographically and bathymetrically widely distributed around the world. The synapomorphy of this order is asters associated with tetractinal megascleres. According to the last revision in the Systema Porifera, five families are included in this order: Ancorinidae, Calthropellidae, Geodiidae, Pachastrellidae and Thrombidae. To date, molecular phylogenetic studies including Astrophorida species are scarce and offer limited sampling. Therefore, phylogenetic relationships within this order are for the most part unknown, hypotheses based on morphology largely untested and the spicule evolutionary processes poorly studied. Using a cytochrome c oxidase subunit I (COI) gene partial sequence and the 5' end terminal part of the 28S rDNA gene (C1-D2 domains) we present the first comprehensive molecular phylogeny of this group. Our sampling included all five families of this order, 29 genera (out of the 38 genera listed in the Systema Porifera) and two sub-genera. We also added to our dataset three 'lithistid' families of Astrophorida affinities as well as two putative Astrophorida (Alectona and Neamphius) still classified today in the Alectonidae (Hadromerida). The COI and 28S (C1-D2) datasets were concatenated in a single matrix containing a total of 152 taxa (89 species) and 1,527 characters. According to our results, the monophyly of the Astrophorida was confirmed and well supported. The Euastrophorida and Streptosclerophorida suborders were both polyphyletic and we propose to formally abandon those names. The Geodiidae, the Ancorinidae and the Pachastrellidae appeared polyphyletic while the Calthropellidae was monophyletic. The Geodiidae was made monophyletic by reallocating Ancorinidae genera to it. Furthermore, a new subfamily of the Geodiidae was revealed, the Caminellinae subfam. nov. The Pachastrellidae genera were distributed in five different clades. Dercitus was reallocated to the Ancorinidae, Characella was incertae sedis along with lithistids, while the rest of the genera clustered in three clades. The Thrombidae and Alectona were the most basal Astrophorida. Some Astrophorida genera were also found to be polyphyletic. Our phylogenetic tree gave us an opportunity to investigate the evolution of Astrophorida sponge spicules, particularly diverse in this order. Our main result was that spicule homoplasy is more common than what we expected: convergent evolution and secondary losses have happened many times, in all the clades, for megascleres and microscleres.

High resolution mapping of community structure in three glass sponge reefs (Porifera, Hexactinellida)

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Glass sponge reefs, unique to the west coast of Canada, are a modern analogue of massive siliceous reefs from the upper Jurassic. Extant reefs represent a living system that allow reef forming patterns and processes to be studied experimentally. Their importance in paleoecology is paralleled by their role in conservation biology as glass sponges are long lived, create habitat complexity for other animals, and are highly susceptible to mobile fishing practices. To date, all reefs have only been studied with remote sensing; a method that does not distinguish between the live, dead and buried sponges. In order to understand the biological patterns of this unique system, we took a small scale sampling approach using stratified grid designs (25m, 12.5m) facilitated by remote operated vehicles (ROV) and high resolution imagery in the field. We created quantified maps of 3 reefs in the Strait of Georgia (Howe, Fraser, and Galiano reefs) which, by using geostatistical analysis and geographic information systems (GIS) allowed us to correlate live sponges with their environment. Glass sponge reefs in the SOG are found on undifferentiated bedrock features elevated from the fine grained depositional sediments that makes up the majority of the seafloor in the SOG basin. In each reef, we found that the glass sponges grow in characteristic patches or mounds 35-72m in diameter. The size of mounds partially reflects the scale at which we surveyed: surveys carried out in a 25m grid show spatial autocorrelation at twice the distance (72m) as surveys carried out at 12.5m (35m). Furthermore, our analysis suggests that were we to survey at an even finer scale, distinct smaller patches would be resolved. The patchiness of the reefs may in part be explained by the distinctive growth pattern of the reef-forming sponges. *Aphrocallistes vastus* forms bushes of oscula 2-5m in diameter and many such bushes form each mound. However, at each reef the entire sponge population occurs within a narrow 30-50m depth range. Therefore, whereas growth and reproduction of individual sponges may structure the mounds, we suggest that reef locations are determined by flow accelerating over the underlying bathymetry. Differences in sponge cover and density of oscula at each reef correlated with the abundance of other animals. The largest abundance of associated animals was found at the reef with the most dense sponge population (Galiano reef), where significantly more crustaceans and fish were found in the presence of glass sponges than in neighbouring areas without sponges. Our high resolution maps together with known pumping and carbon uptake rates of *A. vastus* suggest that 0.4 $Lm^{-2}s^{-1}$ is filtered and 0.96 $gCm^{-2}day^{-1}$ is captured, rates similar to estimates of carbon capture from tropical sponge communities. Our study is the first to quantify small scale biological patterns of glass sponge reefs and establishes a biological baseline that will allow future studies to monitor the sponge populations and determine the key processes involved in reef formation as well as assess the potential impact of trawling fisheries on glass sponge reefs.

Sponges as biomonitors: trace metal accumulation in transplants across a metal gradient

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'Mussel Watch' has been a highly successful North American biomonitoring program. Mussels are not common in SE Australia and their use in this region risks the introduction of pests. We sought to determine the utility of sponges as biomonitors, particularly as modular organisms offer the advantage of identical ramets that serve to reduce intra-specimen variation. We determined levels of trace metals within and between individuals of *Suberites sp.* and *Mycale sp.* - two common estuarine species in the region. We confirmed that within-sponge variation was relatively low for the six trace metals we examined - copper, zinc, lead, cadmium, mercury and selenium. We then transplanted ramets of *Suberites sp.* to 7 locations in a large coastal estuary (Lake Macquarie) with a marked trace metal gradient. Specimens were recovered two months later and all showed growth, in some cases increasing in biomass by more than 200%. Slight increases in copper and zinc were observed from south to north, while lead and cadmium showed substantial increases (>200%) over the same gradient. In contrast, selenium decreased over this gradient, consistent with the large coal-fired power stations at the southern end of this coastal lake. We conclude that sponges show considerable promise as trace metal bioindicators.

How does *Halisarca caerulea* stay slim?

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Evidence is accumulating that some sponges feed on dissolved organic matter (DOM) as their primary food source. DOM can comprise up to 90% of the sponges' daily energy intake, which reveals a peculiar discrepancy. As more than half of their food is assimilated, sponges should double their biomass every three days. Apparently, they do not, since the net increase in biomass in three days is approximately zero. We found that under steady-state conditions, the DOM-feeding sponge *Halisarca caerulea* shows a remarkable and unique proliferation and turnover of their cells, predominantly confined to a single cell population, the choanocytes. The sponge expels its 'old' cells through the process of cell shedding. To our knowledge, these fast cell kinetics (a cell cycle of 5.4 ± 0.4 h was estimated) under steady-state conditions, in combination with high turnover by shedding in the absence of apoptosis, has not been observed previously in any other multicellular organism. A high cell turnover may be a useful strategy for sponges living in oligotrophic conditions, preventing permanent damage to the sponge by environmental stress. Coral reef cavity sponges, such as *H. caerulea*, inhabit low nutrient tropical waters, forcing these organisms to filter large volumes of water and to capture the few nutrients efficiently. H. caerulea maintains its body mass and keeps its food uptake system up to date by constantly renewing its filter system. Interestingly, there are striking similarities between the sponge's absorptive system and the human gastrointestinal tract function and structure. We conclude that studies on cell kinetics and functional morphology provide new and essential information on the growth characteristics of sponges and the regulation of sponge growth in vivo as well as in vitro. The conversion of DOM into expelled cell debris may point to a pivotal role of sponges in the recycling of energy on coral reef ecosystems.

Less spicules, more troubles: Five cryptic species of the *Cliona celata* (Porifera, Demospongiae) complex in South America

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A large number of marine organisms lack proper morphologic characters for clear species description and easy identification. This frequently promotes broad distributional patterns of morphospecies that, in fact, comprise evolutionary independent lineages. In Porifera, the same pattern can be observed, and molecular studies have helped to identify innumerous cryptic species of previously assigned cosmopolitan species. Cliona celata Grant, 1826 is a complex of many species and morphotypes around the world, which includes excavating sponges that contain only tylostyles as megascleres and possess yellow to orange coloration when alive. The present work aimed to verify the taxonomic status of the Cliona celata complex in South America. We used the partial region of the Citochrome Oxidase subunit 1 (CO1) gene of the mtDNA and the Internal Transcribed Spacers (ITS) of the rDNA to estimate the levels of genetic divergence among individuals of C. celata and Cliona chilensis Thiele, 1905 in South America. Additionally, spicules dimensions were quantified to determine if this morphological character was useful for species delimitation. Five distinct clades were found within the C. celata complex with both the CO1 and ITS markers. Based on the large genetic differences found among clades, it is highly likely that each clade encompass distinct species. Two clades comprise the previously assigned C. chilensis, while the other three were formerly recognized as C. celata. Although molecular studies have been demonstrating that the distribution of C. celata is much smaller than previously thought, some species are still widespread. For example, one of the C. celata clades encompasses individuals from Ireland and SE Brasil. Another example is the amphi-South American distribution of one of the C. chilensis clade including individuals from Chile and Argentina, indicating the presence of gene flow between the Atlantic and Pacific oceans via the Patagonian South Cone. Contrary to the molecular results, spicules dimensions were not able to separate the five clades. Although morphometric analysis had statistical support, parameters analyzed were incapable to diagnose clades after post probability tests. Therefore, there is a great need to investigate other morphological characters for species delimitation in the Cliona celata species complex.

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A changing world: The sponge fauna of Ternate (North Moluccas, Indonesia) 1896 vs. 2009

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Scientific museum collections are ideal tools for detecting long-term changes in biodiversity, especially if the specimens are well curated and accompanied by reliable documentation on locality and time of collecting. At present such specimens can be important as reference material for historical studies of marine biota, especially in relation to the mortality and possible local extinction of coral reef species as an effect of the El Niño Southern Oscillation (ENSO) events. For the recognition of global change signals and rational choice of indicator taxa, a solid historical baseline is needed. This may be achieved by the compilation of reliable and validated Indonesian biodiversity data as far back in time as possible. Some Indonesian coral reefs were extensively studied during the past 125 years, especially during expeditions covering major animal and plant groups. One of those collections was done a little bit over a century ago by the German professor Kükenthal who sailed the Moluccas seas and collected about 100 sponges near the island of Ternate. The collection was described by Kieschnick (1896) and revised by Thiele (1900 &1903). These papers are still the most important works of sponge descriptions from Indonesia so far with a total of 51 new species. Some of these species can be found around the coral reefs of the Indonesian archipelago and beyond, while others have never been reported since their descriptions. It is possible, that these sponges only live on the reefs around the island of Ternate as these reefs have never been surveyed again after 1896. However, the city of Ternate has increased dramatically in the past century, for instance the dense mangrove forests have completely disappeared, and it is thus likely that some of the recorded sponges of the 19th century have disappeared due to reef degradation. In 2009 we were able to investigate the reefs around Ternate and this yielded a collection over 300 specimens. In this study we present insights into the sponge composition changes of Ternate and adjacent reefs in the North Moluccas, Indonesia 1896 *vs.* 2009

Seasonal cycles in Adriatic sponges

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Temporal patterns of abundance are recognised to be important in the understanding of the structure of benthic communities in temperate shallow waters. Evidences from the Mediterranean Sea show that in coastal waters, there is a set of species that becomes dormant in summer and another set of species that enters dormancy in winter, with the alternation of two types of floras and faunas. The studies of seasonality of Mediterranean demosponges were until now conduced in the Western basin, while no data are available from Adriatic Sea, a basin characterised by peculiar trophic and hydrologic conditions. Here we show the seasonal trend of abundance, evaluated as covering of the substratum, of two of the most common species of demosponges living on rocky substrates of Conero Promontory (North-western Adriatic Sea): *Chondrosia reniformis* and *Tedania (Tedania) anhelans*. In this area specimens of *C. reniformis* shows size larger than in Western Mediterranean (the largest specimens exceed 1 m2). Little variations of covering around the year were recorded but, during summer, this species shows an extremely high rate of asexual reproduction, via drop-like propagules. In *C. reniformis* oocytes were recorded throughout the entire observation period with a maximal number in August. *T. anhelans* is a typical seasonal species strongly reducing its body size in winter probably forming resting stages to cross the adverse season. During spring time *T. anhelans* progressively grows and starts to develop propagules on its surface; in July the sponge reaches the maximal size and the propagules become branched. In autumn the propagules disappear and the sponge undergoes a process of progressive shrinkage. Larvae occur during summer. The studies conduced in Western Mediterranean indicated that filter feeder organisms undergo a period of high food availability during winter due to the plankton peak, while summer is an unfavourable period of starvation. On the contrary, as already recorded for colonial hydrozoans, the studied Northwestern Adriatic sponges show a maximal of growth rate and metabolic activity in summer and the organisms reach sizes higher than the rest of the Mediterranean. This is probably due to the peculiar hydrological conditions of the Italian coasts of Northern Adriatic where the lower amount of plankton recorded in summer is in the same order of magnitude of the maximal peak of Western Mediterranean. Moreover nanoflagellates, which spread in summer, could represent a further food supply for the Adriatic sponges. On the other hand the winter metabolic decreasing that in *T. anhelans* results in a period of dormancy, is probably related to the water temperature of this area that in the cold season is 5 C° lower than the Western Mediterranean.

Mangrove and coral reef sponges: an untold story about shallow water Porifera in the Caribbean

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The diversity of Porifera associated with mangrove systems in the Caribbean has been increasingly evaluated in the past 15 years. Comprehensive lists of species have been gathered by a handful of taxonomists surveying mangrove islands or lagoons in a few Caribbean sites. Considering that most mangrove ecosystems in the Caribbean are adjacent to coral reef systems, we decided to compare the sponge fauna of these two systems in terms of species richness and taxonomic affinities. A checklist of species from mangrove and coral reef systems at the best-studied sites in the Caribbean was gathered from published, or our own unpublished, data. Validity of species and their classification were checked against the World Porifera Database. The information was gathered in a species-by-country matrix, separately for mangrove ecosystem and for open habitats in coral reef ecosystems, with values of presence/absence. This matrix was analyzed with the biodiversity index Average Taxonomic Distinctness (AvTD) and with cluster analysis (Sørensen's coefficient and taxonomic dissimilarities) to identify similarities among the local faunas within the region. AvTD measures the average degree to which individuals in an assemblage (or list, in this case) are related to each other, based on a taxonomic hierarchy. The analysis was restricted to the sponge fauna from five countries in which we knew that recent comprehensive revisions of Porifera biodiversity in these two ecosystems have been done (Belize, Cuba, Panama, Venezuela, and Jamaica). As a result, 490 valid species belonging to the two sponge classes, Demospongiae (483) and Calcarea (7 species), are listed. Mangrove species richness was 22 for Jamaica, 40 for Cuba, 54 for Twin Cays (Belize), 63 for Bocas del Toro (Panama), 66 for Venezuela, and 123 for the Pelican Cays in Belize. Coral reef open habitats, on the other hand, ranged from 97 in Venezuela, 103 in Belize, 128 in Panama, 129 in Jamaica, and 234 in Cuba. Sponge species richness Caribbean coral reef habitats was double the number of species reported from adjacent mangrove ecosystems (330 vs 160 species respectively). The much higher diversity in coral reef habitats is comprised of 30-60 % more species within each Demospongiae order. Certain Demospongiae orders such as Agelasida, Verongida , Lithistida, Homoscleophorida are basically absent from mangrove ecosystems. Within Demospongiae orders, there are clear divergences among families that radiate within each ecosystem. For example, the Petrosiidae radiates in coral reef open habitats, while the Chalinidae radiates in open mangrove habitats. Overall, significant dissimilarities in sponge faunal composition between mangrove and open reef habitats were consistently found at all five Caribbean localities. Aproximately 60% of open reef species are not shared with the mangrove habitats, and 24 % of mangrove species are not shared with reef habitats. These results give statistical support to the recently proposed hypothesis of the distinct historical origins of these two faunas, with a younger and derived mangrove fauna (<80 my old), and an older (<250 my old) reef fauna. The importance of taxonomy to enlighten the ecological and evolutionary relationships of these faunas is here discussed.

Glass, rocks and clocks: prospects and problems of molecular dating in hexactinellid sponges

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A molecular paleobiological approach integrating paleontology and molecular biology can help in reconstructing the evolutionary history of taxa with fragmentary fossil records or uncertain paleontological classifications, such as Porifera. However, so far this approach has not been applied extensively to any specific sponge group. Glass sponges (Hexactinellida) have been (and still are) important elements of the deep-sea benthos. They have often been involved in reef construction, in both deep and shallower water, through different periods of Earth's history. Although their fossil record, which dates back to the late Precambrian, is relatively rich, fossil information for some groups is sparse, important characters are often not preserved, and relationships between extant and extinct taxa are still poorly understood. Thus, a molecular paleobiological approach is highly desirable for a better understanding of the evolution of this important taxon. Here we use the most extensive molecular dataset assembled so far for Hexactinellida to date their clade divergence times, using fossil calibrations and modern statistical inference methods that do not assume a strict molecular clock. We compare our results to different interpretations of the fossil record, and discuss potential implications for our understanding of hexactinellid evolution. Finally, we discuss some problems with current molecular dating methodology, and point to areas in need of further research.

Climate change impacts on coral reef sponges

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Coral reef ecosystems will be greatly affected by climate change, but the impact of warmer, more acidic water on sponges is unknown. Explants of six coral reef sponges – *Aiolochroia crassa*, *Aplysina cauliformis*, *Aplysina fistularis*, *Ectyoplasia ferox*, *Iotrochota birotulata*, and *Smenospongia conulosa* - were grown for four weeks in tanks with seawater ranging from today's environmental conditions to what is expected by 2100. For each species, final explant size and survival was similar among the various treatments, while attachment to substrate was often quicker in warmer water. For each species, concentrations and compositions of secondary metabolites was examined by NMR spectroscopy to determine how climate change will influence metabolite biosynthesis in sponges.

ALAGerm cells specification in sponges

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The morphological structures named germ cells determinants were revealed in the oocytes of many animals. The preformation mode of germ cells determination is strictly connected with the existence of these organelles. They are localized in the oocyte's cytoplasm and segregated in the certain blastomeres during the embryonal development. These blastomeres descendents will give a lineage of primordial germ cells (PGC). The proteins and the ribonucleic acids which play an important role in post-transcriptional processes are found in these determinants. They provide the maintenance of the totiponency of germ cells. Germ cells determinants have a similar morphology in different groups of multicellular organisms. Their fibrillar material is frequently associated with the mitochondria, the ultrastructure of these formations being reflected in variants of their names - nuage, mitochondrial clouds, dense bodies. Vivid examples of PGC preformation are found in the nematodes, insects and the batrachians. However, there is an epigenetic way of germ cell formation (e.g. Urodeles, Mammalia) when PGC appear for the first time at the later stages of embryogenesis under the specific inductive signals of the neighbouring parts of the embryo. At this mode of germ cell specification the morphological determinants were revealed neither in oocytes, nor in PGC. In sponges, cnidarians and flatworms, morphologically defined PGC appear only in adult animals. Unlike the cnidarians and the flatworms, where the origin of gametes is always connected with the pluripotent i-cells or neoblasts, sponges spermatogonia and possibly oogonia can originate from somatic cells – choanocytes, though the existence of another source of germ cells – totipotent archaeocytes - is not well documented. In view of stated above the fact of detection of typical germ cells cytoplasmic determinants in sponges oocytes seems surprising. We have found such structures in oocytes of fresh-water sponges - *Swartschevskia papyracea* (Haplosclerida, Lubomirskiidae) and *Spongilla lacustris* (Haplosclerida, Spongillidae). At the late stage of oocyte cytoplasmic growth and in the beginning of vitellogenic phase the set of germ cells determinants were found in oocyte cytoplasm. In electron micrographs these organelles appear as electron dense fibro-granular structures surrounded with numerous mitochondria. The ultrastructure of associated mitochondria differs at these stages of oogenesis. In the end of oocyte cytoplasmic growth the mitochondria have dense matrix, at the beginning of the vitellogenesis they become electron-light. According to literary data, the changes in mitochondria structure are due to participation of some mitochondrial components in a formation of germ cells determinants. The presence of "germinal plasma" with specific morphological organelles in the oocytes testifies to maternal factors which can determine the germ cells lineage in sponges. The question arises: what cells are holders of these factors? It is possible, that certain population of the choanocytes and/or archaeocytes maintains totipotency and only this population can perceive an external signal for meiosis transition. The nature of such signal is not known. However, one can assume that the preformation and epigenesis are not alternative but may coexist in sponge germ cell lineage formation.

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Insights into structural biology of marine sponges

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Although in the phylum Porifera the skeleton may be composed of a large variety of minerals, calcium carbonate and siliceous structures very rarely coexist in the same sponge. However, here we represent a unique biocomposite of amorphous silica, crystalline aragonite, and chitin from species of the order Verongida (Demospongiae). The discovery of these nanostructured silica-chitin-aragonite biocomposites as structural scaffolds in verongid sponge skeletons offers also many opportunities for re-examination of the previous views on skeletogenesis and phylogeny of Verongida. Because of their ancient heritage, glass sponges (Hexactinellida) may shed light on fundamental questions such as molecular evolution, unique chemistry and formation of the first skeletal silica-based structures, and the origin of multicellular animals. We have studied anchoring spicules from the glass rope sponge *Hyalonema sieboldi*, which are remarkable for their size, durability, flexibility and optical properties. Using slow-alkali etching of biosilica, we have isolated the organic fraction, which is revealed to be dominated by a hydroxylated fibrillar collagen that contains an unusual [Gly-3Hyp-4Hyp] motif. Amino acid analysis of the collagen isolated from the *H. sieboldi* spicule showed a Pro/Hyp-ratio of 1,33, and approximately a 3:1 ratio of trans-4-Hyp to trans-3-Hyp. These ratios are remarkably consistent with peptide sequence data, with hydroxylation of 33% of those Pro residues in the Xaa position and 100% in the Yaa postion (as 3-Hyp and 4-Hyp respectively) of the [Gly-Xaa-Yaa]n motif. We were able to demonstrate the role of the hydroxylation state of collagen in silica polycondensation. The rate of silica formation was significantly higher in *H. sieboldi* spicular collagen than it was in two samples of collagen which lack significant trans-3-Hyp. We speculate that this motif is predisposed for silica precipitation, and provides a novel template for biosilicification in nature. Our findings suggest that in addition to the previously described silicatein-based biosilicification of sponge spicules, collagen has a key role to play in the formation of the long, flexible, optically pure anchoring spicules of the Hexactinellids. A reconstruction of the evolution of biocalcification as well as of biosilicification with respect to collagen may be a key way to obtain strong evidence of ancient, ancestral programs of biomineralization based on this common template. The bioconstruction of the uniquely large siliceous structures (ten orders of magnitude longer than the spicules of demosponges) was probably enabled by the incorporation of collagen, which can play a role as both a template and provide structural support. This may mean a re-thinking of the role of collagen in the evolution of biomineralization, and almost certainly opens up new strategies for the biomimetic synthesis of silica-based materials.

Morphogenesis in sponges: regeneration as a model

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Sponges branch basally in the metazoan phylogenetic tree and are composed of four distinct lineages. Recent molecular studies propose that Homoscleromorpha are distinct from Demospongiae in which they were traditionally classified and may be regarded as a putative sister group of Eumetazoa. In addition, Homoscleromorpha lineage is notably the only sponge group to possess a basement membrane with collagen IV and specialized cell-junctions, thus possessing true epithelia. In Homoscleromorpha, most morphogenesis processes follow the epithelial type: this is true in the case for embryonic development, metamorphosis, sponge growth, asexual reproduction, and aquiferous system development, and is related to the true epithelial structure of homoscleromorphs. In order to investigate the fine cellular and genetic mechanisms of a morphogenesis it is necessary to have a convenient model. For this purpose, we have combined several microscopy techniques to study mechanisms of regeneration of the ectosome and more precisely, the exopinacoderm and the ostia, in two anatomically similar non-skeletal sponges *Oscarella lobularis* (Homoscleromorpha) and *Halisarca dujardini* (Demospongiae). The unfolding of events during regeneration of both species are similar - there are three main steps: (1) formation of 'the regeneration plug', (2) "the regeneration membrane" development (epithelization of a wound surface), and (3) the end of regeneration - the regenerate presents a functional exopinacoderm with ostia and aquiferous system canals. In spite of this similarity there are deep differences between the two species from a cellular point of view: On the one hand, in *Oscarella lobularis* regeneration passes through morphallaxis, when lost body parts are replaced by the remodeling of the remaining tissue. In this type of regeneration, little or no cellular proliferation takes place during the regeneration process. Moreover, the new exopinacoderm differentiate from both endopinacocytes and exopinacocytes, while new ostia form through invagination process. On the other hand, the regeneration in *Halisarca dujardini* is an epimorphosis that requires active cellular proliferation and dedifferentiation prior to the replacement of the lost body part. This regeneration combines two mechanisms to form the regenerated structure or organ: either a first step of dedifferentiation of cells then followed by redifferentiation, or the intervention of stem cells, archaeocytes that proliferate and migrate to the injured site. Mesenchymal morphogenesis by mesenchymal-epithelial transformations is the main mechanism during *H. dujardini* regeneration. It is of special interest to compare these crucial morphogenetic processes between sponge clades and between them and eumetazoans to understand the origin of fundamental cellular processes during animal diversification.

Horny sponges and their affairs: On the phylogenetic relationships of keratose sponges

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The demosponge orders Dictyoceratida and Dendroceratida comprise keratose sponges, which are (mostly) devoid of their own mineral skeletal elements, but possess a skeleton of organic fibres instead. This paucity of complex mineral skeletal elements makes their unambiguous classification and phylogenetic reconstruction based on morphological features difficult. Here we present the most comprehensive molecular phylogeny to date for the Dendroceratida and Dictyoceratida, and also the Verongida based on several independent mitochondrial and molecular markers. We validate the coherence of all classically (morphologically) recognized orders, families and subfamilies, discuss the significance of keratose morphological and chemotaxonomic characters and suggest the revised definitions for the classification of dendroceratid, dictyoceratid, and verongid higher taxa.

Environmental considerations in sponge farming around tropical coral reefs - impact assessment and environmental management

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Australia's first sponge farm has commenced operation in the pilot-scale, and a second sponge farm is in the planning stages. The regulatory assessment process for these two ventures has created a framework for diligent and science-based evaluation of the potential impacts and environmental management of this new industry within the sensitive reef environments of Torres Strait and the Great Barrier Reef. Potential impacts of commercial sponge densities on water quality have been estimated from hydrology and sponge feeding studies. Possible translocation effects from seed stock selection and movement has been estimated using a population genetics approach. The risk of disease has been reviewed, and seed stock harvest guidelines have been developed based on comprehensive distribution and abundance and wild population dynamics data. The potential for a sponge farm to attract and/or entrap species of special conservation significance, such as hawksbill turtles, has also been assessed through analysis of gut contents of live animals. While the regulatory assessment process is still underway for a proposed sponge farm within the Great Barrier Reef World Heritage Area, relevant authorities have allowed the commencement of sponge farming in Torres Strait using a precautionary risk management approach. This presentation will outline the issues raised by regulatory authorities, and the science-based approach taken to address them, to support the sustainable development of this new industry in Australia.

The sponge fauna of the continental margin of Western Australia

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Western Australia encompasses one third of Australia's coastline and the deepwater habitats off this coastline include canyons and plateaus, seeps, rocky banks, and seamounts. No surveys had previously been conducted on the sponge fauna of southwest Western Australia deeper than diving depths, and limited deep water surveys had been undertaken in northern Western Australia. In 2005 and 2007 the Australian national research vessel Southern Surveyor conducted expeditions to these regions in Western Australia in depths of 100 to 1100 metres. The surveys encompassed a wide range of latitudes from 35.5° to 12.4°S. Specific habitats, including both soft and hard terrains, were identified for sampling from maps generated from multibeam acoustic data. This presentation reports on the sponge diversity and abundance found during these expeditions. The Porifera dominated the invertebrate collections by biomass, comprising 86% of the total catch weight in the southern survey. The largest demosponge was an individual of a species of Biemna which weighed 37 kg. The southern survey yielded 1312 sponge specimens which were identified to 371 operational taxonomic units to species level (OTUs) and the northern survey identified 170 OTUs. Demosponges were most abundant at 100 and 200 metres deep on the shallow outer shelf and were rare at deeper depths on the continental slope. Hexactinellida were found at depths >300 metres and Calcarea at depths <400 metres and mostly at 100-200 metres deep in the south. In the north Hexactinellida were found at shallower depths (>100 metres), and no Calcarea were reported. Combined data from both surveys indicated that the Poecilosclerida were most speciose (113 species) in deepwater in Western Australia followed by the Astrophorida (71) and Haplosclerida (62 species). At family level the Ancorinidae were most speciose with 54 species, followed by the Thorectidae (44) and Irciniidae (34), Phloeodictyidae (27), Axinellidae (25) and Raspailiidae (22 species). We found few species occurred along the length of the coastline sampled (over 23o of latitude), and the majority of species were relatively restricted in their distributions. Seventy four species were common to both the northwest and southwest surveys while 297 species were only found in the southwest and 96 species only in the northwest. A high proportion of singletons (rare species) were reported in the study with many species only been reported from a single station. These studies document the high species richness of the deepwater sponges occurring off Western Australia's continental margin.

Molecular Phylogeny restores the Supra-Generic Subdivision of Homoscleromorph (Dendy, 1905) Sponges

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Homoscleromorpha is the fourth major sponge lineage, recently recognized to be distinct from Demospongiae. It contains <100 described species of exclusively marine sponges that have been traditionally subdivided into 7 genera based on morphological characters. Because some of the morphological features of the homoscleromorphs are shared with eumetazoans and are absent in other sponges, the phylogenetic position of the group has been investigated in several recent studies. However, the phylogenetic relationships within the group remain unexplored by modern methods. Here we describe the first molecular phylogeny of Homoscleromorpha based on nuclear (18S and 28S rDNA) and complete mitochondrial DNA data that focuses on its inter-genera relationships. Our results revealed two robust clades within this group, one containing the spiculate species (genera *Plakina*, *Plakortis*, *Plakinastrella* and *Corticium*) and the other containing aspiculate species (genera *Oscarella* and *Pseudocorticium*), rejecting the close relationships between *Pseudocorticium* and *Corticium*. We also note that the two clades of homoscleromorphs have stable, but distinct mitochondrial genome organizations that differ in gene content and gene order. Among the spiculate species, three main clades are recognized: one containing the *Corticium* species, another grouping *Plakortis* and *Plakinastrella* genera, and the third grouping only two out of four studied species of the *Plakina* genus, revealing the para- or polyphyly of this genus. The validity of these clades is furthermore supported by specific morphological characters, notably the type of spicules. We also investigated more in detailed the Oscarellidae intern phylogeny. Our results show that *Oscarella* species form a paraphyletic assemblage As the result of our studies we thus propose i) to restore the pre-1995 subdivision of Homoscleromorpha into two families: Plakinidae Schulze, 1880 for spiculate species and Oscarellidae Lendenfeld, 1887 for aspiculate species, ii) to discard the genus *Pseudocorticium* and to rename *Pseudocorticium jarrei* in *Oscarella jarrei*. To conclude, Homoscleromorpha will be then composed of 6 genera.

Biosynthetic study of the pyrrole-imidazole alkaloids key member oroidin

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Since their discovery four decades ago, compounds of the pyrrole 2-aminoimidazole alkaloids (P-2-AIs) family have been the source of a large number of chemical and biological studies. They are produced mostly by the Agelasidae and Axinellidae families of marine sponges. They are often characterized by their complex structures, most of them being supposedly derived from the unique intermediate oroidin (1). Some of these secondary metabolites have also been reported to exhibit interesting biological activities which triggered the interest on the biosynthesis and chemical synthesis of this large family of derivatives . Based on the isolation, synthesis or biosynthetic experiments, three hypotheses have been formulated upon the origin of oroidin. In 1999, Kerr and coworkers realized the unique biosynthetic experiment using a cell culture of the sponge *Axinella corrugata* (originally *Teichaxinella morchella*) . They were able to incorporate the two 14C labeled amino acid proline and histidine into stevensine which is closely related to oroidin. Since then, oroidin was most commonly described as the product of condensation between dibromopyrrole carboxylic acid derived from proline and 2-aminoimidazolepropenamine derived from histidine. Few years later, Lindel and coworkers proposed an alternative hypothesis for the 2-aminoimidazole part based on lysine. Taking advantage from isolation and synthetic results, Al-Mourabit and coworkers assumed that oroidin should arise from the condensation of proline and arginine into a diketopiperazine further leading to oroidin after an intramolecular guanidine transfer. Since the origin of the 2-aminoimidazole part of oroidin is still under debate we decided to develop a very sensitive protocol enabling us to study the biosynthetic of oroidin by in vivo experiments using labelled amino acids. The results of our experiments performed on the Mediterranean sponge *Axinella damicornis* gave some important clues towards the understanding of the real biosynthetic pathway for this very important family of marine secondary metabolites.

Matrix-mediated biomineralization in the hypercalcified sponge *Petrobiona massiliana*

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Hypercalcified sponges are the earliest Metazoans engaged in extensive biomineralization as they present the unique characteristic to produce an aragonitic or calcitic massive basal skeleton in addition to their spicular skeleton. These "coralline sponges", which have been the most important reef builders during Paleozoic and early Mesozoic, were believed to be extinct until about thirty years ago, when scuba diving and submersible explorations allowed the discovery of about twenty living species in deep or cryptic habitats. *Petrobiona massiliana* Vacelet & Lévi 1958 is a calcareous sponge producing a massive basal skeleton of high-magnesium calcite. This small species, dwelling in dark submarine caves in the Mediterranean Sea, is a suitable model to comprehend biomineralization processes in calcareous hypercalcified sponges. Transmission (TEM) and Scanning Electron Microcopy (SEM) observations clearly indicate the implication of basopinacocytes in the formation of the underlying basal skeleton. The use of potassium pyroantimonate (precipitating agent of divalent cations) or ruthenium red (dye of acid glycosaminoglycans) in fixation treatments, states further that these wide and flattened cells bordering the soft-tissue are involved, respectively, in the transport/concentration of Ca and Mg ions and in the secretion of an organic matrix of mineralization. After etching, the latter is revealed in Light Microscopy through alcian blue and acridine orange staining and in SEM through cryofracture. Finally, the presence of this organic matrix in superficial and discontinuous early-calcified layers of calcite, also stained with calcein, reveals that the basal skeleton of Petrobiona massiliana is formed by superposition of layers according to a matrix-mediated biomineralization model. The basal skeleton formation, in *P. massiliana*, is thus a highly biologically-controlled process.

Optical Fiber Properties of Antarctic Sponges Spicules

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Biosilica is an intriguing example of natural order and complexity. Siliceous sponge spicules are characterized by a large variety of dimensions and shapes, with an ultrastructure based on silica nanoparticles strictly packaged around an axial filament constituted by a family of proteins called silicateins. This biosynthesis scheme determine the production of a peculiar composite material with remarkable technological properties, like high flexibility and the amazing property to transmit light along its axis. The main consituent of these biological fibers are basically made of the same material of common telecommunication fibers (silica), but, differently from the commercial ones, they are much more resistant to fracture. This is due to their ultrastructural organization into lamellae connected by organic glue in their external region. These highly ordered and unique structures are produced under strictly genetic control in mild environment conditions, drastically different from the industrial process, where high temperatures of manufacture are needed. In this work, we have characterized the optical properties of the spicules of the Antarctic sponge *R. racovitzae* respect to their suggested biological functions. Indeed, given the remarkable transmission properties of biosilica, this material can reveal strategic for the realization of novel fiber-optics systems and for embedment in composites structures based on silicon oxides compounds. Silicoeus spicules used in this work were extracted from a frozen specimen of *Rossella racovitzae* collected during the last Italian Antarctic expedition. In order to assess the performances of the spicules in terms of light transmission, a Tungsten lamp and a He-Ne laser (l=633 nm, maximum power 15 mW) were mainly employed as light source. The results demonstrate that spicules from R. racovitzae are able to transfer both red light and white light. In our experimental conditions it is clearly evident the light trasmission properties of this biosilica structure is remarkably different from the commercial optical fiber. In the sponge case, in fact, light is widely diffused along spicule axis, while in the industrial fibers light is trasported along the cable without appreciable loss of intensity. These data confirm that glass sponges spicules have specific optical characteristics, and the peculiar structure of this composite material is able to determine the different transmittance properties compared to the industrial ones. Moreover, the amazing correspondence of the maximum of transmittance of this natural fiber with the wavelength interval of light irradiance throughout the water column at this depth in the Antarctic seas together to the previously documented presence of diatoms along their axis and their capability to distribute light along the hole spicule body in our opinion suggest a direct action of evolution to select specific biomaterial specialized in light transmission to supply the photosynthetic activity of symbionts.

Altered Secondary Metabolite Production in Aplysina cauliformis in Response to Infection by Aplysina Red Band Syndrome

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Emerging diseases of marine organisms, and sponges in particular, have been reported with increasing frequency in recent years. The health of sponges on coral reefs is important to the overall condition of coral reef communities as sponges perform numerous critical ecological roles. The survival of sponges on reefs is largely due to their production of a plethora of biologically active secondary metabolites that provide protection from a diversity of stressors. Aplysina Red Band Syndrome (ARBS) is a recently described disease affecting common Caribbean rope sponges (*Aplysina spp.*). Although the etiologic agent for this disease has not yet been characterized, it is believed to be a filamentous cyanobacterium, which forms a red band that progresses along the length of the sponge, leaving necrotic tissue behind. ARBS currently affects approximately 10% of *Aplysina cauliformis* on certain Caribbean reefs, and it is not clear whether this patchy distribution is due to mechanisms of pathogen transmission or differential resistance on the part of individual sponges. To investigate whether differences in chemical defenses may explain why some individual sponges are susceptible to ARBS and others are not, we used high performance liquid chromatography to generate chemical profiles from healthy and diseased sponges, and quantified peak areas for 15 individual chemical constituents. Several of these compounds were produced at significantly different concentrations by healthy and diseased sponges. We have identified four compounds previously isolated from *Aplysina spp*. sponges. Two of these compounds (iso-fistularin 3 and aplysamine 1) were produced in significantly higher concentrations by healthy sponges. Two additional compounds (aerothionin and 11-oxoaerothionin) were found only in the diseased sponges. Although concentrations of the feeding deterrent compound iso-fistularin 3 differed between healthy and diseased sponges, extracts from all of the sponges were distasteful to an omnivorous reef fish at natural concentrations. Thus, the observed differences in secondary metabolite production between healthy and diseased sponges do not play a role in feeding deterrence. Instead, differences in chemical constituents may represent antimicrobial chemical defenses that inhibit pathogenesis, inducible defenses in response to pathogenesis, and/or compounds of microbial origin from the pathogen itself or from opportunistic microorganisms.

Effects of parrotfish predation and macroalgae on the growth of bioeroding sponges

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Clionaid sponges, an important bioeroding component in coral reefs, are also known to be strong competitors which are able to overgrow other major benthic organisms such as corals. In the Caribbean, *Cliona tenuis* represents one of the most prominent species. However, the manner in which its populations respond to shifting scenarios in coral reef decline remains a matter of debate. This study aims to examine the major drivers which control the growth of this sponge, and in turn to contribute to the understanding of underlying processes involved in shifting phases in the ecosystem. We evaluated the implications of predation and macroalgal competition as factors of mortality for the *C. tenuis* growth rate, in light of a series of environmental disturbances and the subsequent rise in macroalgal abundance in the Caribbean. The study was conducted at Glover's Reef Atoll Marine Reserve, Belize. Throughout a year of experimental manipulation in situ, growth rates were evaluated as a function of macroalgal (*Lobophora variegata*) competition and fish predation. The experiment was designed to consider presence/absence of the explanatory variables, isolating each factor and their interaction, by means of Predation Exclusion Devices (PEDs) for fish predators and algal transplantation to the surroundings of the sponge. Seventy percent of the sponge perimeter was set in competition with the alga. Predation intensity was recorded using one-hour footage in 10 different individuals. Suggested effects of PEDs on water flow and light availability for the sponge growth were also incorporated into the experimental design. Results showed a significant effect of macroalgae which reduced the sponge size by 81.6% of the sponge growth under control conditions. No predation or interaction effects were detected. Furthermore, the experimental manipulation did not significantly affect the sponge growth when comparing the effects of PEDs on growth. This macroalgae species is proven to be an opportunistic and strong competitor for space, driving the abundance not only of corals, as extensively reported in previous studies, but also for competitive sponges in reefs. In situ observations revealed that parrotfish are the most important macro-predators on this sponge. Among them, *Sparisoma viride* and *Scarus coelestinus* were the species most often observed feeding on the sponge. *S. coelestinus* has an apparently major effect on the sponge, owing to the bite size and intensity per individual (personal observations). However, predation intensity was insignificant (0.5 bites.h-1), therefore linking it to the absence of predation effects on the sponge growth. These observations could be attributable to a previously reported low biomass of S. coelestinus at Glover's Atoll (0.186 g/m2). Following coral cover decline, a previously hypothesized increase in the abundance of Clionaids, as opportunistic competitors, is demonstrated to be dependent on the interaction of processes, such as herbivory (controlling macroalgal abundance), and possibly fishing pressure driving the abundance of predators in coral reefs. However, the latter effects remain to be investigated. These findings clearly establish macroalgae as a factor to be incorporated into models when simulating the effects of coral reef disturbance on sponges.

Unravelling the Hymedesmiidae: aligning molecular and morphological evidence

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The family Hymedesmiidae is very speciose and, particularly in genera such as *Hymedesmia*, the validity of species separated by only small differences in spicule form has been repeatedly questioned. For example several species of the North Atlantic sub-genus *Hymedesmia* (*Stylopus*) are currently synonomised as *Hymedesmia* (*Stylopus*) *coriacea*. The importance of skeleton and presence of chelae as genera characters in this family have also been debated. Using a multi-factor approach combining in situ appearance, traditional morphological taxonomy and sequencing of the D3-D5 region of 28S large ribosomal sub-unit we re-examined north Atlantic species in this family, particularly focusing on *Hymedesmia* (*Stylopus*) species including *Hymedesmia (Stylopus) hibernica*, *Hymedesmia* (*Stylopus*) *primitiva* and specimens which would currently be considered as *H. coriacea*. Small but consistent differences in spicule size and shape and obvious differences in external appearance correlated with the molecular evidence, indicating that several sibling species have currently erroneously been synonymised as *Hymedesmia* (*Stylopus*) *coriacea*. A tree including species of *Hymedesmia*, *Phorbas* and *Hemimycale* showed that the *Hymedesmia* (*Stylopus*) species were not a monophyletic group but were scattered amongst other Hymedesmiidae genera. This molecular evidence supports the theory that separation of genera based on skeletal arrangement is invalid. Furthermore, separation of genera by the presence of chelae and acanthostyles is also erroneous as the molecular evidence suggests these have been lost multiple times. Characters such as ectosomal spicule shape and presence of pore sieves would appear to be significant. Unravelling the mystery of the Hymedesmiidae is likely to require a multi-factoral approach: if we exclude the red herring of skeleton, characters such as surface appearance, previously regarded in some circles as unimportant, are likely to provide important clues.

New mode of sponge displacement in *Amphilectus lobata*

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For a long time the information about sponge motion was insufficient. Presently the known modes of movement are the periodic changes of the body volumes, oscular chimney diameter variations, rearrangement of the choanocyte chambers, 'sliding' of the whole sponge along the substrate due to the pinacoderm activity, asexual reproduction by formation of propagules. All these movements are slow and do not affect essentially entire sponge organisation. Here we describe a new mode of sponge displacement in *Amphilectus lobata* (Bowerbank, 1866) (Demospongia). This sponge is a common epibiont in upper subtidal zone of the White Sea. Sponge movement was studied in laboratory and registered by time-lapse video recording. *Amphilectus lobata* often is found in aggregations of several individuals. Scrupulous survey showed that some of the individuals had partly empty skeleton or incomplete aquiferous system. Usually thin tissue bundles connected such specimens. Under certain conditions certain specimens lose normal structure of adult sponge. Surface pores disappear and the surface becomes smooth. Tissue offshoots appear and stream out of the sponge body in a form of branching 'pseudo-plasmodium'. Each offshoot is attached to the substrate only by its leading edge where cells form adhesive pseudopodia. So the offshoot is stretched between the leading edge and 'maternal' sponge. The structure of leading edge and the whole branching net constantly changed. New branches permanently emerge, submerge and change direction of growth. The offshoots extend for the distance of several centimetres from the maternal sponge with the speed up to 5 mkm/min. The movement and mode of branching correlate with substrate characteristics. The permanent cell streaming in straight and reverse direction occurs inside the offshoots. 'Pseudo-plasmodium' with constantly moving cells stays intact and undamaged over large area. After certain period of stochastic movement the 'pseudo-plasmodium' forms one or several cell condensations where the new sponge differentiation starts. All dispersed cell material concentrates to these points. Shortly after the maternal specimen remains as empty skeleton with several new sponges near it. SEM study shows two distinct cell types in the 'pseudo-plasmodium'. Flattened polygonal cells of pinacoderm cover the bundles from outside. Inner part consists of mass of elongated cells with numerous filopodia. Both macroscleræ and microsclerae are also found inside the offshoots. The spicules move with the speed of cell streaming and can be located close the leading edge. Such mode of *A. lobata* displacement is based on high plasticity of sponge anatomy and ability of cells for quick trans-differentiation and amoeboid movement, and rearrangement of the sponge structure with lose of its entire organisation. The initiating stimuli and governing signals are yet unknown. Possibly the environment factors cause spreading of 'pseudo-plasmodium' that is bound up with the search for the optimal place and sponge asexual reproduction. The mode of displacement on the base of 'coordinated' cooperative cell movement is possible only in the presence of the cell-substrate and cell-cell contacts that are still poorly known for sponges.

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Scales of spatial variation of sponge assemblages associated with mangrove roots in the southern Caribbean

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Fouling assemblages associated with Caribbean red-mangrove roots are very diverse and characteristically dominated by sponges. The species richness (all taxa) in this habitat may exceed 500 for the entire region, the phylum Porifera standing out as the prevailing taxonomic group, with more than 170 species confirmed. Sub-regional inventories, however, show diversities much lower than that, suggesting that mangrove epibenthic assemblages are a highly beta diverse community. Beta diversity can be measured as the variability in species composition among sampling units for a given area and spatial scale. Consequently, this component of diversity is strongly dependent on the scale of observation. For mangrove-root fouling assemblages, various spatial scales of ecological isolation can be identified: a few meters (comparing roots within a cay or continuous forest), hundreds of meters (several cays or forests within a region), and hundreds of kilometers (geographically separated regions). Thus, we evaluated the composition of sponge assemblages associated with mangrove roots using a hierarchical nested design in order to identify the patterns of spatial variability in species composition at different scales. Data were used from two previous studies with different hypotheses but in which spatial scales were appropriately replicated. The surveys were done in mangroves of Belize, Panamá (Bastimentos Marine Park), and two regions in Venezuela (La Restinga National Park and Morrocoy National Park), all separated by hundreds of kilometers. Within these regions, at least three localities, separated by hundreds of meters, were evaluated; and at each locality the presence of sponges on 14-20 roots was registered. In total, 435 roots were evaluated. The data were analysed using univariate and multivariate techniques. Patchiness in species composition was detected at all spatial scales. In both studies, the most important source of variability was the smallest spatial scale (differences between neighbouring roots), suggesting that small-scale processes (e.g. settlement behaviour, competition for space, timing of arrival) could be the main cause that maintain beta diversity in the region. Besides, local and regional communities are not equivalent, both spatial scales explained similar amounts of variation, indicating that medium-scale processes (e.g., dispersal ability of larvae, local environmental gradients, predation) and wider-scale processes (e.g., oceanographic and coastal characteristics of each region) are equally important in preserving beta diversity. These results confirm the perception that mangrove epibenthic assemblages are a highly beta diverse community. Future initiatives in quantifying biodiversity of Caribbean mangrove sponges, or entire mangrove root assemblages, should consider evaluation of different spatial scales if generalization is the main goal. Manipulative experiments are necessary to improve our knowledge of the ecology of this important Caribbean biological system, especially those relating to life histories of species.

Searching for new bioactive substances from south China sea marine sponges

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South China Sea is in the tropical zone where marine biodiversity is abundant. Searching for marine natural products produced by South China Sea marine sponges has been carried out by our group since 2000. These years our continuous efforts on the South China Sea Porifera with the purpose to find new leads for drug discovery has led to isolation and characterization of numerous marine natural products with diverse chemical structures, extraordinary biological and pharmaceutical activities. From the South China Sea sponges *Axinyssa aff. variabilis* and *Lipastrotethya ana*, three unprecedented diastereoisomeric dimers, axinyssenes exhibiting a *bis*-bisabolene skeleton, and a new sesquiterpene, dehydrotheonelline have been isolated. Chemical investigations of marine sponge *Dysidea* sp. have resulted in the identification of a truly remarkable collection of sesquiterpene metabolites. One of these metabolites has an unprecedented skeleton, which may come from rearrangement of drimane skeleton. Another two showed cytotoxic activities against A-549, HL-60, BEL-7402 and P-388 cell lines. A new sesquiterpene quinone, 21-dehydroxybolinaquinone, together with two known related analogues, bolinaquinone and dysidine, had been isolated from the Hainan sponge *Dysidea villosa*. The structure of the new compound was elucidated on the basis of detailed analysis of spectroscopic data and by comparison with related model compounds. Compounds were evaluated for the inhibitory activity against hPTP1B, a potential drug target for treatment of type-II diabetes and obesity, and cytotoxic activity against Hela cell line. The results showed that dysidine had the strongest hPTP1B inhibitory activity with an IC_{50} value of 6.70 μM and had significant cytotoxic activity against Hela cell line with an IC_{50} value of 5.45 μM. New compound showed moderate PTP1B inhibitory activity and cytotoxicity with IC_{50} values of 39.50 μM and 19.45 μM, respectively. In this communication the structures and biological activities of these marine metabolites, which might be useful as biomedical agents, will be described.

When biotas split & meet: Sponge distribution in the crossroads of the Atlantic, Austral and Pacific Oceans

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It has been known for a long time that biogeography in the Southern Hemisphere is no negative image of what happens in the northern part of the globe, essentially because of a rather unbalanced distribution of land masses, but also because the rather common decrease in species diversity towards the poles is not so obvious in the south. What sponges have to say in this respect? Species lists have been published for the Antarctic, as well as for the Argentinean and Chilean coasts. These were updated and reorganized with recently generated taxonomic inventories in all three areas, so that comprehensive assessments for particular Marine Ecoregions (South America) and longitudinal slices (Antarctica) could be used as units in an analysis of faunistic connectivity on the basis of putatively shared sponges. SW Atlantic Ecoregions chosen for the analysis were the Uruguay-Buenos Aires Shelf (+ slope, UBASS, with 94 spp.), North Patagonian Gulfs (NPG, 36), Patagonian Shelf (+ slope, PSS, 75) and Malvinas/Falklands (+ slope, MFSS, 88). The SE Pacific ones were Araucanian (ARAU, 48), Chiloense (CHILO, 48) and Channels and Fjords of Southern Chile (CFSC, 108). Analytical units for the Antarctic were slices 8 (C8, 120) and 9 (C9, 106) in Sará et al.'s assessment of biogeographic traits in Antarctic sponges. These analytical units were compared two by two, and both the absolute number and the percentage of shared species were calculated. It appears that an inverted gradient of higher species richness towards the south is there for sponges, perhaps explained by increasing habitat complexity. SW Atlantic ecoregions shared among themselves, a minimum of 18% (NPF vs. UBASS, NPF vs. MFSS) and a maximum of 26% (UBASS vs. MFSS) of their sponges. In the SE Pacific, connectivity ranged between 38% (CFSC vs. ARAU, CFSC vs. CHILO) and 61% (ARAU vs. CHILO). Antarctic areas shared 38% of their records. Atlantic-Pacific connectivity ranged from 11% (UBASS vs. CHILO, NPG vs. ARAU) to 23% (MFSS vs. CFSC). Antarctic-Atlantic connectivity varied from 3% (C9 vs. NPG) to 14% (C8 vs. MFSS). Antarctic-Pacific connectivity also had a minimum of 3% species shared (C9 vs. ARAU) but the maximum reached only 12% (C8 vs. CFSC). It is worth noting that 54% of all species considered (195/359) occur in a single ecoregion, and only 24% occur in three or more ecoregions (85/359), thus showing good capacity to disperse. Up to 36 species are shared between Atlantic and Pacific Ecoregions, and up to 26 between Antarctic and South American Ecoregions. In total, 46 spp are distributed across the Drake Passage. It will be interesting to investigate whether this is more probably the result of recent range expansion, or relictual Gondwanan affinity. The scanty phylogenetic evidence available for sponges does not recognize yet any deep link between Antarctic and South American species (Cathria procera spp. group, Rhabderemia).

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The aquiferous system of Tethya wilhelma: microanatomy and 3D-topology

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The aquiferous system of the Porifera represents the interface between the sponge and its environment. It is involved in ingestion, excretion, gas exchange, etc. Our knowledge includes general aspects of canal architecture, cellular anatomy and limited models on the flow generated by choanocytes. Some studies indicate complex topologies and flow regimes, but we lack complete 3D reconstructions. Our study aimed at analyzing the canal system anatomy of *Tethya wilhelma* (Demonspongiae, Hadromerida) and at reconstructing the 3D topology. We investigated the cellular components of the aquiferous system by applying a combined SEM- and DIC-microscopy technique, in order to image anatomical and histological details within single specimens. The topology of the canal system has been studied by synchrotron radiation based x-ray micro tomography scans (SR-µCT at GKSS/DESY, Hamburg) of canal system corrosion casts and virtual casts. Entire canal systems were generated from high resolution 3D reconstructions using ImageJ and VG StudioMax. Hierarchical classification of canals and additional morphometric parameters were calculated using CoralSoft software. Tethya wilhelma displays the usual partition into incurrent and excurrent system with inserted choanocyte chambers. The apopinacocytes display short stereocilia. In addition, the apopyle region of the eurypylous choanocyte chambers is unique with two cell types involved: 1. stereocilia-bearing cone cells; 2.the unique apopylar cells, which form a mesh-like grid overspaning the whole apopyle opening, with numerous transcellular pores which can be independently closed or opend. We asume a flow regulative function of this new cell type. The 3D topologies based on micro tomography image stacks were analyzed quantitatively and provided information on canal diameter, length, branching angle and spacing between branches in correlation to a hierarchical classification of canals. Due to the presence of larger bypass elements which interconnect the incurrent and excurrent system avoiding choanocyte chambers and therefore forming ring structures in the canals system, hierarchical classification of canals were applied only to a subsets of the data. Between different hierarchical levels the canal diameter scaled by varying factors between 1.1 – 1.5. Canals included in the analysis ranged from 12 µm to 95 µm in diameter with the latter ones being less frequent. Our results on the microanatomy of the canal system, especially the new type of apopylar cell raises questions on the regulation of flow within *T. wilhelma*. We assume that localized flow modifications by the sponge occur. This is congruent with our 3D-analysis, which demonstrated alternative flow-pathways on a more global scale. Our high resolution 3D models of partial and entire canal systems are an important step towards the first computational fluid dynamics models, which will allow us a deeper insight into the anatomical basis of sponge behaviour and ecology.

Evolution of gene regulatory networks: Early construction of the Pax/Six regulatory hierarchy in sponges

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The evolution of complex body plans from more simple forms has lead to an array of organismal diversity. Modern genomics, phylogenetics, and new developmental biology techniques now make it possible to study evolutionary routes from the simple to the complex. One promising avenue of research is to study the evolution of gene-regulatory networks and their developmental functions. For example, many animals use the Pax/Six/Eya/Dac (PSED) network to specify eyes and other sensory organs. Complex interactions between members of these four gene families, integration of other gene-families into the network, and absence of specific PSED interactions from several developmental contexts make the evolutionary history of this network extremely difficult to trace. To begin to understand this evolutionary history, we examined PSED members in an animal with a relatively simple body plan, the freshwater sponge *Ephydatia muelleri*. While sponges lack sensory organs in the traditional sense, they do have two components of the PSED network: Pax and Six. We have profiled expression of both Pax and Six across developmental stages and used in situ hybridization to localize transcripts in regions around choanocyte chambers and the canal system. We used a newly developed RNAi technique for the freshwater sponge to knock down Pax and Six expression which suggests possible roles for these evolutionarily conserved transcription factors. Additionally, we determined that when Pax expression is knocked-down, Six expression is also reduced. This data, combined with gel-shift data using with a Pax protein construct indicates that Pax regulates Six expression in this sponge. Further, we have used chromatin-binding data to elucidate other targets of Pax in sponges. This work helps further our understanding of the origins of a ubiquitous metazoan gene-regulatory network.

Clionaids provide key insights into ecological and evolutionary forces shaping zooxanthella-symbioses in invertebrates

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We are interested in better understanding host:symbiont relationships in marine sponges that associate with zooxanthella dinoflagellates. While recent work has helped clarify the nature of the association, we still know little of integration between host and symbiont, the ecological consequences of that integration, nor the stability of these associations under environmentally stressful conditions. We have been studying the relationship between *Cliona varians* and its Clade G zooxanthellae. Here we present data on photophysiological aspects of the association, physiological integration of the partners, and genetic regulatory pathways utilized during acquisition and maintenance of symbionts. We contextualize these findings in light of current increases in sea surface temperature, and highlight how sponges provide essential and novel ways to think about zooxanthella symbioses. Our surveys of the diversity of zooxanthellae harbored by sponges, as well as the surveys of other sponge biologists, indicate that sponges form symbioses with a broad diversity of zooxanthellae. We will present detailed data on the diurnal changes in photophysiological performance of the zooxanthellae found in *C. varians*, and the consequences of the photophysiology for host performance. We will also discuss data on the integration of the host and symbiont in terms of materials transmitted between partners, and the evolutionary consequences of these physiological linkages. Finally, we will present data on the genetic regulatory pathways that are likely involved in the sponge:zooxanthella symbiosis. Given that zooxanthellae perform vital physiological functions for several sponge species, there is growing need to understand these symbioses given projected increases in sea surface temperatures.

Threats to the system ? Beyond the 'Systema Porifera'

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'Systema Porifera' (SP) established the beginnings of an accessible sponge systematics using the rank-based Linnaean nomenclature as defined by the ICZN. The rules and recommendations of the ICZN apply only to the taxonomic nomenclature of animals, unrelated to any particular biological classification. As such it does not impinge on the rights of scientists to develop new ways of making taxonomic decisions, ensuring the nomenclature is independent of the systematics used, thus providing stability to nomenclature even if the classifications might change significantly. The classification presented in the SP was based on the then-current (2002) understanding of presumed evolutionary relationships between taxa, which even 8 years later has changed substantially for some groups based especially on the growing availability of substantial new molecular datasets, both in numbers of species extracted and the genes sequenced. While it is absolutely certain that this classification will evolve and progress further, the nomenclature will remain stable, and changes in the future will be transparent through a trail of synonymies. Threatening this stability is the (possible) implementation of the PhyloCode in 2010, based on a different systematics paradigm with nomenclature rigidly fixed to a strict set of phylogenetic rules in which every clade is defined by a statement concerning its perceived ancestry and descent. PhyloCode clades do not have mandatory ranks or any necessity to be named (although this is allegedly optional). Similarly, while the escalating availability and use of DNA technologies have been a boon to taxonomy in terms of its improved profile, its relevance and even the continued economic and political survival of the taxonomic agencies themselves, the data they have produced have not always integrated well into a rank-based classification system. There are instances where these data have produced well-supported clades, yet these remain undefined and/or as yet unsubstantiated by corroboratory phenotypic evidence. Whilst the latter aspect is most likely to be contended or resolved retrospectively, by incorporating other genes into analyses and/or by revisiting morphological homologies, the resolution of naming and incorporating the well-supported unnamed clades into the rank-based classification is not yet settled. The rapid emergence of online world biodiversity databases (such as OBIS, GBIF), are set to revolutionise the accessibility of taxonomic data for a multitude of society's purposes, with several nomenclatural lexicons that conform to ICZN (e.g. WoRMS, AFD, EOL, Catalogue of Life), poised to give biological meaning and perspective to the otherwise bland GIS-based specimen point data. These nomenclatural lexicons are by nature hierarchical, based on ranks, and thus the success of these online initiatives firmly rests with the rank-based nomenclature. Because the rules of the ICZN apply only to family level taxa and below, with higher taxonomic nomenclature treated by a much more limited set of provisions, we offer here a reconciliation to deal with these clades that will not jeopardise what the SP has painstakingly strived to achieve: to provide nomenclatural stability within a potentially rapidly changing evolutionary classification.

Novel nuclear coding genes for sponge phylogeny

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Very few molecular markers have been employed to solve sponge evolutionary relationships at the family level. Ribosomal DNA markers (18S and 28S) and the COI gene, the barcoding marker, are most commonly used. Inherent problems with the mitochondrial genome (in particular saturation and base composition issues), and ribosomal genes (in particular lack of variability) have prompted molecular systematicians working on other taxonomic groups to employ nuclear coding genes as phylogenetic markers. Because RNA is not often preserved in museum specimens, sequencing genomic DNA rather than cDNA is desirable. Within genomic DNA long exons are choice markers since introns and intergenic regions are fast evolving and difficult to align. Most markers developed until now, such as the elongation factor-1, were often too conserved to solve evolutionary relationships among sponges that are slowly evolving. We here present three new variable nuclear markers (ALG11, SO and DYRK3) that were successfully amplified in all four sponge classes (Calcarea, Demospongiae, Hexactinellida, and Homoscleromorpha). The protein alignment of these three genes is 728 amino-acid long and the combination of the three markers provides a phylogenetic resolution that is similar to the results obtained with the 18S rRNA gene but which contradicts that obtained using the COI gene marker. The novel markers hold great potential in resolving intra-class relationships of sponges. These three genes complement and corroborate the phylogenetic inferences observed with the nuclear ribosomal genes. Given the incongruent signal observed between COI and the 18S rRNA and ALG11, our results suggest that the inclusion of the mitochondrial COI in multi-gene phylogenies should be examined carefully. Finally, our markers might also help to solve evolutionary relationships of other slow evolving animal phyla such as cnidarians.

Floating dispersal through asexual reproduction of psammobiontic sponge in coral reefs of the Ryukyu Islands, southern Japan

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A psammobiontic sponge, *Spheciospongia inconstans* (Dendy, 1887) (Demospongiae, Hadromerida, Clionaidae), burrowing their body in sandy sediments, distributes in shallow lagoon of tropical and subtropical Indo-Pacific. Observation of asexual reproduction of this sponge in the Ryukyu Islands, southern Japan, revealed unexpected dispersal strategy for this benthic animal. Asexual reproduction (bud formation) proceeded as follows; Phase 1: bud formed as small tip and elongated on upper surface of the sponge. Phase 2: expansion and maturation of the bud. Phase 3: probable apoptosis in the connected region between the bud and the sponge. Phase 4: bud detached from body of the sponge, float and drift. Lots of stranded buds were observed at the beach of the Ryukyu Islands on late summer, which were unsuccessfully settled buds. And formation of an exhalent opening was observed on the detached part of successfully settled buds Floating mechanism is possibly explained by entrapment of bubbles produced by photosynthetic activity of symbiotic zooxanthellae. Bubbles were effectively trapped in pits and thin sheath of buds. Such architecture was also found from voucher specimens labeled as different species deposited in Natural History Museum London. Floatation and drifting of buds may contribute to the wide distribution of S. inconstans in shallow waters of the Indo-Pacific although the drifting period is unknown. According to observation of type specimens, mysterious subglobular sponge *Spheciospongia globularis* (Dendy, 1922) is considered as just a bud of *S. inconstans*, and thus junior synonym of the species. This is the first report of floating buds from sponge.

Bioactivity and secondary metabolite variation in relation to biotic and abiotic factors: chemical ecology study of a new Mediterranean Oscarella species (Porifera, Homoscleromorpha)

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The genus *Oscarella* belongs to a small and challenging sponge clade, Homoscleromorpha, whose phylogeny is still debated and which has been little studied in terms of chemistry. Identification at species level is often troublesome in the highly diversified *Oscarella* genus (14 species listed in the World Porifera Database) which lacks a skeleton, a fundamental character of sponge taxonomy. A new Mediterranean species of *Oscarella*, currently being described, grows mainly in semi-dark caves and coralligenous communities where it co-occurs with two other common species, *Oscarella tuberculata* and *Oscarella lobularis*. Up to now, it has been reported along the coast of Provence and Corsica in France, in the south of Spain and Croatia. In general, *Oscarella* species can be a dominant component of communities and may constitute a particular facies. Therefore, they seem to be good competitors for space. *Oscarella sp. nov.* has often been observed to colonize other massive sponge species as well as gorgonians and bryozoans. This capacity to overgrow different biological substrates could be inferred by an efficient secondary metabolism and the chemical defenses it confers. *Oscarella sp. nov.* was identified and described using morphological, cytological, chemical (metabolic fingerprint) and genetic (COI marker) characters. Growth patterns detected by the analysis of in situ pictures taken during a photogrammetric monitoring (2002-2007) reveal a highly dynamic species. The life cycle and the natural fluctuation of two metabolic indicators with a high energetic cost, reproductive effort and bioactivity, were studied on a subsequent temporal series of samples (2007-2009). *Oscarella sp. nov.* is gonochoric and ovoviviparous, having a seasonal life cycle with an annual reproduction peak during spring (April-June). Significant variation of crude extract bioactivity was observed in time and we attempted to explain it by correlations with other factors such as reproductive effort, substrate type, season, water temperature and secondary metabolite expression. A significant decline in bioactivity and therefore chemical defense capacity was observed during the period of embryogenesis and larval development. A negative correlation between bioactivity and water temperature could also be evidenced. Bioactivity variation was not observed to vary depending on substrate type. The metabolic fingerprint analysis revealed variation in the expression level of several major metabolites, but no particular metabolic phenotype pattern could be associated to the individuals displaying high bioactivity or high reproductive effort. It seems that optimization of resource allocation, clearly observed in the peak of the reproductive season and probably influenced by a combination of interacting biotic and abiotic factors, shaped the temporal trends observed in bioactivity variation.

Diversity and affinities of the sponge fauna in the deep Southern Ocean (SO): New findings from the Antarctic ANDEEP-SYSTCO, ANT XXIII/8 Expeditions (2002-2008)

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Introduction: As part of our Antarctic Porifera project (DFG-project ANTPOR, with WG Wörheide, Munic Univ., JA 1063/14-1, WO 896/9-1), the sponge collections from several recent Antarctic expeditions are described and analyzed. We intend to obtain a better understanding of the zoogeographic-phylogenetic affinities and evolutionary history of Porifera in the deep SO. Here we present some of the new, mostly still preliminary results from the ANDEEP I-III and SYSTCO campaigns (2002-2008). Methods: Sponges were sampled by various benthic sampling gears, especially Agassiz trawl, epibenthic sledge and Rauschert-dredge. Taxonomic analysis was performed by skeletal and histological preparations study in LM and SEM. Ecologic analysis was done by means of several computer programs including MS excel and Past. Results & Discussion: From the deep Weddell Sea and surrounding areas, we so far identified more than 110 sponge species, of which at least 22 are new to science. They belong to the Porifera classes: Demospongiae, 77 spp. (12 new), Hexactinellida, 26 spp. (7 new) and Calcarea, 7 spp. (3 new). Most of the Hexactinellida in the ANDEEP-SYSTCO collections belong to the family Rossellidae. On the Antarctic shelf and slope, the circum Antarctic genus *Rossella* is dominant, whereas at abyssal depths it is replaced by species of *Bathydorus* and *Caulophacus*. Another diverse group in the deep Southern Ocean is the family Euplectellidae. Contrary to the largely endemic shelf fauna, the Antarctic deep-sea Hexactinellida comprise several cosmopolitan species, and show affinities to the deep Atlantic fauna. However, in the case of *Lonchiphora sp. nov.*, collected at 2180 m in the Weddell Sea, we find the closest relative to be a poorly documented species from the Pacific Sagami Bay (Abstract Göcke & Janussen, this volume). Within the ANDEEP-SYSTCO sponge collection, many of the our findings of Cladorhizidae (carnivore sponges) are new to science and most of them new to the SO. The same is true for the first deep-sea Calcarea discovered in the Antarctic Ocean at bathyal and abyssal depths. Cluster-analysis of the sponges sampled during SYSTCO-expedition exhibit some distinct tendencies: The deepest station (5000 m) shows no species overlap with others, whereas the Stations between 500 and 3000 m show some similarities. The composition of the Antarctic sponge fauna changes successively with increasing depth, but keeps cohesive similarities down to ca. 3000 m. In the depth zone of about 4000-5000 m, a distinct abyssal fauna is found. Zoogeographic links and different regional affinities exist, depending on the sponge taxa investigated. These preliminary results need to be tested by comparative morphological and molecular methods. Further detailed analysis of the entire sponge faunas from the ANDEEP-SYSTCO Expeditions will be a major part of our upcoming work.

Acknowledgements: The authors are obliged to the scientific teams of the ANDEEP-SYSTCO expeditions, especially to the initiator and scientific leader Angelika Brandt (Hamburg). Thanks are due also to the crew and captain of RV "Polarstern", and to the expedition leaders. We further thank the scientific sponge community for discussions and collaboration. DFG is acknowledged for financing this research project (JA 1063/14-1).

Spatio-temporal modelling of spicule growth in sponges

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In sponges there exists an enormeous variety in spicule morphologies. Recently new insights have become available in spiculogenesis. In the formation of spicules several proteins (e.g. galactin, silicatein) are involved. In the center of the spicule there is an axial filament that consists predominantly of silicatein, which serves as a point of nucleation in spiculogenesis. This enzyme catalyzes the synthesis of biosilica. The galectin forms aggregates to which silicatein molecules bind. The silicatein-mediated silica formation is strongly increased by the association with the galectin. Furthermore the growth of spicules is limited by the local availabilty of silicic acid. In the growth process of the spicule typically a concentric pattern of growth layers emerges in which layers of silica are deposited alternatingly with layers of organic material. Here we aim to develop a model of the growth process of a spicule and the absorption of local silicic acid. Silicic acid is the main nutrient for the spicule formation in a marine sponge which has three chemical species in seawater (H4SiO4,H3SiO4 -,H2SiO4 2-). The only way for transporting silicic acid from water to the boundary of the sponge cells is by diffusion, while in the uptake of silicic acid by the cell a transporter is involved. Due to the uptake of silicic acid by sponges, the chemical system (including silicic acid species, H+ and OH-) will become out of equilibrium. We model this as a reaction-diffusion system using a system of partial differential equations in order to calculate the concentrations of chemical components as a function of the distance from the center of the cell. We solve this system in three dimensions and as a boundary value problem. This model also provides the uptake rate on he surface of the cell. The amount of absorbed silicic acid species can be used in a growth model of spiculogenesis. We couple a spatial model of spicule growth and a model describing the concentrations of the proteins involved in spicule formation, with a model of the transporter of the uptake of silicic acid from the environment over the cell membrane. The growth of the spicule is simulated using an accretive model, where local growth velocities are controlled by the local concentraties of simulated chemical species.

Diversity of mitochondrial gene arrangement in hexactinellid sponges

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Within the phylum Porifera, our understanding of phylogenetic relationships at class, order, and family levels has shifted frequently in the past. Depending on whether morphological or molecular data were used and which characters were compared while creating a phylogeny, the three classes—Calcarea, Demospongiae, and Hexactinellida—have shifted both in their relationships to each other and to other basal metazoan phyla. Morphological phylogenies hypothesize that the three classes within Porifera are monophyletic, while molecular phylogenies support paraphyly, with on class more closely related to the rest of the Eumetazoa than to the other two sponge classes. A correct understanding of higher-level systematics of sponges is essential for making hypotheses about the evolution of early metazoans. In addition, molecular phylogenies may also resolve persistent discrepancies in morphological studies and contribute to an accurate phylogeny for sponges. Unfortunately, conventional methods for producing molecular phylogenies, such as using mitochondrial gene sequences or ribosomal RNA regions, fail to provide the resolution necessary to study higher taxonomic relationships. Even though mitochondrial gene sequences are conserved among sponges and thus cannot provide such resolution, gene order is highly variable. A data set of mitochondrial genomes was used to compare gene order and study relationships between orders within the class Demospongiae. We have expanded on this data set to allow comparisons among two of the three classes within Porifera. We sequenced two novel mitochondrial genomes from two new species of glass sponges collected from the abyssal northeast Pacific Ocean. We compiled the mitochondrial genomes of *Bathydorus sp. nov.* and *Docosaccus sp. nov.* with previously published mitochondrial genomes and used the resulting phylogeny to evaluate the two competing hypotheses in sponge systematics: whether the three classes within Porifera are monophyletic or paraphyletic.

Endemic Lubomirskiidae of Lake Baikal: molecular-biological approaches to study evolution, biosilification and diversity of associated microorganisms

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Lake Baikal is the oldest (estimated age is about 30 million years) and deepest (maximum depth is 1637 m) freshwater lake in the world with high level of endemism. In comparison with other ancient lakes Baikal has richest fauna of endemic sponges which are unique model for study evolution, biosilification and symbiotic relationships. Nuclear and mitochondrial genetic markers were used to study evolution of baikalian endemic sponges species flock. Analyses of 18S rRNA and COXI gene revealed that Lubomirskiidae have common origin with other freshwater sponge families. Based on paleontology data and sequences of mitochondrial COXI gene it was shown that rate of sequence evolution of this gene in sponges is about 0.1-0.4 % nucleotide substitution per million years and is low in comparison with other Metazoa. According COXI sequences time of divergence between Lubomirskiidae species was estimated to be approximately 0.5-4 million years ago. Analyses of the internal transcribed spacers of rDNA of 25 species of the suborder Spongillina indicated that the endemic family Lubomirskiidae is monophyletic and have common origin with *Ephydatia muellery* (Spongillidae). Interspecific differences between Spongillidae species exceed intraspecific variability that indicate the utility of ITS region for species identification. At the same time genetic distances between species of Lubomirskiidae are equal to intraspecific and intragenomic variability supporting the hypothesis of a recent species divergence within Lubomirskiidae or question their specific status. Special proteins – silicateins – are involved in the process of sponge spicule morphology determination and silica deposition. In our study the exon-intron structure of four silicatein-α isoform: -α1, -α2, -α3 and -α4 from endemic baikalian sponge *Lubomirskia baicalensis* was determined and characterized as well as silicatein-á gene fragments of several freshwater sponge species. Phylogenetic analysis made on the basis of different known silicatein sequences comparison shows that freshwater sponge authentically differ from marine group. It is shown that freshwater sponge silicateins always demonstrate homology with one of the four known isoforms of *L. baicalensis*. Evidently these isoforms are orthologous proteins and all silicatein genes were evolved from the ancestor gene as result of thee duplication events. Further investigation of the silicatein gene sequences and structure also gives a good promising for the development of molecular marker for the closely-related species identification. Sponges harbor large number of diverse bacteria, majority of which are uncultivated. In contrast to marine sponges, freshwater-sponge microbial communities are poorly studied. For the fist time we described phylogenetic diversity of microorganisms associated with Lake Baikal endemic sponge *L. baicalensis*. By constructing of full-length 16S rRNA gene library, RFLP-analysis and clone sequencing we have shown abundant bacterial diversity existed in this sponge. A total of 102 bacterial 16S rRNA clone sequences making up to 22 operational taxonomic units (OTUs) were analyzed. About 36% of OUT were affiliated to Actinobacteria, which is a potential source for drug research. Clones affiliated with alpha-Proteobacteria, beta-Proteobacteria, Verrucomicrobia, Nitrospiraceae and Bacteroidetes were also observed. Thus Lake Baikal sponges are the good source for investigation of microbial communities as potential producers of biologically active secondary metabolites.

Calcareous sponges redefine the limits of mtDNA evolution in the animal kingdom

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Mitochondrial DNA (mtDNA) is one of the favorite molecular markers in animal phylogenetics due to several intrinsic and methodological advantages. Although early studies depicted animal mtDNA as a remarkably uniform molecule (a "frozen genome"), a more comprehensive sampling of mitochondrial genomes especially from non- bilaterian animals, has revealed their extensive diversity. In particular, the newly sequenced mtDNA from various Porifera clades (including Demospongiae, Homoscleromorpha, and Hexactinellida) displayed multiple unique features and several opposite evolutionary tendencies. Remarkably, no genuine mitochondrial sequence has been reported from Calcarea to date. To rectify this deficiency, we obtained partial mitochondrial sequences for two calcarean species *Clathrina clathrus* and *Leucetta chagosensis* (Calcinea) by traditional PCR approach and EST library screening. Our analyses revealed an unusual combination of features in the calcareous sponge mtDNA. In addition, the rate of mitochondrial sequence evolution in this group was found several orders of magnitude higher than in other animal groups, hindering any attempt to reconstruct phylogenetic relationships among sponge classes using mitochondrial data. The combination of unusual mitochondrial features in calcareous sponges puts into a new perspective the extent of possible mtDNA evolution in the Animal Kingdom. Given that incomplete nature of the mitochondrial genome presented here, and that we only explored two representatives from the subclass Calcinea, we expect that future studies of calcareous sponge mtDNA, and particularly from the second subclass Calcaronea, will uncover even more exceptional features in these remarkable animals.

Calcareous sponge biomineralization: ultrastructural and compositional heterogeneity of spicules in *Leuconia johnstoni* Carter 1871

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In contrast to siliceous sponge spicules, the biomineralization in calcareous sponges is poorly understood. In particular, the existence of a differentiated central core in some spicules has been a subject of controversy for a long time. Here we combine high spatial resolution observations and analyses, including NanoSIMS (Secondary Ion Mass Spectrometer), Raman microspectroscopy, SXM (Scanning X-ray Microscope), AFM (Atomic Force Microscopy), SEM (Scanning Electron Microscopy) and TEM (Transmission Electron Microscopy) to investigate the composition, mineralogy and ultrastructure of the giant tetractines of *Leuconia johnstoni* Carter, 1871 (Baeriidae, Calcaronea) and the organization of surrounding cells. We demonstrate that a compositionally distinct core is present in these spicule types. The core measures ~3.5 to 10 µm in diameter and is significantly depleted in Mg compared with the adjacent outer layer in the spicule. Measured Mg/Ca ratios in the core range from 70-90 mmol/mol compared to ~125-130 mmol/mol in the adjacent calcite envelope. This core is also lightly enriched in S. However, this heterogeneous distribution of Mg and S is not reflected in the mineralogy and the microstructure. Raman spectroscopy demonstrates that giant tetractines (as well as others spicule types) are entirely composed of calcite and that no other phase, such as ACC (Amorphous calcium carbonate) is detected. SEM examination of slightly etched spicules (0.1% formic acid during 2 min 15 sec) indicates an ultrastructure organized hierarchically in a concentric pattern, with layers less than 250nm in width inside layers averaging 535„b260nm. No change in structural pattern corresponds to the Mg/Ca variation observed. At the nanometer length scales, AFM and TEM observations show a nano-granular organization of the spicules. A network of intraspicular organic material containing proteoglycans (stained by Ruthenium Red) is intercalated between nanograins ƒ160-130 nm in diameter. Combined with observations of sclerocyte cells in the process of spiculogenesis, it is proposed that the compositionally distinct core is produced by a sub-apical sclerocyte "founder cell" that controls axial growth, while the envelope is secreted by lateral sclerocytes "thickener cells", which control radial growth.

The *Amphimedon queenslandica* genome and the origin of development

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We have recently analysed the genome of the demosponge *Amphimedon queenslandica* from an evolutionary perspective, with a view to reconstructing the first evolutionary steps that led to the first animals. Through comparison with other ancient metazoan branches, we have attempted to map a chronology of genetic innovations in early animals, particularly focusing on developmental novelties. More than adult morphology, development reconciles sponges with the rest of the animal kingdom. It is hence not too surprising to see that many developmental genes are present in *Amphimedon* and thus likely arose with the first metazoans. We have analysed the striking amount that *Amphimedon* and other metazoans have in common – e.g. many neural and muscle developmental transcription factors, all major developmental signaling pathways, most components of epithelium formation and innate immunity. We have also catalogued what *Amphimedon* is missing, which indicates a gradual enrichment of the developmental program and body plans in early metazoans. Hence, molecular data is providing independent support for what morphology tells us – that sponges are not so different from other metazoans, but rather form part of a continuum of increasing complexity in the first branches of the metazoan tree.

Mitochondrial genomics and phylogenomics of sponges

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Sponges and bilaterian animals shared their last common ancestor in the Precambrian, and both groups underwent major radiations in the early Cambrian. Since then, they have taken different evolutionary routes. Bilaterian animals diversified into a multitude of body plans, conquered all environments, and became one of the dominant life forms on the planet. Sponges stayed on the bottom of the sea and retained their morphology and filter-feeding lifestyle. Multiple changes occurred in the genomes, both nuclear and mitochondrial, of bilaterian animals. The changes that occurred in the genomes of sponges are largely unexplored. Here we analyze >50 mitochondrial genomes representing the four major groups of sponges (Calcarea, Hexactinellida, Demospongiae, and Homoscleromorpha) and all major lineages within Demospongiae and investigate major trends in mtDNA evolution in these groups. We show that both the tempo and the mode of mitochondrial genome evolution vary greatly among these groups. In fact, the extent of mitochondrial genome diversity observed in sponges exceeds that found among all bilaterian animals. We discuss the possible evolutionary forces that shape mitochondrial genomes in sponges and other animals and present a phylogenetic reconstruction of sponge relationships based on mitochondrial genomic data.

Investigating the specificity of larval settlement in the Caribbean Orange Icing Sponge *Mycale laevis*

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Despite intense competition among benthic sessile organisms on space-limited coral reefs, the commonly observed association between the Caribbean Orange Icing Sponge *Mycale laevis* and scleractinian corals has been hypothesized to be mutualistic. As a result of this putative mutualism, does the sponge specifically settle in response to the presence of live corals? To investigate this association, larvae of *M. laevis* were collected using traps at Bocas del Toro, Panama. Settlement assays demonstrated that larvae of *M. laevis* did not preferentially respond to the presence of live coral in terms of metamorphosis or settlement compared to other substrata. Moreover, benthic surveys conducted at coral reefs off Key Largo, Florida, and Bocas del Toro showed that *M. laevis* occurred on other kinds of substrata, such as dead coral and other species of sponges. Why then is the sponge-coral association commonly observed? The response of *M. laevis* to predation is offered as an alternative explanation. Previous experiments have demonstrated that *M. laevis* is chemically undefended, and readily eaten by spongivorous fishes on the reef when exposed. The sponge thus gains an obvious advantage from associating with corals- that of a predation refuge, an advantage that is also conferred by other reef substrata. In overfished areas where sponge predator densities are low, the proposed mutualism breaks down and *M. laevis* actively competes with live corals for space. The apparent association of *M. laevis* with live corals is driven less by a mutualistic relationship, and more by post-settlement mortality due to predation and substratum availability.

Northern Philippine sponges: taxonomic identification, associated marine microorganisms and bioactivity

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The presence of marine microorganisms was investigated on sponges collected in Pangasinan, Philippines. Marine microorganisms from fourteen sponges have shown initial bioactivity when tested against *Pseudomonas aerigunosa*, *Klebsiella pneumoniae* and *Staphylococcus aureus*. Taxonomy of the sponge and identification of marine microorganisms based on molecular method is presented in this paper. The fourteen sponges belong to eight orders and the marine microorganisms isolated from these sponges belong to six classes and these are dominated by *Pseudomonas* of the class Gammaproteobacteria. It also showed highest inhibition among the microorganisms on the test organism *Staphylococcus aureus*.

The vast potential of transcriptomic approaches for advancing poriferan genetics research

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The marine sponge mesohyl represents a complex microcosm with a plethora of microbial species that potentially interact with the sponge host and among each other. In some sponge species microbial biomass can reach above 50%, with over a hundred different microbial ribotypes present, including bacteria, fungi and protozoans. At the genetic level, knowing the complete nucleotide genomic sequence of any given organism does not guarantee complete elucidation of basic physiological processes. For a more precise understanding, the post-genomic era has begun to focus on the active gene expression patterns of mRNA transcripts (transcriptomes) and encoded proteins (proteomes) from the DNA genome. At this point in time, analysis of Poriferan transcriptomes and meta-transcriptomes is at a relative infancy. To address major questions circulating around sponge biology and symbiosis models (including uncultivable microbial symbionts), molecular genetics approaches will be the most efficacious, especially when using advanced high-throughput DNA sequencing technologies, such as "454" pyrosequencing. This paper describes the current state-of-the-art transcriptomics and meta-transcriptomics research on marine invertebrates, and its growing relevance to various aspects of sponge biology including physiology, microbiology, genomics and natural products. Preliminary results in our laboratory show that many unique mRNA transcripts are generated within the complex microcosm of sponge mesohyl. In an effort to enrich for potentially unique microbial symbiosis or secondary metabolite (SM) gene transcripts between lithistid sponge Discodermia dissoluta ecotypes producing varying levels of SM's such as discodermolide, suppression subtractive hybridization (SSH) PCR was applied to generate an EST library. Out of approximately 5000 total EST sequences, close to 30% appeared to be novel or hypothetical, showing no matches to current sequence database entries queried by BLAST. After various bioinformatics analyses, several ESTs showed distant similarities to secondary metabolite sequences, while others showed relationships to symbiosis, virulence, stress response and TGF-beta cell growth pathways. For example, a tenascin-like sequence was identified, which is a protein that can modulate cell adhesion, migration, and growth. Quantitative "Real-Time" qPCR then validated differential gene expression of two hypothetical Discodermia transcript sequences. Our research also anticipates the generation of at least one baseline sponge meta-transcriptome using current Titanium 454 GSFlx (Roche) pyrosequencing methods, which has the capacity to sequence up to 0.5 Gigabase pairs, with individual read lengths of ~ 300 bp. Possible sponge specimens will stem from local Florida and Caribbean waters, such as the model sponge *Axinella corrugata*, which produces secondary metabolites and has a wide diversity of microbes, including Vibrio sp. related to food-borne pathogens. Alternatively, *Amphimedon compressa* has been shown by fluorescent in situ hybridization to hold bacteria related to coral microbial pathogens (see K. Negandhi poster). Overall, the cataloguing and characterization of more sponge mRNA sequences is expected to provide an important baseline profile of both eukaryotic and prokaryotic transcriptomes within a symbiosis context for this important basal metazoan.

Sponge fauna from the Late Eocene of southern Australia - reconstruction based on loose spicules record

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Despite the fact that Eocene sponge fauna from southern Australian, i.e. from the Vincent Basin near Adelaide, Bremer Basin and Norseman region in SW Australia, is known since very long, it remains still poorly studied. Especially nonlithistid demosponges that are represented mostly by loose spicules remain unstudied taxonomically. The study of morphological types of these spicules revealed that the most abundant morphotypes of the spicules are monaxons, but various triaens, calthrops, sterrasters, spirasters, spherasters sanidasters and pycnasters are also frequent. Minor dischaele, discorhabds, anthasters, pynakids, sigmas, discorhabds and flattened exotylostyles are present either. Only rare hexactinellid spicules, mostly dermal pinnules were noted. Comparison of the Eocene demosponge spicules with Recent ones shows that among nonlithistid demosponges, representatives of Astrophorida (e.g. families Pachastrellidae and Geodiidae) dominated during the Eocene in this region, but representatives of Poecilosclerida (families Mycalidae, Guitarridae, Latrunculidae and Coleospheaeridae) also occurred. Hadromerids (families Placospongidae and Clionidae) and spirophorids (family Samidae) were less common members of this Late Eocene sponge fauna. Worth of mentioning is the occurrence in this Late Eocene Australian fauna of exotylostyles and flattened exotylostyles identical with those of Recent *Mycale* (Rhapidotheca) species known today only from Northern Atlantic, but found earlier also in the Miocene deposits of this region. In general, the southern Australian fauna can be compared to that from the New Zealand described by Hinde & Holmes (1892) from the Eocene of Oamaru diatomite, but the New Zealand sponge fauna seems to be more diversified. The common presence of amphidiscs and other hexactinellid spicules, suggests relatively deepwater setting in the case of New Zealand, while southern Australian sponge fauna, where the hexactinellids are very rare, indicates more shallow-water environment.

Microorganisms are not responsible for the disease-like syndrome affecting the marine sponge *Ianthella basta*

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A disease-like syndrome is currently affecting a large percentage of *Ianthella basta* over a large geographical area, with 43% and 66% of surveyed sponges displaying symptoms in the Palm Islands and Torres Strait, respectively. Symptoms of the syndrome include discolored, necrotic spots leading to tissue degradation, exposure of the skeletal fibers and disruption of the choanocyte chambers. To ascertain the role of microbes in the disease process, a comprehensive comparison of bacteria, viruses, fungi and other eukaryotes was performed in healthy and diseased sponges. All techniques indicated a very low diversity of microbes with both healthy and diseased sponge communities dominated by three strains: an Alphaproteobacteria, a Gammaproteobacteria and a Group I Crenarchaea. Bacterial cultivation, community analysis by DGGE (bacteria and eukarya), sequencing of 16S rRNA clone libraries (bacteria and archaea) and direct visual assessment by electron microscopy failed to reveal any putative pathogens. In addition, infection assays could not establish the disease in healthy sponges even after direct physical contact with affected tissue. These results provide substantial evidence that microbes are not responsible for the formation of brown spot lesions and necrosis in *I. basta*. Future research will investigate the role of environmental factors and auto-immune dysfunction in the onset of brown spot lesions and necrosis. Preliminary findings suggest that elevated sea water temperatures have little effect on *I. basta*, with similar microbial community compositions between treatments.

Biogeography of Porifera from Brazilian oceanic islands

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The Brazilian oceanic islands (BOI) have strategic values concerning its environmental, scientific and economic significance. Sponges (Porifera) are important animals that structure and maintain biodiversity. Yet, the taxonomic knowledge of this group is still scarce in these islands. This is the first study about the sponge biogeography of the BOI. Four out of five Brazilian oceanic islands were studied: Atol das Rocas (AR), Arquipélago de Fernando de Noronha (FN), Arquipélago de São Pedro e São Paulo (ASPSP) and Ilha da Trindade (IT). A total of 138 species of sponges was listed to these areas. A species x areas data matrix was constructed with AR, FN, ASPSP, IT + neighbor areas (from literature data): Caribbean (CA), Coast of Brazil (CB), Western Coast of Africa (WA); in a total of 1142 species. These species were distributed as follows: 70 (AR), 76 (FN), 26 (ASPSP), 23 (IT), 346 (CB), 602 (CA) and 310 (WA). Based on this matrix, a Parsimony Analysis of Endemicity (PAE) and a Cluster Analysis were developed. The PAE generated only one area cladogram, with a high Consistency Index (CI) = 0,913; showing biogeographic affinities among the fauna of the BOI with the Caribbean and the Coast of Brazil. The Cluster Analysis showed two groups: (1) coastal areas and (2) BOI, but the similarity indexes obtained were low. The minimal Endemism Rate calculated for these islands were 11.5% (ASPSP), 8.7% (IT), 7.1% (AR) and 2.6% (FN). Oceanographic, geographic, and geomorphologic features of each of these islands were recorded from the literature and personnel observations, trying to understand the patterns of diversity, endemism and connectivity of sponges. The geomorphologic and environmental differences between AR and FN might act as filters for sponge species, as they are geographically close but harbor different assemblages of sponges. In ASPSP, the presence of species shared with the NE Brazil and Caribbean, plus the total absence of any species exclusively shared with WA indicate the influence of the Equatorial Under Current as a way for colonizers coming from the coast. In Ilha da Trindade, it is necessary to raise the collection effort to develop a better understanding of the spongofauna, but the low number of species recorded could be caused by a high degree of isolation coupled with environmental impacts. The present results agree with the Theory of Island Biogeography: smallest and more isolated islands have lower species diversity and higher endemism rates. The harboring of endemic species and the low degree of similarity among the Porifera fauna in Brazilian oceanic islands reinforce the need for conservation of unique genetic stocks.

Spiculous skeleton formation as a new model to clarify pattern formation in demosponges: The roughly spaced spicule holding up (SHU) points and the identification of spicule carrying cells

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To clarify the cellular and molecular mechanisms involved in pattern formation of demosponges, we recently focused on spiculous skeleton formation during the asexual reproduction (gemmule hatching) of the freshwater sponge *Ephydatia fluviatilis*. For the first step, we especially focused on the process of holding up the spicules like the poles of a tent. In demosponges, it is known that siliceous spicules are formed intracellularly in megasclerocytes, and are thought to be released when they mature. Then they are held up at certain points by basopinacocytes. Although the patterned spiculous skeleton of Hexactinellids (including *Euplectella*.sps) is well known, the patterns of the spiculous skeletons of demosponges remain largely unknown. In this study, we explored whether there is some pattern in the spiculous skeleton of *E. fluviatilis* and if there is, what types of cells are involved in this pattern formation. In previous studies, we could detect megasclerocytes using *EfSilicateinM1 /M2* (Funayama et al. 2005, Mohri et al. 2008). We recently showed that megasclerocytes are directly differentiated from archeocytes by identification of *EfPiwiA/B* as genes expressed in archeocytes (Funayama et al. 2010). In this study, we succeeded in tracing the movements of spicules in live sponges during gemmule hatching by establishing a method to fluorescently label spicules and using time-lapse video recording. The roughly evenly spaced pattern of newly held-up spicules was demonstrated using this video and whole-mount *in situ* hybridization of a type of collagen gene (*EfColS1*) which is specifically expressed in basopinacocytes that hold up spicules. These results suggest the presence of the mechanisms that regulate the roughly evenly spaced spicule holding up (SHU) points. How mature spicules are carried into SHU positions was next one of the important long-standing questions of sponge biology that could now be approached. We demonstrated the presence of a specific type of cells, spicule-carrying cells [designated as "transport cells" according to the prediction by Weissenfels (Weissenfels 1978)], using video recording of moving spicules with attached cells and by identification of a Sox homologue (*EfSoxB*) as a gene marker for the cells attached to spicules. The identification of transport cells and their marker gene paved the way for future studies to reveal the molecular mechanisms involved in spiculous skeleton pattern formation, such as how future SHU points are determined, and how transport cells recruit to future SHU points.

The contractile pinacoderm –new insights from 2D- and 3D-imaging and quantitative image analysis

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Research activities on sponge contraction over the last 150 years provided basic knowledge, mainly on Demospongiae. Contractions occur spontaneously, follow endogenous rhythms and in some cases sponges reduce their body volume by >70 %. External mechanical and chemical stimuli induce contraction, implying integrative capabilities. However, sponges lack muscle cells and neurons. The nature of the sponge contraction-integration system is still a matter of debate, i.e. whether contractile cells in sponges are dependent or independent effectors. To address such questions properly, the nature of the non-muscular contractile effector cells in the Porifera must be revealed. Two competing hypotheses were postulated: (1) mesohyle-mediated contraction originating from fusiform smooth-muscle-like actinocytes ('myocytes'). (2) epidermal contraction originating in pinacocytes. The aim of the study presented here is to test the two hypotheses on the sponge contractile effector for members of Demospongiae, Calcispongia and Homoscleromorpha. This is achieved by combining volumetric microtomography, quantitative time-lapse imaging, histology and ultrastructure. We studied living specimens of *Tethya wilhelma* (Demospongiae, Hadromerida), *Oscarella lobularis* (Homoscleromorpha, Homosclerophorida) and *Clathrina clathrus* (Calcispongia, Calcinea) in aquariums using digital time-lapse imaging. Specimens of all species were fixed under defined conditions in contracted and expanded state. Each species and state was analyzed using microtomography/virtual 3D reconstruction/volumetric analyses, semi-thin-sectioning/DIC-microscopy imaging of sections combined with SEM imaging of the remaining corresponding section plains, and ultra-thin sectioning and TEM imaging. Volumetrics and virtual 3D reconstruction provide strong evidence for pinacoderm mediated contraction in all three species. The volume of the canal system elements displays strong changes which account for almost the entire body volume changes. The mesohyle only slightly contracts or is possibly compressed. Some areas of the mesohyle in *T. wilhelma* even expand during contraction. Semi- and ultrathin sections did not reveal any sphincter-like structures or actinocyte layers structurally related to the contracting canals and lacunae. The surface-to-volume ratios decreases during contraction in all three species studied. The most prominent change is found for the pinacoderm-to-mesohyl ratio, which is biologically important for the sponge in order to exchange particulate and soluble matter as well as gases. Our results strongly support the presence of pinacodermal contraction in Demospongiae, Calcispongiae and Homoscleromorpha. Contractile endo- and exopinacocytes seem to be responsible for most of the macroscopic contractions which result in the strong reduction of body volumes. The partial expansion of mesohyle in *T. wilhelma* accounts for elastic energy storage in the mesohyle. From the kinetics of time-lapse imaging of whole contraction cycles, it seems likely that an actively contracting mesohyle element (actinocytes in Demospongiae?) might be involved as pinacocyte antagonists which are involve in expansion, eventually in conjunction with hydrostatic pressure mediated by differentiated choanocyte activity. Even though Hexactinellida seem to lack extensive contractility, our results indicate that a contractile pinacoderm is part of ground pattern of the Porifera. Contractile epithelial layers might even be an autapomorphy of the Metazoa.

Identification of suitable nuclear gene markers for reconstructing non-bilaterian relationships

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Phylogenetic analyses of 18S ribosomal DNA in the early 1990'ies questioned the traditional morphology-based view of Porifera as a monophyletic group, however, without providing strong support for their paraphyly ever since. In spite of rapid accumulation of molecular data during the last two decades, relationships among the early-branching metazoan taxa still remain controversial. Single-gene phylogenies proved to be unreliable for resolving the branching pattern among the main poriferan linneages due to the lack of informative sites, different gene histories, systematic bias, or stochastic error. Results of the analyses of concatenated multi-gene alignments have not come to any agreement concerning the phylogeny of this group, either. The output of this kind of analyses strongly depends on taxon and gene sampling, gene orthology assessment and tree reconstruction methods. However, some recent phylogenomic studies support monophyletic Porifera after all. The goal of our study was to further test the hypothesis of sponge paraphyly using phylogenomic analyses of different novel subsets of nuclear genes. We constructed a local database consisting of whole genome sequences and expressed sequence tag-data for 67 species including two placozoan, 16 poriferan, 13 cnidarian, three ctenophora, 22 bilaterian species and 11 outgroup species from Choanoflagellida, Ichthyosporea, and Fungi. Several different bioinformatic approaches have been employed for assembling subsets of gene markers for subsequent phylogenetic analysis. For example, one multi-gene set included sequences selected from our local database based on automated sequence orthology assignment to the predefined orthologous groups from the KOG database, using our OrthoSelect pipeline. Another concatenated sequence alignment included only those genes that were present in metazoan genomes in single copy. These datasets were analysed using maximum likelihood and Bayesian methods. Comparison of the resulting tree topologies showed that gene sampling has major effects on phylogenetic inferences.

Integrated mariculture with sponges - the MIRA:CLE concept

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Aquaculture trials were done in the East Aegean Sea with *Dysidea avara* and *Chondrosia reniformis* to test the possibility to grow these sponges in the vicinity of sea-based fish farms. Culturing sponges in the vicinity of fish farms may have two benefits; the sponges may grow faster due to an increased availability of organic food and the pollution caused by the fish farms is remediated by the filtering activities of the sponges. An initial trial was done to compare growth of the two sponge species under floating fish cages to growth in a natural, pristine environment. After being one year in culture, nearly 100% of all explants of *D. avara* survived. Growth was highest underneath the fish cages, but growth rates were low compared to earlier studies. For *C. reniformis* survival at the pristine site was 100%, and growth was estimated at 800% per year. All explants cultured underneath the fish cages died due to smothering with sediment. After the initial trial, a new, cost-saving and growth promoting method for *D. avara* was tested at the fish farm location. Explants were grown on PVC pins that were mounted into a metal frame. Growth of the sponges on the pins was eight times faster than that of sponges growing on threads. We conclude that culturing *D. avara* under floating fish cages is feasible when using the new methodology. Currently, we are testing and upscaling new methods to grow *C. reniformis* under floating fish cages for the commercial production of sponge collagen and fish feed ingredients. This study is part of a larger concept termed MIRA:CLE (Marine Integrated Renewable Aquaculture: a Closed-Loop Exploitation). Within MIRA:CLE, integrated sponge/fish mariculture is combined with large-scale cultivation of seaweeds. The seaweeds take up inorganic nutrients produced by the fish, whereas the sponges take up the organic (particulate) components in the effluent. Harvested seaweeds can be used for the production of biofuels and fish food, sponges can be used to produce feeds and fine chemicals. MIRA:CLE offers a unique opportunity to realize a sustainable production of seafood and biofuels and concurrently mitigate negative effects of ocean acidification and eutrophication on vulnerable coastal ecosystems such as coral reefs and shellfish banks.

An innovative multi-locus PCR-based assay to assess temperature stress related gene expression in the Great Barrier Reef sponge *Rhopaloeides odorabile*

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An accurate multi-locus PCR-based assay, capable of reproducing gene expression profiles of 22 genes of interest (GOIs) and 3 reference genes (FGs), has been designed for the Great Barrier Reef sponge *Rhopaloeides odorabile*, previously exposed to thermal stress. This technique utilises the Beckman Coulter GenomeLabTM GeXP Genetic Analysis System; GOIs employed in this study are known to interfere with the following functional processes: cytoskeleton/skeleton rearrangement (A-, B-tubulin,actin, gelsolin, profilin, prolidase, villin, radial spoke protein), signal trasduction (calmodulin, YWHAH, YWHAQ), chaperone (cyclophilin, HSP70, HSP90), protein synthesis/degradation (cyclophilin NIMA-interacting 4, elongation factor - Tu, poluybiquitin, ribosomal protein S9, ubiquitin conjugating enzyme), oxidative stress (ferritin) and detoxification (glutathion-S-transferase). Experimental conditions were set at 27°C (ambient), 31°C and 32°C and sponges were sampled after 1, 3, 7 and 14 days. Biological and technical triplicates were performed for each sampling time and temperature to assess the reliability of the system and expression levels of the aforementioned genes were evaluated. Results were expressed as fold changes relative to the 27°C treatment at each time point. Sponges at 32°C died after 3 days but all other specimens showed no visible signs of stress. After 1 d at 31°C and 32°C a decreased expression of actin, cyclophilin and ferritin was observed. This reaction, following thermal stress, is consistent with a molecular shut-down response and has also been detected previously in similar stress conditions. After 3 days at 31°C and 32°C, actin and cyclophilin were up-regulated, while a decreased expression was observed for calmodulin, ferritin, glutathion-S-transferase, radial spoke protein and ribosomal protein S9. Expression of HSP90 is increased in sponges at 31°C but decreased in sponges at 32°C. These results suggested that different genes within each of the functional classes behave differently to thermal stress. For instance, both actin and the radial spoke protein are involved in cytoskeletal rearrangement, yet, after 3 d at 32°C these genes exhibit opposite expression levels (increased *vs* decreased respectively). Whilst HSP 90 was up-regulated in sponges at 31°C for 3 d, after 7 d the expression level of all chaperone genes was consistent with the ambient controls. Actin remained up-regulated in sponges after 7 d at 31°C while cyclophilin, B-tubulin and radial spoke protein were down-regulated. After 14 d at 31°C, actin and ferritin showed opposite expression levels (up and down-regulated respectively), with the remaining genes showing no deviation from specimens kept at ambient temperature. Overall, results indicate that 31°C does not represent a significant thermal stress to *R. odorabile* and that a molecular adaptation occurs in sponges exposed to a 31°C treatment. These findings are consistent with previous experiments based on necrosis, pumping rates or symbiotic disruptions- indicating a very strict physiological threshold between 31°C and 32°C in *R. odorabile*.

Niche partitioning by sponges in a temperate rocky reef system

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Sponges are often a dominant component of hard substratum environments, with many species co-occurring in the same habitat and living in close proximity. Since many sponge species appear to co-exist and depend on the same food resource, niche partitioning might be expected to occur if food is ever limiting. To date niche partitioning has not been demonstrated for sponges. This study examined the diet of seven sponge species occupying the same habitat on a temperate rocky reef to determine if differences exist in the picoplanktonic organisms that are being consumed by the different sponge species. We collected *in situ* water samples from the exhalent water stream of sponges and compared the picoplanktonic organisms in the samples to those collected immediately adjacent to the sponges, using flow cytometry. Three populations of picoplanktonic organisms were identified (heterotrophic bacteria, *Prochlorococcus*, *Synechococcus*), which are well known components of the natural diet of temperate demosponges. The cells removed with the highest efficiency by all species were *Prochlorococcus* (average across all species 72 ± 18%), although the retention efficiencies of heterotrophic bacteria (average across all species, 40 ± 32%) and *Synechococcus* (average across all species, 54 ± 22%) cells varied considerably between the species. Our results demonstrate that different sponge species have different retention efficiencies for different types of picoplankton and we suggest that this is the first report of intra-phyletic food particle niche partitioning among sponges. We hypothesise that this ability of different sponge species to remove different picoplanktonic organisms with varying efficiencies represents selective resource utilisation and niche partitioning, allowing co-existence of different sponge species in the same habitat.

Harnessing secondary metabolites from marine sponge-associated microbiota by genomic mining and cultivation

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Marine sponges (phylum Porifera) are considered to be the most prolific producers of marine natural products and are well known to harbor dense and diverse microbial communities. We investigated the secondary metabolites produced by the sponge microbiota by cultivation and genomic mining. Selective isolation for actinomycetes yielded two new actinomycete species namely, *Saccharopolyspora cebuensis* and *Streptomyces axinellae* producing novel macrolactams (cebulactams) and tetromycin derivatives, respectively. Furthermore, genomic mining for biosynthetic gene clusters encoding for polyketide synthases (PKS) and non-ribosomal peptide synthases (NRPS) were also employed. Genomic library construction and sequencing of *S. cebuensis* led to the identification of the PKS involved in the cebulactam biosynthetic pathway. A widely distributed NRPS system have also been identified to be present in several sponge species, ascidian and seawater with an intriguing similarity to the NRPS cluster from a cultivated sponge-associated actinomycete. These results highlight the potential of using genomics and cultivation-dependent approaches towards the discovery of natural products from sponge-associated microorganisms.

A high diversity Middle Eocene freshwater sponge fauna from the Giraffe Pipe crater lake, Canada

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Although the fact that today fresh-water sponges are a significant element of biotas in all terrestrial water bodies and are well studied worldwide (Pronzato & Manconi 2002), very little is known about their geological past. Marine sponges are known since Precambrian but the first occurrence of freshwater sponges is from the Late Jurassic of the USA. Freshwater sponges are more common in the fossil record only since the Eocene (Pisera & Saez 2003), from where 3 species were recorded. Recently, Middle Eocene (about 40 MY old) lake sediments from the kimberlitic crater in northern Canada known as Giraffe Pipe, have been discovered, containing a rich siliceous microfossil biota, including freshwater sponge spicules. Apart from sponges, there are rich and diversified diatoms (Bacillariophyta), chrysophytes (cysts, scales and bristles) and euglyphid scales (Euglyphidae, Thecamoebians). Many of these forms have pronounced modern affinities (Siver & Wolfe 2009). Sponges are represented by loose spicules only; together with megascleres, numerous morphotypes of amphidiscs and non-amphidisc gemmuloscleres have been recognized, as well as microscleres. Based on the morphology of amphidisc gemmuloscleres, at least 8-9 species can be recognized, and 3-4 species with non-amphidisc gemmuloscleres. They belong to several different genera, the most common being *Ephydatia*, but *Spongilla* and *Heteromeyenia* were also present, as well as some other still unidentified genera. Most sponges belong to the family Spongillidae Gray 1867, but some spicules point also to the presence of the Recent family Potamolepidae Brien 1967, earlier know in the fossil state from the Miocene of Japan (Matsuoka and Masuda 2000). There is a clear differentiation in species composition in various samples that contain one to five species. This diversity is the highest known in the fossil record and can be compared with that of present-day Canada where about 15 species were recorded (Ricciardi & Reiswig 1993). Such high diversity indicates that radiation of freshwater sponges predates the Middle Eocene. This unique fauna of sponges, with clear Recent affinities, is crucial in understanding the evolution of freshwater sponges and their Recent distribution characterized by numerous cosmopolitan species and genera. This contribution is a part of US NSF Grant No.0716606.

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One step at a time: slow but steady progree towards a complete glass sponge fauna for New Zealand waters

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The beginnings of a survey of the deep water glass sponges (Hexactinellida) around New Zealand were made by the Challenger in 1874, but recent collections show that the task is far from complete. Over the last several years we have reviewed New Zealand institutional collections to determine species identifications and to begin description of the new species discovered. We are nearing completion of the first part of the project, covering the Orders Hexactinosida, Aulocalycoida, and Lychniscosida, and provide an overview here of the situation in general. Historically, the biodiversity of hexactinellid sponges in New Zealand was seen to be low. In 1904, Hutton listed only 7 hexactinellids in reports from New Zealand waters. Nearly 90 years later, Dawson (1993) listed 16 hexactinellid species from New Zealand waters, an increase of 229%. By 2008, partly as a result of surveys around New Caledonia and description of those collections by K. Tabachnick and co-workers, the New Zealand species listing had grown to 22 named species. Now in 2010, we have amassed a considerable survey of ca 300 dictyonal or 'hard' hexactinellids, collected mostly by NIWA's RV Tangaroa and RV Kaharoa and a variety of other vessels in their dredging operations over the last 60 years. Through these we have increased the dictyonal hexactinellid diversity from 4 species to 25 species and 2 subspecies (27 taxa), and increase of 675%. Most (18 of 27) of the taxa are new, with a reasonable number (5 of 18) previously known species but new to New Zealand waters. The new taxa are spread rather evenly over several families: 4 new species and 2 new subspecies of Farreidae; 5 new species of Euretidae; 4 new species of Tretodictyidae, 2 new species of the newly reinstated family Auloplacidae, and 1 new Aulocystidae (Lychniscosida). Of particular interest is the very recent discovery of *Psilocalyx wilsoni* Ijima, 1927, alive and well on the Chatham Rise and a Bay of Plenty seamount, previously only known in New Zealand waters as a fossil in the Chatham Island Tutuiri Greensand (late Paleocene–early Eocene, ca 60–53 Ma). Several dictyonine families remain as yet unrepresented in the New Zealand area, but we conclude that while the survey of dictyonine species in this area is mature, it is not yet complete. We also have well over 200 species of Rossellidae, Euplectellidae, Leucopsacidae, Hyalonematidae, Pheronematidae and Monoraphididae, which will progress towards description in the near future. The prognosis for compiling a total fauna of glass sponges around New Zealand waters is good as NIWA continues to pursue major collections throughout the EEZ. In particular the advent of DTIS imagery and refined collection techniques has assisted with greater ease of identification of the soft Hexactinellida in particular.

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Phylogenetic analyses of deep-water sponges associated with coral reefs in Europe provide new insights for their protection

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Large-scale phylogenetic analyses are essential to explain the origin and evolution of marine biodiversity. They have proved very useful for establishing baselines of species diversity and the management of marine resources. An increasing number of studies are focusing on species boundaries, distribution and on processes that control species ranges in popular shallow-water coral reefs. However, less attention has been paid to deep-water coral and sponge ecosystems, despite the fact that they thrive along our European coasts. Within the present study, we used a comparative phylogenetic/phylogeographic study of broad range sponge species to test the hypothesis of different lineages of cold-water coral associates along the European margins. The molecular diversity within *Hexadella* species (Porifera, Verongida, Ianthellidae) and *Plocamionida* (Porifera, Poecilosclerida, Hymedesmiidae) retrieved from the European shallow-water environment to the deep-sea coral ecosystems was assessed in order to get insights into (i) the genetic variation of these species within, between and outside reefs (ii) the connectivity or isolation of these reefs along the European margins. Deeply divergent clades congruent across the mitochondrial (COI) and nuclear gene fragments (28S rDNA and the ATPS intron) suggest the occurrence of several cryptic species, while widely distributed specimens within these clades suggest an old or putative recent connection between the reefs. Once revealed by molecular markers, an 'a posteriori' search of diagnostic phenotypic characters and description of these new cryptic species is the next step for taxonomists. Moreover, these sharp genetic breaks as well as connections in the deep-sea coral habitats stress the need to protect multiple lineages of cold-water coral reefs along the European margins.

Metabolic processes in marine sponges related to their associated microbial community

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Marine sponges are ancient metazoans common on benthic substrates around de world. Sponges are filter feeders that efficiently remove organic particles in the 0.5 to 2 microns size range from the water mass, although they can also remove some larger particles at a lower efficiency. As a result of the oxidation of the particulate organic matter ingested by the sponges, release of dissolved compounds such as DOC, DON, ammonia, and phosphate would be expected. The high filtration and efficient particle retention rates of sponges suggest their potentially relevant role in benthic-pelagic coupling. Most marine sponges establish a persistent association with microbes including archaea, bacteria and protists. Microbial symbionts are hypothesized to contribute to the health and nutrition of sponges in different ways such as producing protective antibiotics, acquisition of limiting nutrients and processing of metabolic waste. Most of the microbial diversity associated to marine sponges has been based on 16S rRNA and functional genes library construction, or metagenomics used to infer alternative metabolic routes to the sponge metabolism. This metabolic routes included photoautotrophic C fixation with the subsequent NH_4^+, NO_3^-, NO_2^- and PO_4^{3-} assimilation, photoautotrophic N_2 fixation and chemoautotrophic C fixation by NH_4^+ and NO_2^- oxidation or nitrification, by methane oxidation or by sulfate reduction as energy sources. The main fate of all these routes is to increase microbial biomass. However, despite the wide phylogenetically and physiologically diverse microbes associated with sponges, their role remains largely unknown. We quantified fluxes of dissolved compounds on three common sponge species *Dysidea avara*, *Agelas oroides* and *Chondrosia reniformis* (Desmospongiae) which cohabite in the coralligenous community in the Mediterranean sea. We hypothesized that the ability of sponge to handle dissolved compounds and the resulting nutrient flux is related to particular metabolic processes mediated by the associated microbes. Then, marked differences in fluxes of dissolved compounds could be expected from species hosting contrasting microbial associates. However, we observed that similar metabolic processes can be achieved by sponges exhibiting totally different associated microbial community.

Germlineage in sponges

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Germ cells carry the hereditary information for the next generation, and therefore these cells must remain totipotent –should retain the capacity to differentiate into each and every cell type of all the different organs. The genetic machinery that all animals use to originate germ cells is now being unraveled, as well as the biological mechanism by which the germline is segregated from the somatic line. However, these mechanisms are far from being understood in basal invertebrates. These animals possess strong regenerative capacities based on large populations of totipotent cells that could also generate gametes (for instance: I-cells in cnidarians and neoblasts in planarians). In sponges, the most ancient metazoans, archaeocytes are considered the totipotent cells, and are also thought to be the anlagen (origin) of gametes in many instances. Choanocytes have been also suggested as giving rise to gametes and are also now recognized as pluripotent cells using molecular methods. To identify the germline in sponges, we sequenced three different genes that are usually involved in creating and maintaining the germline in the rest of metazoans: vasa, nanos and PL10. We used a diversity of sponges to reflect different embryonic mechanisms, *Ephydatia muelleri*, *Eunapius fragilis*, and *Sycon coactum*. For the selected genes, we designed RNA probes to reveal their expression in the tissue of *E. muelleri*, and performed whole-mount *in situ* hybridizations on gemmule-hatched sponges. Our results suggest that archaeocytes express these germline genes and that these will become germ cells. In the light of this new finding we suggest that the germline is not segregated early in the development in this lineage of Porifera.

Establishing an indigenous enterprise for the aquaculture of *Coscinoderma* spp. in Australia

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Kailag Enterprises Ltd is an indigenous company based in the Torres Straits of North Queensland, Australia, with a focus on commercial aquaculture of bath sponges including *Coscinoderma* spp. A ten hectare farm has been established at Yorke island, Torres Straits, to produce bath sponges for export and domestic markets in partnership with sponge retailers. Torres Strait Islanders have an intricate culture associated with the sea and a long history of involvement in the early pearling industry which has now drastically reduced in size. The sponge farm is now providing new jobs for young people on Yorke Island in an alternative marine based enterprise, and represents an exciting economic development opportunity for the region. The establishment of the sponge farm is based on significant scientific investigations into the ecology & distribution of sponges in North Queensland, as well as the successful development of aquaculture technology appropriate for the species, the site and the local people. Most of the stock husbandry and monitoring work is done using SCUBA to maintain sponge explants on vertical rope lines. Biofouling problems have been resolved with the encouragement of grazing fish to clean the sponge lines. Kailag Enterprises Ltd is currently investigating the potential for harvest of bioactive compounds in addition to quality bath sponge products.

Chemical and structural defense in the sponge *Melophlus sarassinorum* – A holistic approach

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Sponges are well known for their ability to deter potential predators. While most studies have focused on chemical defense, evidence for structural defense, or co-occurring chemical and structural defenses are still rare. We conducted field and laboratory experiments under different predator regimes to investigate the anti-predatory defense mechanisms of the sponge *Melophlus sarassinorum* from the tropical Pacific. Specifically, we tried to identify the natural predators of *M. sarassinorum* via transplant experiments and video monitoring and investigated whether chemical and/or structural defenses were specifically adapted against different predator groups. Furthermore, we tested if the allocation of defenses to the sponge ectosome and choanosome were in accordance with the optimal defense theory (ODT). Our results demonstrated that *M. sarassinorum* had evolved chemical and structural defenses to deal with predation. While chemical defense was evenly distributed in the entire sponge body likely to reduce predation by macro-predators, structural defense was allocated only to the ectosome to prevent tissue loss by meso-predators and collateral damage. The endoderm of the sponge contained higher protein levels, but revealed no structural defense. We conclude that the defensive mechanisms of *M. sarassinorum* are in accordance with the ODT and that some sponges have evolved discrete tissues to adapt to different ecological pressures.

Speciation, biogeography and phylogeny of the sponge genus *Latrunculia*, within the Southern Ocean

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A comprehensive revision of the genus *Latrunculia* was published recently based on re-examination of all type material and new collections. This revision brought the number of known species worldwide to 30, over half of which occur in the Southern Hemisphere around Antarctica, the south eastern coastline of South America, South Africa, New Zealand and Australia. In this study the phylogenetic history of the genus is constructed with cladistic techniques, using morphological characters 1) to represent a hypothesis on the phylogenetic relationship, 2) to discuss the biogeographic history and 3) to understand speciation events of the genus in the Southern Ocean. Based on the phylogenetic analysis three possible monophyletic clades are recognizable in the genus *Latrunculia* and that many of the Southern Oceans species are exclusively shallow-water taxa. Southern Ocean species are distinguished by consistent interregional morphological differences, leading to the conclusion that these species are probably complexes of closely related species derived from former widespread (Tethyan and/or Gondwanan) ancestors. It is possible that these species have evolved in shallow waters independently of the deeper water species. Frequent extinction events during the evolution of the genus, since at least the Oligocene, may explain the inferred disjunct sister-group distributions.

Cell cycle analysis of primary cell cultures of *Haliclona oculata*

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Marine sponges are a very rich source of bioactive compounds, which have the potential to provide future drugs against important diseases such as cancer, a range of viral diseases and inflammation. However, the limited availability of sponge biomass hampers the development of these potential drugs into commercially available products. An approach to gain more sponge biomass can be *in vitro* sponge cell culture. Despite many efforts by different research groups, no one has been able to develop a continuous sponge cell line and also the number of primary sponge cell cultures developed is very limited. A possible explanation can be that the sponge cells are in a resting non-proliferative state. Conventional methods to measure viability (e.g. trypan blue, FDA and PI) only give information about percentage of viable and dead cells, and do not provide information about the proliferation of the cells. Therefore we analyzed the cell cycle distribution of sponge cells, which makes it possible to discriminate between cells in a growing, resting or apoptotic phase. The method is based on the fact that during a reproductive cell cycle, cells successively are in the G1 phase, DNA replication phase (S), G2 phase and mitosis phase (M). If a cell is not dividing it is in the so-called G0 phase. During the G2 phase, the cell contains twice as much DNA as in the G1/G0 phase and this we can detect by staining the nuclei with propidium iodide and quantifying the amount of DNA per cell with flow cytometry. Also apoptotic populations can be detected, since apoptotic cells fragment their DNA, which results in a sub-G1 peak. To confirm apoptosis as detected with flow cytometry we performed an additional assay, which involved measurement of caspase 3/7 activity. Caspases are a family of cysteine proteases and play an essential role in apoptosis and can therefore be used as indicators for apoptosis. These methods were tested on cells from the sponge *Haliclona oculata*, collected from Lake Grevelingen in the Netherlands. We found that right after dissociation already 20% of the cells were apoptotic and that the majority of cells were in the G1/G0 phase and only a small fraction was in the G2/S phase, which agrees with the fact that based on cell counts no proliferation was observed in primary cell cultures. In future research we will study whether cultivation conditions like nutrients and mitogens can increase the fraction of proliferating cells and decrease apoptosis. Next to this, we want to measure the cell cycle distribution of cells taken from sponge larvae, since we expect larvae to contain more dividing cells. These cells could next be a good starting point for a proliferating sponge cell culture.

The sponge microbiome: insights into the diversity, distribution and origin of sponge microbial symbionts

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Marine sponges are well known for their association with highly diverse, yet very specific microbial communities, although many previous molecular studies mainly described the abundant members. The recent development of next generation sequencing technologies allows much deeper insights into the diversity of microbial communities and therefore also the description of rare members. We applied 454 pyrosequencing to 32 different sponge species (representing 9 orders and 13 sponge families) from all over the world, thereby generating more than 35.000 high quality 16S rRNA gene sequence tags. Our data show that the sponge microbiome consists of at least 19 different bacterial phyla. While some, such as Chloroflexi, Acidobacteria, Actinobacteria and the candidate phylum Poribacteria, are commonly detected in sponges, others were previously only rarely or not found in these hosts including Fusobacteria and the candidate phyla TM7, OD1, WS3, and OP10. These results will be further analyzed to determine the degree of similarity between microbial communities in sponges and to reveal potential biogeographical distribution patterns. One of the central current questions in sponge microbiology is where the sponge symbionts originate from. We used a comprehensive set of 16S rRNA gene sequences obtained from sponge reproductive stage (oocytes, embryos, and larvae) to provide insight into vertical transmission of microbial symbionts. Overall, the microbial diversity in sponge offspring resembles that in adult sponges. We found 28 phylotypes present in both adult sponges and their respective offspring and distributed over 11 microbial phyla which indicates that the adult microbial community is collectively transmitted through reproductive stages. Our analysis reveals the importance of vertical transmission to maintain specific sponge-microbe associations, although horizontal transfer might also occur in some sponge species or for some specific microbial lineages.

Sponge gardens of Ningaloo Reef, Western Australia are biodiversity hotspots

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Two major biodiversity projects in depths of 18-145 metres have been carried out under the governance of the Western Australian Marine Science Institute and the Commonwealth Environmental Research Centre. Surveys were undertaken on the continental shelf of Ningaloo Marine Park (NMP), Western Australia, an area that encompasses 2435 km^2. These surveys revealed that while much of the area is composed of sandy sediments and rhodolith fields supporting little or no macro-epibenthos, locally dense and diverse filter feeding communities exist. Dredge samples from these richer habitats were distinctly dominated by demosponges, both in abundance and diversity. Both projects consisted of multidisciplinary research, the first using side scan sonar, *in situ* video recording, sediment characterisation, and epibenthic sampling between 2006-2008, and the second in 2008 added multibeam sonar to the methods used before. Due to the enormous collection of sponges generated from the first project only dominant sponges (>1 kg wet weight) were identified at the Western Australian Museum, resulting in 155 identifications to operational taxonomic units to species level (OTUs). During the second project 670 specimens were resolved to >220 species OTUs by national and international sponge taxonomists in a workshop held in Perth in February 2010. Many of these species may be new to science or new records for Western Australia. The second project aimed at identifying all 670 specimens, regardless of size, and most specimens were small individuals of <100 g that often differed taxonomically from specimens in the first collection. For any one dredge tow of 100 metres length, the total wet weight of small and large specimens combined could constitute up to 100 kg of biomass. Data resulting from the two projects have greatly enhanced the biodiversity values associated with Ningaloo Marine Park, where sponge diversity was previously unknown. The richness of the observed filter feeding communities adds additional weight to the increasing perception of Australia as a global hotspot for Porifera biodiversity.

Unique properties of silicatein-silintaphin-shaped sponge skeletons: from biosintering to light transmission/ photosensitivity

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The material ("biosilica") that builds the spicules of the two classes of siliceous sponges, the Demospongiae and the Hexactinellida, is characterized by exceptional properties: high fracture toughness, stiffness and bending strength, and a chemical purity that is adequate to quartz glass (melting temperature of >1,800°C). The siliceous sponges are unique in their ability to form this material at low temperature (0-30°C) and near-neutral pH. In the past few years, the principle molecules involved in biosilica formation have been identified: (i) Silicatein, a (sponge-specific) silica anabolic enzyme (and the first enzyme at all that catalyzes formation of an inorganic polymer). Silicatein shows both silica esterase activity and silica polymerase activity. Several silicatein isoforms have been identified, which are capable to self-assemble to fractal-like structures at a defined molar ratio of the isoforms. The kinetic parameters of the enzyme have been identified. Silicatein is present in the axial filaments of spicules from demosponges and hexactinellid sponges. (ii) Silica degrading (catabolic) enzyme: silicase. (iii) Silica transporter. (iv) Silintaphin, a silicatein interactor protein. The latter protein has a structure-directing activity: it is able to assemble silica nanoparticles to rod- or fibre-like structures. Silintaphin-1 forms the "core" of the silicatein axial filaments. Siliceous spicules show a surprising phenomenon: "Biosintering". In demosponges the appositionally formed biosilica layers fuse during maturation of the spicules; in hexactinellids fusion of the central, concentrically arranged lamellae is observed. Sintering usually needs high temperatures, while in sponges this process is observed at ambient conditions. The strong reduction in activation energy in biosintering is assumed to be caused by proteins integrated in the inorganic silica matrix. Recently we demonstrated that in giant basal spicules (lengths up to 3 m) from the hexactinellid Monorhaphis chuni each of the silica lamellae is composed of several sub-lamellae and slats, which are traversed by an organic scaffold (NanoSIMS analyses). Thus the silica nanospheres formed by silicatein are in close contact with those proteins that facilitate their fusion to larger nanospheres. An exciting property of spicules is their ability to act as optical fibres. Spicules have a high refractive index core and a surrounding low-refractive-index cylindrical tube, allowing light transmission in the core by total internal reflection. Based on the light-reactive behavior of siliceous sponges, we searched for a light-generating (luciferase) and a light responding (cryptochrome) receptor system. We succeeded to clone both proteins as well as a luciferin-regenerating enzyme from the demosponge *Suberites domuncula*. The expression of cryptochrome is stimulated by exposure of the animals to light and shows a day/night rhythm. From our data, we propose that the poriferan spicular network might be part of an alternative photosensory system, consisting of a luciferase (light source), the spicular system (light transmission) and a cryptochrome (photosensor/receptor).

Acknowledgements. This work was supported by grants from the European Commission (projects BIO-LITHO, Mem-S, and MCITN BIOMINTEC), and BMBF (Center of Excellence BIOTECmarin). References: Schröder et al 2008 Nat Prod Rep 25:455-474. Müller et al 2009 Cell Mol Life Sci 66:537-552. Müller et al 2009 Appl Microbiol Biotechnol 83:397-413.

Combining chemical and microbial ecology to investigate rapid tissue reduction and recovery in the sponge *Aplysinella* sp.

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We observed a pronounced, yet reversible tissue reduction in the tropical sponge *Aplysinella* sp. under non-experimental conditions in its natural habitat, after transfer into seawater tanks, as well as after transplantation from deep to shallow water in the field. Tissue reduction resulted in the formation of small "reduction bodies" tightly attached to the sponge skeleton. Although volume loss and gain were substantial, both tissue reduction and regeneration were often remarkably rapid, occurring within few hours. Microscopic analysis of the reduction bodies revealed morphological similarities to previously described sponge primmorphs, with densely packed archaeocytes and spherulous cells enclosed by a thin layer of epithelial-like cells. Denaturing gradient gel electrophoresis (DGGE) revealed pronounced changes in the sponge-associated microbial community upon tissue reduction during laboratory and field experiments and following changes in ambient conditions after transplantation in the field. Generally, the microbial community associated with this sponge proved less stable, less abundant, and less diverse than those of other, previously investigated Verongid sponges. However, one single phylotype was consistently present in DGGE profiles of *Aplysinella* sp. This phylotype clustered with γ-proteobacterial sequences found previously in other sponge species of different taxonomic affiliations and geographic provenances, as well as in sponge larvae. No apparent changes in the total secondary metabolite content (per dry weight) occurred in *Aplysinella* sp. upon tissue reduction; however, comparative analysis of intact and reduced tissue suggested changes in the concentrations of two minor compounds. Besides applying various investigation methods, we were not able to discern a clear picture as to the ecological value of the observed tissue reduction.

Bacterial fingerprints of marine sponges with different bacterial transmission strategies

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Many marine sponges host diverse populations of microorganisms that are often vertically transmitted from mother to oocyte or embryo. However, not all sponges obtain all consistently associated bacteria via vertical transmission. To study the impact of the mode of transmission we studied the bacterial fingerprints of three sponges that live in the same environment (= the same rock) by denaturing gradient gel electrophoresis (DGGE) combined with sequencing of reamplified excised DNA fragments. Detailed studies have shown that *Petrosia ficiformis* does not vertically transmit its associated bacteria and this species was taken as a model for horizontal transmission. The bacterial fingerprint of *Corticium candelabrum* adults closely resembles the bacterial fingerprint of *C. candelabrum* embryos and this sponge was included as a model for vertical transmission. *Crambe crambe* is in contrast to the other species a low-bacterial-abundance sponge and was included to detect unspecific environmental acquisition of bacteria. We found that *P. ficiformis* and *C. candelabrum* harbour associated bacteria similar to other high-microbial-abundance sponges, while *C. crambe* shows some overlap with local seawater bacteria. In addition, the associated bacteria of *P. ficiformis* were closely related to bacteria that were associated with other sponges for which vertical transmission was proven. This implies that the mode of transmission has no major impact on the bacterial profile of marine sponges.

Where's the glass? Biomarkers, molecular clocks, and microRNAs suggest a 200-Myr missing Precambrian fossil record of siliceous sponge spicules

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The earliest evidence for animal life comes from the fossil record of 24-isopropylcholestane, a sterane found in Cryogenian deposits, and whose precursors are found in modern demosponges, but not choanoflagellates, calcareans, hexactinellids, or eumetazoans. However, many modern demosponges are also characterized by the presence of siliceous spicules, and there are no convincing demosponge spicules in strata older than the Cambrian. This temporal disparity highlights a problem with our understanding of the Precambrian fossil record – either these supposed demosponge-specific biomarkers were derived from the sterols of some other organism and are simply retained in modern demosponges, or spicules do not primitively characterize crown-group demosponges. Resolving this issue requires resolving the phylogenetic placement of another group of sponges, the hexactinellids, which not only make a spicule thought to be homologous to the spicules of demosponges, but also make their first appearance near the Precambrian - Cambrian boundary. Using two independent analytical approaches and data sets – traditional molecular phylogenetic analyses and the presence or absence of speciﬁc microRNA genes – we show that demosponges are monophyletic, and that hexactinellids are their sister group (together forming the Silicea). Thus, spicules must have evolved before the last common ancestor of all living siliceans, suggesting the presence of a significant gap in the silicean spicule fossil record. Molecular divergence estimates date the origin of this last common ancestor well within the Cryogenian, consistent with the biomarker record, and strongly suggests that siliceous spicules were present during the Precambrian but were not preserved.

Sponge mortality in the Florida Keys, USA: patterns of species response and population recovery

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In the early 1990s, widespread mortalities decimated sponge populations over hundreds of square kilometers of shallow lagoonal waters in the middle and upper Florida Keys, USA. Circumstantial evidence implies the mortalities were caused by blooms of the picoplanktonic cyanobacterium *Synechococcus* sp. Fortuitously, sponge community surveys were initiated prior to the mortalities and the recovery of the sponge populations was monitored through 2006. Consequently, we can now provide an unprecedented long-term and detailed view of sponge population dynamics following a set of environmental conditions that caused a widespread mortality. Due to the co-occurrence of several hurricanes in the study area over the study period, we are now also able to provide insight into how hurricanes affect sponge population dynamics. The results documented that there are many different patterns of abundance. The sponge assemblage at any one time may differ on a relatively short-term basis. While other deeper-water sponge communities have been shown to be stable with little change over long periods of time, our results show a much different picture for this shallow-water sponge community. Our long-term results document that species respond in different ways at different sites, and population change is dynamic, sometimes dramatic and sometimes unpredictable. We were able to document that a relatively small number of large, long-lived, species dominate the sponge community in terms of volumetric biomass. These species exhibited gradual and consistent recovery. However, our study conclusively documents that this is not a rapid process. Restoration of sponge population biomass in our study area was at least a 10-15 year process. If such mortality events re-occur on even a decadal time scale (which may be happening) the result could be chronic reduction of sponge community biomass. We saw a very different population response to hurricanes. The massive strongly attached and long-lived sponges were more resistant to damage from hurricanes than smaller opportunistic species. Hence sponge community biomass will recover from the affects of hurricanes much more rapidly than from the affects of mortalities induced by the blooms.

The cox1 gene of Tetillidae: a hot spot for mitochondrial intron insertions

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Animal mitochondrial introns are rare: they have only been found in two sponges (a tetillid and a plakinid), several Hexacorallia, all Placozoa sequenced to date and one annelid. Given their sporadic distribution and their known mobility in fungi, it has been assumed that these introns have been horizontally transmitted. However, a vertical transmission followed by many losses has also been proposed for a cox1 intron present in sponges and Faviid cnidarians. To better understand the mode of transmission of mitochondrial introns in sponges, we studied the distribution of cox1 introns among 13 representatives of the family Tetillidae (Spirophorida). Among the 13 tetillid species, four were found to possess group I introns in their cox1 sequence. Remarkably, these four introns belong to three different intron-forms, i.e., they are inserted at three different positions in the cox1 alignment and are predicted to have distinct secondary structures. Furthermore, an analysis of the endonuclease ORF (LAGLIDADG) encoded within these introns suggests that their LAGLIDADGs belong to different lineages. Thus, the pattern of intron presence observed in Tetillidae can only be explained by at least three independent intron insertions. Two of the LAGLIDADG lineages were also found to have cnidarian representatives. To evaluate whether cox 1 introns were vertically or horizontally transmitted in sponges and cnidarians, co-phylogeny tests were performed to compare the evolutionary history of the LAGLIDADG ORFs with that of their cox1 hosts. The results of the co-speciation analyses suggest that events of horizontal transfer are necessary to explain the incongruence between the LAGLIDADG and cox1 topologies. Our results thus imply that the family Tetillidae is characterized by rampant horizontal intron insertions.

Genomic signatures and molecular interactions of a sponge-bacteria symbiosis

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The poor culturability of sponge-associated bacteria has limited our understanding of the functional factors that mediate the symbiotic relationship with their host. We therefore explored the bacterial community of sponges by a combination of cultured-independent methods including large-scale shotgun sequencing and screening of environmental DNA libraries (metagenomics) as well as molecular probing. As a model case, we investigated the marine, temperate-water sponge *Cymbastela concentrica* as it possesses a spatially and temporally stable bacterial community that is distinct from the surrounding bacterioplankton. Metagenomic data of the sponge-associated bacterial community revealed a number of functional genes to be over-represented when compared to the planktonic community. These include transposable elements and phage defence mechanisms (such as CRISPRs) that would imply a need to control the genomic content of the sponge-associated bacterial community. Abundant genes involved in vitamin production, ammonium assimilation and carbon monoxide oxidation indicate specific metabolic interactions between the bacteria and their host. We have also discovered a large array of unique ankyrin repeat tetratricopeptide repeat proteins that are related to proteins found in intracellular bacteria. Molecular characterisation of these repeat proteins indicated that they might interfere with eukaryotic phagocytosis and hence might prevent symbionts from being consumed as food. Reconstruction of partial genomes of uncultured, bacterial symbionts also allowed us to link all these functional traits to specific types of organisms. This genomic reconstruction resulted in the description of a novel delta-proteobacterium as a symbiont of photosynthetic bacteria and showed that sponge-symbionts themselves are subject to complex bacteria-bacteria interactions. The specific genomic signatures and functions identified here have until now not been recognised to be involved in mediating the interaction of bacteria with their sponge host or to maintain the bacterial community composition under the particular biological, chemical and physical conditions given by the sponge environment. Our dataset delivers therefore novel markers to monitor the status of the sponge's bacterial community and hence could assess how abundance or expression of those functional genes impact on the symbiotic relationship between bacteria and sponges.

The Great White *Abyssocladia* and other Down Under wonders: three new remarkable carnivorous sponges from New Zealand

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The deep-sea carnivorous sponge fauna appears to be remarkably diverse, especially in the Pacific where most specimens collected turn out to be undescribed taxa. In particular, New Zealand's Chatham Rise, the Kermadec Trench, and Macquarie Ridge, display a high number of new species and possibly new genera that are presently in the process of description. Three are described here as new species. *Abyssocladia carcharias* sp. nov. has the shape of a pedunculate disc with radiating filaments, and is characterized by three types of special multidentate isochelae, probably deriving from anchorate chelae. *Asbestopluma* (*Asbestopluma*) *anisoplacochelae* sp. nov. is erect and cylindrical with lateral expansions. In addition to the usual *Asbestopluma* microscleres, this species displays large microscleres which we call anisoplacochelae. These microscleres bear a plate-like central tooth similar to that of the placochelae of Guitarridae, but with two dissimilar ends. *Asbestopluma* (*Asbestopluma*) *desmophora* sp. nov. is an erect dichotomously branched sponge, that has densely packed desmas in the enlarged fixation base. This use of desmas to reinforce the attachment base is not unprecedented as they are found in other (presumably) carnivorous sponges such as *Esperiopsis desmophora*, *Euchelipluma arbuscula*, and *Asbestopluma* (*Helophloeina*) *stylivarians*, but it is the first record in the subgenus *Asbestopluma*. Our interest in the taxonomy of south western Pacific deep-sea carnivorous sponges is revealing a rather surprising and considerable diversity. Unfortunately, this increase in diversity does little to clarify their taxonomic position. While most carnivorous sponge genera have been traditionally classified in the poecilosclerid family Cladorhizidae, our results confirm that several carnivorous sponges share potential affinity with other families including Guitarridae and Esperiopsidae, and that the Cladorhizidae itself is polyphyletic. If carnivorous sponges are polyphyletic, one of the outstanding problems is their relationship with the typical members of the poecilosclerid families. What is the relative phylogenetic weight of the characters common to all Cladorhizidae and other carnivorous sponges as they are presently recognized (characters such as stipitate, branching, or stick-like morphology, the global presence of mycalostyles and particular forms of microscleres etc), versus the appearance of new highly ornamented multidentate isochelae such as in *Abyssocladia carcharias* sp. nov., and ones that resemble existing isochelae (such as the anisoplacochelae in *Asbestopluma* (*Asbestopluma*) *anisoplacochelae* sp. nov.)? At present these characters are apomorphic rather than synapomorphic and thus of little value in determining phylogenetic affinity.

The origin and sources of food for cavity sponges on coral reefs based on stable isotope and fatty acid analyses

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We studied stable isotope ratios ($\delta^{13}C_{org}$ and $\delta^{15}N_{org}$) and fatty acids of 12 'cavity' sponge species and their potential food sources to determine the main origin (open sea- and/or reef-derived) and sources (plankton-, and/or reef-derived coral mucus, crustose coralline algae) of their food on the narrow fringing coral reefs of Curaçao, Netherlands Antilles. Carbon and nitrogen isotope signatures suggest that suspended particulate matter (mainly phytoplankton and bacterioplankton) is not the main food source for the sponges examined. A previous study showed that cavity sponges are mainly consuming dissolved organic matter (DOM). Results therefore imply that the DOM derived from phytoplankton and recovered by bacterioplankton (you are what you eat with respect to $\delta^{13}C_{org}$) is not an important source of DOM for cavity sponges. The more than 2‰ lower $\delta^{13}C_{org}$ values in different plankton size fractions than in sponges as well as the less than 3‰ difference in the $\delta^{15}N_{org}$ values between plankton and sponges substantiate the insignificance of the trophic link between examined sponges and plankton supplied from the open water to the reef. Interestingly, the $\delta^{15}N_{org}$ stable isotope signatures of coral mucus of 2 coral species and the tissue of calcareous coralline algae, present at entrances of coral cavities, were in trophic range of sponges (3-4‰ less than sponges, i.e. 1 trophic level). Mixture of coral mucus and benthic alga derived organic matter results in $\delta^{13}C_{org}$ values within the trophic range of examined sponges (0-2‰ less than sponges). In addition the presence of characteristic fatty acids (FAs) of coral mucus of *Madracis mirabilis* and coralline algae such as 20:4ω6 in sponges point to reef-derived sources. Moreover, 20:5ω3, which is an abundant FA in reef organic matter (e.g. in mucus of *Montastrea annularis*), was also recovered in sponges, suggesting that the source of this FA is again the reef benthos. Not all sponges showed the same FA profile. On the contrary, we established low similarity in fatty acid composition profiles of sponge species. Particularly the relative abundance of key FAs varied between sponge species as well as the abundance of bacteria specific fatty acids. This suggests that trophic niche segregation may be an important factor facilitating co-existence of these filter feeders in cryptic habitats. Our results show that cavity sponges mainly rely on reef derived food and therefore play an important role in conserving food and energy produced within the reef system.

The World Porifera Database: ambitions, status, limitations

Van Soest RWM

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Van Soest RWM, Boury-Esnault N, Hooper JNA, Rützler K, de Voogd NJ, Alvarez B, Hajdu E, Pisera AB, Vacelet J, Manconi R, Schönberg CHL, Janussen D, Tabachnick KR, Klautau M, and Picton B (editors) 2010. The World Porifera Database. The World Porifera Database (WPD: http://www.marinespecies.org/porifera/) is an online searchable catalogue of all names of Recent Porifera erected since 1759. The catalogue is part of the World Register of Marine Species (WoRMS) hosted by the Flemish Institute for the Sea (VLIZ), Oostende, Belgium. The WPD aims at being the world standard for sponge names and the world portal for internet access to Porifera information. With its expert team of editors, the WPD acts to stabilize and regulate the use of sponge names in science and society. It serves as a tool for taxonomy by facilitating inventories of taxa, literature references, distributional data, and knowledge gaps. A great advantage over traditionally published inventories is the continuous updating that takes place with each new item of taxonomic information that becomes available in the literature. Currently, the WPD contains approx. 20,000 taxon names of which approx. 8,500 are considered valid. All names are linked in a hierarchical classification based on the Systema Porifera. All names are provided with literature references to original descriptions and authoritive redescriptions of the taxon, an indication of the distribution of the taxon, and often an illustration of the habit, and an edit history. Many literature sources are made available to users by simple downloading of PDFs. A start has been made with collecting information (depositories, localities and registration numbers) of types and other important specimens. The WPD is adopted by GBIF, Catalogue of Life / ITIS, OBIS, EOL, CoML, GenBank and the Sponge Barcode Project / BOL, and a string of subscribers to these organizations, so Porifera names are increasingly matched with those in the WPD. Several regional Porifera identification tools (e.g. The Sponge Guide, The Marine Species Identification Portal, CoralPedia), and regional databases (e.g. the Australian Faunal Directory, and the Encyclopedia of the Marine Life of Britain and Ireland) are linked to the WPD. Short-term ambitions are to complete (type-)specimen-, distribution- and validity information, the further addition of non-original unaccepted combinations, and the linkage to servers with habit and microscopic images. Priority will be given to images of type specimens and reliably identified and vouchered material. Above all, the WPD initiative is contributing to alleviate the taxonomic impediment of sponges.

Molecular diversity of the marine actinobacteria genus *Salinispora* isolated from Great Barrier Reef marine sponges

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The discovery of new compounds of biopharmaceutical significance can be hindered by many factors. In the case of bacteria, identifying unique strains in initial isolation studies is the first of many hurdles. The obligate marine genus *Salinispora* is unique. It is the first actinobacteria to be reported as having species-specific chemical diversity (1). This suggests that being able to identify phylotypes, based on 16S rRNA gene sequencing, should provide an indicator to the uniqueness of an isolate and its potential for the discovery of new compounds. In this study, rep-PCR fingerprint analysis and ribonuclease P RNA (RNase P RNA) gene sequencing have been applied for the first time to investigating the diversity of the marine actinobacteria genus *Salinispora*. These methods will be used in conjunction with 16S rRNA gene sequencing to screen for bacterial clones and assist in the clarification of phylotypes from a collection of *Salinispora* isolates derived from Great Barrier Reef (GBR) marine sponges. Using sponges collected during the Great Barrier Reef Seabed Biodiversity Study (2) isolates from the actinomycete genus *Salinispora* were isolated. The initial screening study of the sponges generated 441 Salinispora-like isolates. To dereplicate the number of isolates being identified via sequencing, DNA fingerprints generated through repetitive extragenic palindromic PCR (rep-PCR) were employed. The family of rep-PCRs uses short, conserved, palindromic repeat sequences as primer sites. These sites are located throughout the genome and have a very high rate of genomic migration (3). From 441 presumptive-*Salinispora* isolates recovered from GBR marine sponges, 47 unique rep-PCR fingerprints were identified. Representative isolates of the 47 unique fingerprints were identified using sequencing of the 16S rRNA and RNase P RNA gene sequencing. The 16S rRNA gene sequences identified two species of *Salinispora*: *S. arenicola* and "*S. pacifica*" within the collection. The use of RNase P RNA gene provided support for phylotypes, which were based on the 16S rRNA gene sequencing results. This has resulted in the identification of phylotypes within the species "*S. pacifica*" that are being reported for the first time. Within the species S. arenicola, RNase P RNA gene sequencing was able to identify a sub-species clade not identified by 16S rRNA gene sequencing. The use of rep-PCR fingerprinting and RNase P RNA gene sequencing have been applied as complementary techniques to 16S rRNA gene sequencing. This has enabled the dereplication of *Salinispora* isolates and distinguishing of new phylotypes. Within the *Salinispora* genus this is considered to be of potential significance for recognition of secondary metabolite diversity.

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Sponges (Porifera) as living metazoan witnesses from the Neoproterozoic: biomineralization and the concept of their evolutionary success

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Prior to and at the beginning of the Neoproterozoic (600 Ma) the earth surface comprised initially insoluble silicates and carbonates. The silicate weathering-carbonate precipitation cycle proceeded especially during the Sturtian and Varanger-Marinoan glaciations and resulted in the formation of soluble calcium carbonate and soluble silica, under consumption of atmospheric CO_2. These minerals were leached into the waters of the rivers, lakes and oceans, where they again precipitated into new minerals (MINERALIZATION) as part of the sedimentary rocks. Simultaneously with the mineralization of calcium carbonate and silica biologically driven or controlled biogenic deposition of minerals by metazoans began; that evolutionary novelty was first seen in sponges (BIOMINERALIZATION). The formation of an inorganic skeleton, built of ortho-silicate, facilitated the development of highly structured and large metazoans. Surely other, soft body, metazoans (might have) co-existed with the Porifera prior to the Varanger-Marinoan ice age; however only the skeleton-forming sponges were powered enough with biological defense systems to resist extreme climatic/environmental changes. Fossil skeleton-free demosponges (Ceratosa) are not known. It must be stressed again that the silica-skeleton forming Porifera are the only biotic witnesses of the Neoproterozoic period that survived the Marinoan glaciation, a discovery which has recently been confirmed. The present-day Porifera and, very likely, also the Ediacaran sponges had the genetic tool kit to express proteins required for the formation of a distinct metazoan body plan. It was the enzyme silicatein that permitted the sponges to form a hard skeleton. Even though the Porifera comprise the simplest body plan, their biomineral structure formation is already highly complex. The formation of siliceous spicules in sponges is genetically controlled. Data demonstrate that at suitable concentrations, silicate induces a battery of genes that are involved in the shaping of the spicules, e.g. collagen, silicatein and myotrophin. The earliest siliceous sponges were free-moving (example: *Choia*) and became only sessile at a later stage (example: *Paraleptomitella*). During that transition the apical opening, the oscule, became wider. In parallel, the large spicules of the freely-moving sponges acted as stabilizing pillars against rotation or tumbling, whereas those in sessile sponges functioned as root or anchor spicules that can reach sizes of up to 3 m. It is concluded that it was the hard skeleton of the siliceous sponges that contributed to the successful evolution and survival of the Porifera during the last 500 million years.

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Deep sequencing reveals exceptional diversity and alternative lifestyles of bacterial sponge symbionts

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Marine sponges contain complex bacterial communities of considerable ecological and biotechnological importance, with many of these organisms postulated to be specific to sponge hosts. Testing this hypothesis in light of the recent discovery of the rare microbial biosphere, we investigated three Australian sponges by massively parallel 16S rRNA gene tag pyrosequencing. Here we show bacterial diversity that is unparalleled in an invertebrate host with more than 250,000 sponge-derived sequence tags being assigned to 27 bacterial phyla and revealing up to 2996 operational taxonomic units (95% sequence similarity) per sponge species. Replicates of the same sample type contained similar microbial communities, irrespective of the phylogenetic level examined, demonstrating (i) that different individuals of the same sponge species from the same environment harbor similar microbiomes, and (ii) that different sponge species from the same environment possess distinct symbiont communities. Of the 33 previously described "sponge- specific" clusters that were detected in this study, 48% were found exclusively in adults and larvae – implying strict vertical transmission. The remaining taxa, including "Poribacteria", were also found at very low abundance among the 135,000 tags retrieved from surrounding seawater. These scenarios have important implications for our perception of the evolution of sponge-microbe associations, because they could explain the widespread distribution of these symbionts in many different sponge hosts without the need to postulate an ancient association. Taken together, the two postulated transmission patterns of the "sponge-specific" clusters resemble those described for primary and secondary symbionts of insects and call for future in depth analyses of co-evolution of sponge hosts and their symbionts.

Consequences of reproducing on the edge of species distributional limits for marine sponges

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The question of why species do not occur everywhere and what defines a species range remains an elusive pursuit for ecologists. It is clear that environmental conditions contribute to species distributions with successful expansion of boundaries relying on a dynamic mix of dispersal ability, habitat adaptation and organism performance. Established paradigms, central to niche theory, suggest optimal habitats support centres of abundance with movement towards the periphery (often into sub-optimal habitats) requiring a higher investment in physiological maintenance, leading to poorer organism performances and declining abundances. Indeed, the ability to balance the trade among key life history traits (e.g. growth (maintenance) and reproduction) often sets the stage for how well a species performs across variable habitats. Reproduction is a key life history trait, the success of which is fundamental to individual and population performance. To establish a complete understanding of reproductive (geographic) variability, larval supply and ultimately population performance it is important to quantify reproduction across a broad spectrum of environmental (habitat) conditions. For sponges, it is equally important to establish dispersal and settlement capabilities of larvae across their species range. Using the sponge *Rhopaloeides odorabile*, this study quantified reproductive output (RO) using histological sections from sponges across the shelf-reefs of the central Great Barrier Reef (GBR). Because larval dispersal and settlement play key roles in population success, experiments to quantify larval vagility and settlement for *Rhopaloeides odorabile* were also undertaken. Of importance, is the gradient of water quality across the geography of this study where coastal reefs exhibit more turbid conditions than offshore reefs. There were stark differences in reproductive effort between coastal and offshore sponges with numbers of reproductive females and reproductive output being higher with increasing distance from the coast. Of interest, for this gonochoristic species was the finding that males showed consistent levels of RO across the shelf reefs. Larvae from offshore sponges were larger than those produced from coastal reef populations and although both larval competency and settlement were similar, post-settlement survivorship was higher for larvae from offshore populations. The results are discussed under the proposal that sponges exposed to turbid coastal waters of the GBR face heavier maintenance requirements than sponges located on offshore reefs resulting in marked differences between reproduction and larval recruitment, and ultimately population dynamics for this sponge.

Diversity patterns and zoogeography of the Northeast Atlantic and Mediterranean shallow-water sponge fauna

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Recognizing and understanding present-day biodiversity patterns and how these relate to contemporary and past climate is pivotal to predict the effect of future climate on marine biodiversity and promote adequate conservation policies. Sponges constitute an important and dominant component of the marine benthos and are therefore an excellent model group for such investigations. In this study we assessed the diversity patterns and the zoogeographical affinities of the Northeast Atlantic and Mediterranean shallow-water demosponge assemblages. Data on the distribution of 745 species throughout 28 areas was compiled from the literature and used to build a presence/absence matrix. Diversity patterns were assessed from estimates of species richness (S) and taxonomic distinctness (AvTD). The Mediterranean Sea proved to be more diverse both in terms of species richness and taxonomic distinctness (S=539, AvTD=94.74) than the Northeast Atlantic (S=480, AvTD=92.42) and the two regions together were found to constitute a diversity hotspot harbouring approximately 11% of the global demosponge diversity. We found an Atlantic N-S and a Mediterranean NW-SE gradient of increasing taxonomic distinctness that is strongly correlated to both contemporary ($R2=0.5667$; $p<0.01$) and historical values ($R2=0.7287$; $p<0.01$) of sea surface temperature at the Last Glacial Maximum (LGM). The zoogeographical affinities examined through classification (cluster analysis) and ordination (non-metric multidimensional scaling) based on the Bray-Curtis similarity index, revealed the presence of three groups approximately corresponding to the Northern European Seas, Lusitanian and Mediterranean provinces outlined in the 'Marine Ecoregions of the World' classification system. Geographical distance and oceanographic circulation were shown to constitute important factors in shaping the zoogeographical affinities among areas. The vast majority of the species occurring in the Northeast Atlantic and the Mediterranean (67% and 57%, respectively) was shown to have restricted geographical ranges, as single-area or narrow-range (2-3 areas) endemics, which raises some concerns regarding their conservation.

Potential of antitumoral (+)-discodermolide production by the Caribbean marine sponge *Discodermia dissoluta*

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The Caribbean marine sponge *Discodermia dissoluta* produces (+)-discodermolide, a potent polyketide antitumoral that has reached human clinical trials. Probably owing to local upwelling of colder waters, at Santa Marta, Colombia, *D. dissoluta* is found at shallower depths (about 12-25 m), than in other Caribbean areas, where it dwells deeper than 30 m. This makes it logistically easier to conduct field research on natural or biotechnological supply of (+)-discodermolide. Through extensive exploration, we found that *D. dissoluta* is located in places with relatively low wave-exposure, in patches at the scale of tens to hundreds of meters. It prefers horizontal to inclined, well illuminated hard reef and rocky substrata. Where it occurs, quantification in 4 m-radius circles showed densities of 2-5 individuals per 50 m^2. Point-count spatial analyses showed that *D. dissoluta* lives aggregated at the scale of decimeters to a few meters, possibly related to short-distance larval dispersal or fragmentation. Size distribution shows that about half of the individuals in the population are small (<50 mL), with a few reaching very large sizes (up to 2000 mL), consistent with high recruitment and high mortality in small and mid sizes. Tagged individuals showed natural growth rates of around 60% in volume per year, but partly harvested individuals (90% of their volume removed) only recovered about 30% of their pre-harvesting volume in this period. After standardizing a methodology for sample extraction, purification and HPLC quantification, it was found that the natural concentration of (+)-discodermolide ranges from 5 to 89 ig.g^{-1} of wet sponge, variation not consistently related to local biotic or abiotic factors. From all of the above, it is clear that the population cannot sustain direct harvesting for (+)-discodermolide production. To evaluate if *in-situ* culture is feasible, sponge fragments were hung from lines in net pockets in a structure placed at 18 m depth. After 6 months, survivorship was high (93%), growth was moderate (mean 20% to 39% in volume for three separate experiments), and (+)-discodermolide concentration increased on average 21% to 80%. These results are promising, but it is necessary to try alternative culture systems that increase biomass and compound production and to establish a stock of fragments that can make the system sustainable for research and production. We are also currently working with cell dissociation and separation techniques to identify the cells responsible for (+)-discodermolide production and to establish an *in-vitro* culture system.

Toward intensive aquaculture of marine sponge of *Hymeniacidon perlevis*: Closing the life cycle under controlled environments

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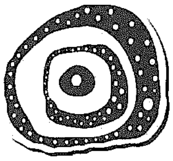
Sponges are the richest sources of bioactive natural products among marine invertebrates. The development and applications of sponge-derived bioactives are critically hindered by insufficient supply of wild sponge biomass, i.e., "the supply problem". Intensive sponge aquaculture through artificial seed rearing may provide an alternative sustainable supply of sponge biomass; however the knowledge of sponge life cycle and the technology for artificial seed production are still limited under controlled aquaculture environments. Over the past 10 years, our group has focused on the development of scientific understanding and technology for intensive sponge aquaculture, mainly using an intertidal marine sponge *Hymeniacidon perlevis* as a model. The long-term field observations reveal the natural life cycle of *H. perlevis* to have four different developmental stages of the dormancy at winter (December to February), the resuscitation at spring (March to April), the bloom at summer (May to August), and the decline at autumn (September to November). Adult sponges release larvae during August to October, and can produce gemmules during the decline phase. Based on this observation, a protocol has been successfully established to collect sponge explants from the field and to harvest sponge larvae under laboratory-controlled environments. The larval release is an asynchronous event. The attached sponge explants release 5 times more larvae than the unattached. Over 12 days of release, the mean release rate was 7.2 larvae g^{-1} wet sponge day^{-1} for attached sponges. Over 7 days of release for unattached sponges, the mean release rate was 2.6 larvae g^{-1} wet sponge day-1. Light stimulated the sponge larvae release. The highest number of sponge larvae (195.8 larvae g^{-1} wet sponge) was released at 18 °C, while only 48 and 51.7 larvae g^{-1} wet sponge were released at 14 °C and 25 °C, respectively. After releasing, larvae swam to the water surface column and stayed for 3-24 hours, and moved to explore suitable substrate to settle. Larval settlement was favored in dark condition, with the highest percentage of larvae settled at 22 °C. Then the settled larvae undergo metamorphosis, finally develop into juvenile sponges. To rear the early juvenile sponges, the effects of food, temperature, water flow and light on the growth and survival were examined. Temperature changes have striking effects on juvenile growth. Juvenile sponges grow faster when they are shifted to higher temperatures (18 to 23°C) than when they are shifted to lower temperatures (18 to 4 to 23°C) or kept at a constant temperature (18°C). Periodic water flow and light cycles favor early juvenile sponge growth. Under the optimal conditions, the morphology of juvenile sponges turned to strip from disc shape, the area of juveniles increased about 29 times. To further rear the juvenile sponges to mature sponges and to embryo-bearing sponges under controlled environment is still a challenging task, but promising results will be presented from our recent studies. More R&D attentions are deserved for this technology to provide a low-cost solution to "the supply problem" of sponge biomass for biotechnological applications.



POSTERS

A cost effective technique for measuring relative water movement for studies of benthic organisms

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Water movement has been shown to affect the structure, population and community composition of the marine environment, and as such the ability to monitor and record water movement is important in marine research. This study describes a cost-effective, repeatable method for measuring relative water movement both spatially and temporally using an 'off the shelf' accelerometer. The technique involves suspending the accelerometer in the water column near the seafloor; movement of the apparatus by the water column is recorded as changes in acceleration relative to earth's gravity ($9.8\ m\ s^{-2}$). Water movement recorded by the presented technique was highly correlated ($r = 0.907$, $P < 0.01$) with a recognised commercial device (wave rider buoy). Deployment tests revealed that total wave height showed the most significant relationship ($r^2 = 0.83$) with the technique. Use of the technique in a field situation allowed the detection of small scale water movement patterns within the Houtman Abrolhos Islands off Western Australia, and the quantification of the relative differences in water movement among coral monitoring sites. Overall the technique is a cost-effective way of obtaining basic long term temporal water movement data at small spatial scales (100's meters) or in areas lacking this information.

Molecular markers for sponge population studies in the genomics era. Are microsatellites still the best choice?

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Microsatellites are hyper variable, polymorphic, tandem repeats that have been successfully used in population genetics of animals and plants during the last decades. Conversely, sponge researchers continue searching new, intra-species variable genes for studies of phylogeography, gene flow, imbreeding, clonality, and other population descriptors, while only four sponge species have been targeted for population studies by using microsatellites up to now. The main reasons for the underuse of microsatellites in sponges has to be searched in its species-specificity and the consequent time-consuming methodologies that were involved in the necessary construction of microsatellite enriched libraries. Moreover, these procedures gave a low yield of sequences-containing microsatellite, which compromised microsatellite selection for suitability in population genetic studies. The genomic era has changed our approach to the search and primer design of species-specific microsatellites. Sequencing a small part of the genome can provide thousands of microsatellite sequences. Implementation of already existing software, for searching repeats, allows a more stringent microsatellite selection by discarding those sequences that deviate from single mutation models. Here we exemplify this issue by comparing microsatellite yield and quality, and time-effort from preliminary results obtained by using either enriched gene libraries construction or mass

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Distribution of Nickel in the tissue of *Cliona viridis* from the coast of São Miguel (Azores)

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Marine sponges can selectively accumulate considerable amounts of trace elements even when their levels are low in the surrounding environment. In particular, *Cliona viridis* has been shown to accumulate high levels of nickel, in a manner which seems to be independent of the amount of this compound found in the sediment and in the water column (Araújo et al., 2003). The protein responsible for this bioaccumulation has been identified and partly characterized (Esteves et al., 2006). However, it is not clear what is the role of this accumulation in the sponge metabolism. The aim of this work was to find out whether there is a spatial pattern in this accumulation, and if it is associated with any particular structure or tissue location in the sponge. If Ni accumulation is associated with a particular area within the individuals or with a specific site within its cells, this may enlightenwhich metabolic pathways are involved in the Ni accumulation processes. Samples from *Cliona viridis* individuals were collected near the south coast of S. Miguel. Scanning electronic microscopy coupled to element percentage determination were carried out using energy dispersive X-ray spectroscopy (EDX) in order to quickly screen for Ni variations among different sponge individuals and to compare Ni accumulation at separate locations within the same sponge (JEOL scanning electron microscope with an elemental composition detection limit of 0.1%). Several fields were chosen for elemental determination per specimen. Results are in agreement with the expected accumulation of nickel since Ni levels ≥ 0.1% were present in 84.1% of the 44 spectra analyzed. From all the areas in study only 7 did not shown Ni concentrations above 0.1% and 10 spectra had Ni ≥ 1%. In fact, the median Ni concentration reached 12.3% at one of the fields analyzed (with a maximum read of 17.1% Ni). In this case the high Ni concentration was associated with high levels of Fe, and moderately high levels of Si. This Si enrichment indicates the proximity to a sponge spicule. However, thus far, no consistent pattern of association between sponge structures and Ni accumulation was detected. In fact, in other areas where Si level was high, Ni level was highly variable (Ni values ranged from medium amounts to below detection level). Our results seem to indicate a heterogeneous, apparently random distribution of Ni accumulation, at least in the surface of the sponge. The data here presented is still preliminary and we are in the process of enlarging the study sample to include sponge individuals from different spots within the Azores to further test the absence or to confirm the existence of a possible pattern for Ni accumulation in *Cliona viridis*.

A new *Clathria* (Microciona) (Porifera: Demospongiae: Poecilosclerida: Microcionidae) associated with the sea urchin *Hesperocidaris asteriscus* from the Punta Sal Region, Northern Peru

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Until the end of the twentieth century, the sponge fauna along the Peruvian coast has been poorly investigated with barely twenty records, mostly abyssal Hexactinellids. In 2007 the Project ESPER (Esponjas del Perú) started, followed in 2008 by Project EsponjAS (Esponjas de América del Sur), both with the same objectives: to make an inventory of the shallow sponge fauna along the coast and to encourage local investigations. In this work, we describe the association between the sea urchin *Hesperocidaris asteriscus* Clark, 1948 and a new *Clathria* Schmidt, 1862 species found in the northern region around Punta Sal in three localities: Bajo el Cardo 1, Bajo el Cardo 2 and Balneario Punta Sal. For each locality, linear transects (10 x 2 m) were performed, recording all sea urchins and collecting at random 10 specimens of *H. asteriscus*. A total of 377 sea urchins were registred, all of them had some spines regularly covered with a sponge of the genus *Clathria*. For the 30 specimens collected, the number of spines covered with sponges varied from 18.2 % to 75.7 % with a mean value of 47.7 %. This sponge had a red color in life with a velvety surface and a thickness never exceeding 5 mm. Dissociated spicules were observed in scanning electron microscopy (SEM) and transversal ground sections of the sea urchin spines covered with the sponge were observed in light microscopy. Megascleres were principal massive (sub)tylostyles with microspined base, fusiforms and slightly curved (50-330/3-20 µm) and auxillary thin (sub)tylostyles, straight, frequently fusiform and slightly curved (70-340/< 2 µm); microscleres were smooth toxas of two categories (179-262 µm and 13-260 µm) and palmate isochelae (8-18 µm). The size of the spicules and their organization differentiate this sponge from the 12 known *Clathria* from the Eastern Pacific. *Clathria* (*Microciona*) sp. nov. is the first species of this genus described for Peru. It will be interesting to compare it to the orange crustose sponges found on the primary spines of *Eucidaris thouarsii* (Valenciennes, 1846) recently reported for shallow-water specimens collected off the Pacific coast of Panama. This opens up the possibility that the new *Clathria* reported here may extend its range further north in the Tropical Eastern Pacific.

Taxonomic revision and molecular phylogeny of the Family Leucettidae de Laubenfels, 1936 (Calcarea, Calcinea)

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Leucettidae sensu Borojevic, 1968 is one of the most complex families of the order Clathrinida. The family is diagnosed by leuconoid aquiferous system, well-developed choanoskeleton composed of triactines and/or tetractines, and thin cortex composed of spicules similar to those of the choanoskeleton. Originally, Leucettidae was composed of three genera *Leucetta* Haeckel, 1872, *Pericharax* Poléjaeff, 1883, and *Teichonella* Carter, 1878. Nowadays, only *Leucetta* and *Pericharax* are considered as valid genera, since *Teichonella* was synonimyzed to *Leucetta*. Recent studies, however, suggest that a possible synonymy between *Leucetta* and *Pericharax* cannot be discarded due the great similarity that both genera exhibit, differing only by the presence of subcortical lacunes in *Pericharax*. The principal difficulty concerning the taxonomic history of Leucettidae are the synonymies produced by some authors that considered the diagnostic features present in the original descriptions of *Leucetta* and *Pericharax* as plastic characters and then, invalid to distinguish species assigned to these genera. The family is considered cosmopolitan with circumtropical distribution. Forty-eight species have already been assigned to Leucettidae. However, presently only 25 species are recognized as belonging to the family: 21 to *Leucetta* and four to *Pericharax*. Given the complexity of Leucettidae, the purpose of this study is to verify the validity of the species currently attributed to this family, and also to check its monophyletism and the possible synonymy between *Leucetta* and *Pericharax*. Our molecular analyses (ITS) showed the monophyletism of Leucettidae, and that *Pericharax* is a junior synonym of *Leucetta*. The taxonomic revision revealed that the family Leucettidae is monogeneric and *Leucetta* is composed of 20 species, distributed in the Atlantic, Pacific and Indian Oceans.

Acanthoisochelae and the classification of Guitarridae (Poecilosclerida: Mycalina)

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Spiny isochelae-like microscleres known in the literature as acanthoisochelae have been reported from the genus *Guitarra*, with some of these being used as distinctive characters to separate species. With different shapes and sizes reported from each species, a spectrum of acanthoisochelae forms has now been described for this genus. A taxonomic revision of Australian species of *Tetrapocillon* also revealed a great range of shapes and sizes of, acanthoisochelae, which had previously only been reported in one species in this genus, *T. atlanticus* Van Soest. We found at least three forms of acanthoisochelae in species of *Tetrapocillon*. The first form (bipocilla-like) consists of a straight shaft supporting a concave helmet-like alae. The length and thickness of both the alae and shaft in this form is variable, from thin and long to short and thick. Clusters of spines in different stages of development are distributed either in regular or irregular patterns along the shaft and over the alae. The second form (sigmoid-like) consists of an arched shaft supporting a reduced to non-existent alae covered irregularly with clusters of long and stout spines of variable size. Included in this second form are c- to circular shapes with rudimentary alae almost connected centrally. A third transitional form has a straight shaft supporting undeveloped helmet-like alae of which the elongate extremities in some cases nearly touch each other. Different combinations of these forms of acanthoisochelae are present in the species of *Tetrapocillon* we examined. Bipocilla-like forms are found in all studied species (i.e. *T. novaezealandiae* Brøndsted, *T. minor* Pulitzer-Finali, *T. atlanticus* Van Soest and in a new species from tropical Australia). Sigmoid-like and the transitional form are observed only in *T. minor*. The variability of transitional forms within and between individuals of this latter species was remarkable. In addition, the size of all forms of the acanthoisochelae observed in *T. minor* is tiny, less than 5 µm, and they can only be seen, and studied in detail, with SEM. The great diversity of acanthoisochelae observed among species of *Guitarra* is now reported in *Tetrapocillon*. Our observations suggest that the use of this particular microsclere as a character to establish phylogenetic relationships among these genera could be debatable as these forms of acanthoisochelae are clearly either not homologous, or homoplastic. Detailed morphometric and developmental studies of acanthoisochelae would be necessary to conclude whether or not they can be used as reliable characters to distinguish species within these two sister genera and also to resolve phylogenetic relationships within the family Guitarridae.

Cyanobacteria-Sponge Symbioses: An evolutionary genomic approach

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Marine sponges harbor a diverse microbial community varying from archaea and bacteria, which constitute the greater proportion of their body biomass. Cyanobacteria represent the major and the most common members of sponge-associated bacterial community and are abundant symbionts of coral reef sponges. The study of cyanobacteria-sponge association can provide valuable insight to understand the diversity of Proto-Eukaryote symbioses. In this study we performed a genetic assessment to characterize the evolution of cyanobacteria-sponge symbioses. The intertidal sponge species *Crambe crambe* and *Halichondria* sp, belonging to the class Demospongiae, showing one of the highest distributions, constituting 85% among the total sponge diversity worldwide, were sampled at different geographical locations in Portugal. The cyanobacterial symbiont and its localization in the marine sponge *C. crambe* (Poecilosclerida) and *Halichondria* sp (Halichondrida) were examined using Fluorescent microscopy and Transmission Electron Microscopy. Unicellular coccoid cyanobacteria (*Xenococcus* like morphotypes) distribution is observed throughout the mesohyl, a central mass of cells and skeletal elements of the sponge body. The genetic relationship of symbiont and host partner was investigated using the 16S rRNA gene marker and phylogenetic tree reconstructions were based on neighbor joining, maximum-likelihood and Bayesian analyses.

A Gland-like cell-type in the Demosponge *Collospongia auris*?

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Gland cells are considered as an autapomorphy of the Epitheliozoa. Thus, sponges are regarded to posses no gland cells, and pinacocytes seems to secrete substances rather non-specifically. But some sponges secrete an epipinacodermal layer which shed from time to time. One example is the tropical marine lamellate species *Collospongia auris* (Demospongiae, Dictyocerida, Thorectidae), frequently found in aquariums. Our aim was to analyse the anatomy of *Collospongia auris*, using µCT datasets, as well as the layer and the secreting cell types, using DIC microscopy, histochemistry, CLSM, SEM and TEM. In addition we want to compare the results with former specimen of different sponge families (Order Dictyoceratida, Dendroceratida) and other basal metazoa, like Placozoa (*Trichoplax adhaerens*) and Cnidaria (*Aurelia aurita*).The exopinacoderm is interspersed with conspicuous bottle-shaped vesicular cells, reaching into cavities of the collagen-rich ECM into which small filopodia protrude; a similar type in the inner mesohyl is surrounded by numerous cyanobacteria. The pocket-like structures of the vesicular cells around the cyanobacteria suggest a strong interaction (i.e. substance exchange). We assume that the cyanobacteria provide a big part of the nutriment, because of a significant lower amount of choanocyte chambers in *C. auris* in comparison to other sponges. The vesicular cells seem to move towards the exopinacoderm. During this process the cells become flask-shaped and get in touch with the exopinacocytes. These cells completely lack cyanobacteria in their surrounding. At the contact point with the exopinacodem a small pore arises, forming an opening to the surface. The cells show a typical exocrine unicellular gland morphology, which is supported by ultrastructure: a large amount of mitochondria, distinct ER in-between vesicles and around the large nucleus, and a notable golgi complex. The sponge possess 3 different types of the vesicular cells, which vary in the diameter of the inclusions. The histochemistry of the cells and the epipinacodermal layer are shown and possible functions discussed. Whether or not this secretory cell type can be addressed as a sponge gland cell type as suggested by morphological homology criteria will have to be proven by biochemical and molecular studies.

The new species complex *Clathrina* 'brasiliensis' (Porifera, Calcarea)

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Species belonging to the *Clathrina* 'cerebrum' complex are characterized by their white cormus with regular and tightly anastomosed tubes, and skeleton composed of triactines, tripods, and tetractines with spines. Currently, four species are recognized: *Clathrina aspina* (Brazil), *C. brasiliensis* (Brazil), *C. cerebrum* (Mediterranean Sea) and *C. paracerebrum* (Western Mexico). Recently, a large international effort to inventory the markedly underestimated sponge diversity in the coastal south-eastern Pacific (Chile and Peru) and south-western Atlantic (Argentina) was carried out to figure patterns of endemism, biogeographic connectivity and biotic affinities with neighbouring faunas, such as the Tropical Southwestern Atlantic Province. Populations morphologically similar to *C. brasiliensis* were found in southern South America. Given the existence of cryptic diversity in the group, the aim of this work was to verify the taxonomic status of these populations. We performed molecular (rDNA-ITS sequencing) and morphological (spicules size, shape and distribution) analyses using 14 individuals of *Clathrina* cf. *brasiliensis* from Argentina and Chile, 12 of *C. brasiliensis* from Brazil, and as outgroups three Brazilian individuals of *C. aspina*, and three of *C. cerebrum* from the Mediterranean Sea. Molecular results (p-distance) showed that the Brazilian population supposed to be *C. brasiliensis* was separated into two clades, with p-distance of 2.6%. The populations from Argentina and Chile diverged 1.3-1.6% from the two Brazilian clades. Surprisingly, no genetic divergence between the Argentinean and Chilean populations was detected. Shape, number and distribution of spines on the apical actine of the tetractines were the most informative morphological features distinguishing the species initially called *C. brasiliensis* (Brazil) and *C.* cf. *brasiliensis* (Argentina and Chile) from all others within the *C. cerebrum* complex. However, they were not diagnostic for either the Brazilian, or the Argentinean/Chilean clades. Our results revealed the existence of a new species complex, '*C. brasiliensis*', with three cryptic species: *C. brasiliensis* and *Clathrina* sp. nov. 1, from Brazil, and *Clathrina* sp. nov. 2 from Argentina and Chile. *C. brasiliensis* and *Clathrina* sp. nov. 1 live in sympatry in Rio de Janeiro, while populations of *Clathrina* sp. nov. 2 are separated by a distance of over 3,500 km (47°S in Argentina and 23 and 33°S in Chile). The existence of the two cryptic species in Brazil can be explained by sympatric speciation. On the other hand, the cryptic species present in Argentina and Chile could have diverged from the Brazilian ones by allopatric speciation (the Prata River and the large continuous sandy coast in the south of Brazil are probably effective biogeographic barriers). The most intriguing pattern observed is the lack of any sizeable molecular divergence between Argentinean and Chilean populations. We propose two possible explanations for these results: (1) Anthropogenic transport or (2) Asexual reproduction. It is necessary to check these hypotheses in future investigations combining genetic population, ecology, reproduction, and larval behaviour studies.

Information flow through the Porifera Tree of Life project

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Large-scale projects addressing aspects of biodiversity face significant challenges in informatics. This challenge arises for two main reasons: 1) biological entities are complex, meaning that their information content is high and relatively difficult to describe, and 2) biological information is housed in resources that are distributed geographically throughout the world. The U.S. NSF-funded Porifera Tree of Life project aims to enhance understanding of poriferan phylogeny through the sampling of several thousand specimens representing thousands of taxa. This poster describes our efforts to create an efficient system for linking information generated within the project (e.g., collection meta-data, specimens, subsamples, molecular sequences, sequence contigs, morphological observations, intellectual contributions, etc.) with existing sources of information residing outside the PorToL project (such as the World Porifera Database, the Encyclopedia of Life, Sponge Barcoding Database, museum catalogs, etc.). Two guiding principals are that: 1) data be captured electronically only one time, as this is time-efficient and likely to minimize errors, and 2) the value of any particular observation is enhanced if it is readily attached to both upstream and downstream pieces of information (e.g., that a DNA extract be linked to pictures and collection data from which the original specimen was collected, as well as to sequences or phylogenetic analyses that were only possible due to the particular DNA extraction). Achieving these goals is difficult for a variety of reasons, including a lack of ability for various distributed data sources to be readily linked. We present a workflow diagram that illustrates the successes, limitations, and future development of our current system for tracking information flow through the Porifera Tree of Life project.

Structure of sponge communities in polluted waters of Rio de Janeiro, SE Brazil

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In this study sponge communities were used as biomonitors of the industrial and domestic pollution in Guanabara Bay and in coastal islands of Rio de Janeiro, Brazil (SW Atlantic). The composition and structure of sponge communities were estimated in six sites along a pollution gradient, ranging from the heavily polluted Rio-Niterói Bridge at Guanabara Bay to the slightly polluted Redonda Island 10 km offshore. Quantification was made through SCUBA diving in 20 quadrats 0.25 m^2 each per site from April 2007 to March 2008. The highest species richness and diversity were found at the intermediate site Comprida Island (25 spp., $H = 2.12$ $bits.ind^{-1}$) and the lowest were found at the most polluted site Rio-Niterói bridge (6 spp., $H = 1.30$ $bits.ind^{-1}$). Sponge density increased from 27 $ind.m^{-2}$ in Rio-Niterói bridge to 68 $ind.m^{-2}$ in Redonda Island. The most common species inside the bay (*Halichondria* sp., *Hymeniacidon heliophila* and *Cliona aff. celata*) were rare in the less polluted coastal islands, while *Tedania ignis* showed the opposite trend. These taxons can be used as indicator species in the SW Atlantic. Although the structure of sponge communities in Rio de Janeiro may also be influenced by other factors such as salinity and wave action, it is a useful indicator of the pollution levels in tropical coastal ecosystems.

Chitin in the fibrous skeleton of *Aplysina gerardogreeni* (Verongida: Demospongiae)

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The recently discovered chitin/collagen composite fiber skeletons found in the aforementioned Verongida sponge species have apparently evolved independently from Dictyoceratida and Dendroceratida. However, additional studies on the numerous other species of the order Verongida are required to provide further support to this hypothesis. Here, we presented a detailed study of the structural and physico-chemical properties of three dimensional skeletal scaffolds of the marine sponge *Aplysina gerardogreene* (Verongida: Demospongiae). Modern spectroscopic as well biochemical methods were applied in order to unequivocally prove the of alpha-chitin in investigated specimens. We show that these fibrous scaffolds have a multilayered design and are made of chitin. Chitin has been extracted from the sponge by subjecting it to the chemical treatment specified below. These extractions included step-by-step treatment as follows: an acidic extraction, an alkali-based extraction, an optional hydrogen peroxide treatment, and washing steps using distillated water before and after each treatment step. The present study of *A. gerardogreeni* revealed the presence of a chitin-based scaffold closely resembling the shape and morphology of the original sponge. Taken together with our previously published data concerning the species *V. gigantea*, *I. basta*, *A. crassa* and numerous representatives of Aplysinidae family we have now good reason to assume additionally that such chitin-based scaffolds are characteristic for the order Verongida and not just for a small number of singular species.

Indonesian anchialine lakes: natural laboratories of sponge evolution?

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Anchialine lakes are small bodies of water with no surface connection to the sea, but that have maintained a marine character through a network of small submarine connections to the adjacent sea. Anchialine lakes are unique marine habitats that could provide new insights into complex ecological and evolutionary processes in the marine realm. As a result of the relative isolation from the open sea, the anchialine lakes harbor isolated populations, endemic subspecies, and new species. The marine flora and fauna of these lakes are, however, sparsely documented. The objectives of the present study were to locate and document unexplored anchialine lakes in Indonesia and to record the sponge species within these lakes. In parallel to the sponge community survey we used molecular analyses to assess the level of genetic connectivity and divergence between the different marine lakes and the outside sea. Surveys of the lakes and adjacent coastal habitats showed that algae and sponges were the most dominant taxa in terms of biomass and diversity. Over 1000 sponge specimens, belonging to at least 60 genera, were collected from anchialine lake, mangrove, and reef systems located in East Kalimantan and West Papua in Indonesia. The observed sponge species could be divided into three groups (1) species restricted to one lake (2) species shared by almost all lakes, but unknown from the adjacent sea, and (3) widespread species, known from various locations in the Indo-Pacific reefs. In this presentation we will discuss our preliminary results which show some analogies to island systems in terms of species-area relationships, possible high endemicity, and genetic isolation of populations.

Marine island life? On the phylogeography of sponge populations isolated in anchialine lakes

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Anchialine lakes are small bodies of seawater that are entirely surrounded by land and have maintained a marine character through narrow subterranean crevices or porous stone to the open sea. Anchialine lakes are unique marine habitats that could provide new insights into complex ecological and evolutionary processes in the marine realm. As a result of the relative isolation from the open sea, the anchialine lakes harbor isolated populations, and endemic subspecies. The existence of multiple independently derived populations in anchialine lakes and of their putative ancestral populations in the adjacent sea provide an unique opportunity to study the evolutionary history as well as evolutionary rates of ecologically important groups such sponges. This study has investigated four key sponge taxa, the dendroceratid *Darwinella aff. gardineri*, the hadromerid *Suberites diversicolor*, the poecilosclerid *Biemna aff. fistulosa*, and the spirophorid *Cinachyrella aff. australiensis*. We sampled populations from four anchialine lakes in Indonesia (two in East Kalimantan and two in West Papua) that flooded during the Holocene with different dates of flooding from 8000 to 5000 years ago. We have used molecular analyses to assess the level of connectivity and divergence between the different anchialine lakes and the outside sea. We will present preliminary data addressing the following questions: (1) To what extent are sponge populations in anchialine lakes isolated from eachother and from the populations outside in the adjacent sea? (2) Whether there is evidence for parallel evolution of sponge taxa in the different anchialine lakes?

From dark chocolate to café au lait – how many flavors of *Placospongia* (Hadromerida: Placospongiidae) do we have in the Indo-Pacific?

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Species belonging to the genus of *Placospongia* are common within tropical waters of the Indo-Pacific occurring in a wide array of dark-chocolate to crème colorings with either branching or encrusting growth forms. Presently there are three valid species in the Indo-Pacific: *Placospongia carinata* (Bowerbank, 1858) (type locality 'South Sea') *Placospongia melobesioides* Gray, 1867 (type locality Borneo), *Placospongia anthosigma* (Tanita & Hoshino, 1989) (type locality Sagami Bay, Japan). In 1900 Thiele described *Placospongia mixta* from Ternate (Indonesia), which subsequently was synonymized with *P.carinata* by Vosmaer & Vernhout in 1902. To determine the taxonomy of the species of *Placospongia* in the Indo-Pacific I examined the holotypes of these four species and over 200 specimens of *Placospongia* collected from various locations in Indonesia (including the Vosmaer & Vernhout material), Seychelles, India, and Singapore. Based on this extensive examination I conclude that *P.mixta* is in fact a valid species. All *Placospongia* species are characterized by selenasters and tylostyles in two size-classes: the large with blunt points, the small with sharp points. *Placospongia* melobesioides is distinguished from the other species by an absence of spirasters and microrabds, and the presence of spherules. *Placospongia mixta* and *P.carinata* both contain spirasters and microrabds, and are differentiated from each other by the length of the tylostyles, the shape of the spirasters and the presence/absence of spherasters. In addition to these three species, I have found an undescribed species containing microrhabds, but not spirasters, spherules or spherasters. The four different species could not be distinguished by color or growthform. *Placospongia* melobesioides and *P.mixta* are common in the reef environment, while *P. carinata* appears to be very rare in the reefs, but is highly abundant in the marine lakes Haji Buang and Kakaban in East Kalimantan, Indonesia. *Placospongia* anthosigma was not found in any of the examined collections from the tropical Indo-Pacific, this species may be restricted to more temperate waters. In conclusion, the grand total for the Indo-Pacific is at present five species of the genus *Placospongia*.

Distributional patterns and spatial interactions between sponges and macroalgae at New Zealand rocky reefs

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Sponges are effective spatial competitors, together with other invertebrates such as cnidarians and tunicates. Although sponges are abundant in many habitats, under some circumstances they appear to be restricted to cryptic habitats, particularly in high-light areas that are dominated by macroalgae. Interactions and spatial competition between sponges and algae are not clearly understood, nor is the reason why sponges are restricted to cryptic habitats in the presence of macroalgae. The aim of our work is to assess if the spatial distribution patterns of sponges are dependent on algal populations. Subtidal sampling was conducted in a variety of zones at a number of sites in New Zealand including an assessment of the environmental factors at each site. A series of 0.25 m^2 quadrats at 5 m and 15 m depth were placed on different surface angles covering a range of surface inclinations (horizontal, inclined, vertical and overhanging surfaces). Other parameters, such as substrate rugosity, turbidity and physical characteristics of water column, were recorded at each site. The percent cover of first layer (canopy-cover) together with the coverage of sponges, algae and other encrusting organisms were estimated in each quadrat. We evaluated the differences in coverage of organisms between sites, depths and inclinations. In general, the percentage cover of sponges was higher with increasing depth. Macroalgae dominated the shallowest depth (5 m) on the majority of reefs, except at sites with steep walled channels were macroalgae were virtually absent and sponges dominated the substrate. Data from the different sites are presented in order to examine differences in the observed patterns at local and regional scales in order to better understand the factors controlling sponge distribution and abundance patterns.

Interactions between nudibranchs and sponges in Rio de Janeiro State, Brazil (SW Atlantic)

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Nudibranchs are well known as the main invertebrate predators of sponges, but other aspects of the interactions between nudibranchs and sponges such as egg deposition and camouflage remain comparatively less studied. Of the 103 species of nudibranchs known from Brazil, 40 are listed from Rio de Janeiro state, but they are known only from taxonomic studies, with little information on their ecology. The goals of this study were to determine the types of interactions between nudibranchs and sponges at the coast of Rio de Janeiro state, SE Brazil; to identify the interacting species; and to determine the degree of specificity of these interactions. Field observations and collections were conducted through free diving and SCUBA diving in 2008-2009 in five localities: Angra dos Reis, Rio de Janeiro, Arraial do Cabo, Cabo Frio and Búzios. We found 16 species of nudibranchs, 13 of which (61 individuals) belonging to the suborder Doridina and 3 species (6 individuals) of the order Aeolidina. All Doridina species were associated to sponges (18 species), while the Aeolidina were associated to tunicates. Predation was the most frequent type of interaction between nudibranchs and sponges (77% of the species). Nine nudibranch species (70 %) used sponges as substrates for egg deposition. Two species (15%) were camouflaged on the sponges, and five (38%) showed aposematic coloration. Three associations were very specific, viz., *Chromodoris binza* on *Chelonaplysilla erecta*, *Hypselodoris lajensis* on *Dysidea etheria*, and *Rostanga byga on Mycale microsigmatosa. Jorunna* sp. preyed upon four species of the order Haplosclerida, and five species of nudibranchs showed non-specific interactions. As in other regions of the world, such as the Indo-Pacific, sponges in SW Brazil are of fundamental importance for the biology of dorid nudibranchs, which use them as food, substrate for reproduction and protection against predators.

Sponsors: CNPq, FAPERJ, Petrobras.

Characterization of the culturable sponge-associated bacterial consortium from two marine sponges: *Diacarnus erythraeus* and *Theonella swinhoei,* as first steps towards obtaining bioactive marine natural products

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Since the 1970s more than 20,000 structurally diverse compounds have been discovered in the marine environment, with sponges (Porifera) being one of the most prolific sources. Many of these compounds have been shown to possess a wide variety of bioactivities (e.g., antibiotic, anti-tumor, antifungal, anti-parasite, and antiviral). However, concentrations of the desired compound in these organisms are usually low, which frequently hampers the possibility of production on a commercial scale. Sponges are known to harbor diverse communities of bacteria, and a growing body of evidence indicates the involvement of these 'sponge-associated bacteria' in the production of some of the compounds. These sponge-associated bacterial communities include Actinobacteria, which are known to be a rich source for novel drugs, with more than two-thirds of known naturally-occurring antibiotics produced by one order (Actinomycetales). It is therefore no surprise that Actinobacteria are of great interest to the pharmaceutical industry and immense efforts are being made to isolate novel representatives of this group for natural product research. We examined the bacterial community structure of two sponge species, adult *T. swinhoei*, and adult and larvae *D. erythraeus.* The species were harvested from the natural coral reef and from a marine-based culture facility, and were chosen for their metabolite content. We analyzed the culturable and unculturable bacteria, by comparison of 16S rRNA gene sequence profiles of cultured bacterial isolates, and clone libraries. Here we report on the culturable isolates. Special emphasis was placed on culturing Actinobacteria. Phylogenetic analysis of identified bacteria was performed by constructing phylogenetic trees (by class) at the genus level. Overall, 303 isolates were identified, using the 16S rRNA gene. Phylogenetic analysis revealed their grouping into five classes: Actinobacteria, Bacilli, Alphaproteobacteria, Gammaprotobacteria and Flavobacteria. Although the bacterial community cultured from these two sponge species did not differ by class, some differences were noted at the genus level. In both sponges, the Actinobacteria made up the largest number of cultured isolates and showed the greatest diversity. A total of 127 (80 from *T. swinhoei*, 34 from adult *D. erythraenus*, and 13 from its larvae) actinobacterial isolates, belonging to 13 families and 16 genera; 36.8% of *D. erythraenus* isolates, represented by nine different genera and 45.7% of *T. swinhoei* isolates represented by thirteen different genera. *Micrococcus sp.* was the most abundant genus cultured (20% of total isolates in *D. erythraenus* and 36.2% in *T. swinhoei*). Isolates were cultured, frozen and await high-throughput screening (HTS) in future research.

Marine-based cultivation of sponges for obtaining bioactive marine natural products

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The marine environment has yielded numerous compounds, with sponges (Porifera) providing one of the most prolific sources. These have been shown to possess a wide variety of bioactive properties (e.g., antibacterial, anti-tumor, anti-inflammatory, anticoagulant, anti-malarial, and antiviral). The discovery of such properties has revealed great potential for biotechnological applications, including development of novel drugs for pharmaceutical purposes. However, the desired compounds are usually present in minute concentrations, and synthesis is frequently highly complex and expensive. Harvesting wild sponges is also unrealistic. This 'supply problem' hampers the possibility of production on a commercial scale. One suggested solution to this problem is that of sponge biomass culture on artificial structures. The simplest and most cost-effective method is the culture of sponge fragments in a marine-based system. Growth rate and survival may vary, depending on the species, season, and mariculture conditions. Sponges grown under different conditions have been shown to yield different quantities of the desired metabolite. We evaluated the compatibility of four demosponge species (*Diacarnus erythraenus, Theonella swinhoei, Negombata magnifica* and *Amphimedon chloros*) for marine culture. All species were chosen for their metabolite content. Fragments of all sponges were maricultured on two artificial structures (frames), at two depths (10m and 20m). Commencing in December 2005, the experiment lasted 767 days; during which the fragments' mean growth rates, survival, and the presence of the desired compound (NMR analysis) were monitored every 6 months. The results revealed very high survival rates for *D. erythraenus*, with 72% of all individuals surviving the entire duration of the experiment at 10m depth, and 70% at 20m. *N. magnifica* survival rates were 58% at 10m and 54% at 20m after 18 months; however, 82% of these later died within a month as a result of an infection (August 2007). *T. swinhoei* and *A. chloros* survival rates were much lower, with only 17% and 18% (respectively) of all individuals surviving the duration of the experiment. Growth rates of the last two were also low (mean±S.E, averaged for both depths), and stood at -27%±8% and 52%±22% (respectively). *A. chloros* and *T. swinhoei* were found to be unsuitable for culture. In contrast, the highest annual growth rates were measured for *N. magnifica* (308.6%±41% at 10m and 298.4%±39.7% at 20m), with no significant differences between depths (P=0.475). *D. erythraenus* had significantly lower growth rates (60.1%±18.3% at 10m and 76.1%±18.8% at 20m, P< 0.001). Chemical analysis (by NMR) showed that all sponges produced the desired secondary metabolites. In addition, no significant differences (P=0.103) were found in Latrunculin B content, between fragments of wild *N. magnifica* (0.71% of dry weight) and maricultured ones (0.91% at 10 m and 0.96% at 20 m). The results of this study show the potential of two of the sponges (*D. erythraenus* and *N. magnifica*) for mariculture, with the latter providing greatest promise.

Are sponge biodiversity patterns consistent with the bioregions proposed to represent different areas of marine biodiversity in New Zealand?

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Preserving biodiversity is important for maintaining ecosystem functionality and stability. The New Zealand biodiversity strategy (2000) aims to create a network of Marine Protected Areas in order to protect a proportion of all the marine habitats present in New Zealand and the biodiversity within them. It has been suggested that mainland New Zealand can be divided into two biogeographic provinces, which can be split further into eleven bioregions. Bioregions cover relatively large areas of water, and contain characteristic, geographically distinct assemblages of natural communities and species. They are developed to aid in biodiversity conservation planning. New Zealands marine bioregions have been designed using macroalgal communities and large-scale benthic structural groups. This work investigates if the boundaries of the regions are appropriate for sponges, which are the largest contributor to total biomass for encrusting invertebrates in New Zealand. The patterns of sponge biodiversity were compared at the species, genus and family level using the average taxonomic distinctness between bioregions, and also the variation in taxonomic distinctness from the PRIMER statistics package. This method is particularly applicable to historical presence absence data sets across space and time because it is less affected by the variation in sampling effort around the country. Over 800 sponge species and over 3000 sampling periods were included in the data from all over New Zealand waters to a depth of 200m. Sponge biodiversity varied significantly over depth and matched the biogeographic regions and provinces to some degree however there were also many small areas found to be hotspots of biodiversity within bioregions.

Patterns of temporal sponge assemblage variability on the Wellington South Coast.

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To date, there have been conflicting reports regarding the stability of sponge assemblages, for example, past research from the Mediterranean, the Atlantic and Caribbean has shown sponges to be temporally stable, yet more recent work from the Atlantic, Caribbean and the Pacific suggest that more rapid changes in sponge assemblages are common. Here we compare the patterns of to determine if there are any consistencies in the rates of assemblage change at seasonal and inter-annual scales along the Wellington South Coast and we describe how identifying consistencies or differences in assemblage variability can help understand the factors controlling patterns of natural variability. Sixty-five species were found along the Wellington South Coast with forty three found within the permanent survey areas. There were significant levels of spatial variation between the locations surveyed even though they were all at the same depth and aspect, species richness and substrate type. There were no significant seasonal variations, however there was a tendency for fewer sponges to be present at all three sites during the summer possibly due to spatial competition with ascidians, algae and other dominant spatial competitors, These data will be compared with that from other regions in order to examine the consistency of the relationships.

Sponge assemblages in the mesophotic zone of the Tyrrhenian and Ionian Seas

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Remotely Operated Vehicle (ROV) exploration has opened new perspectives in the study of deep benthic fauna, providing new and detailed physical records and allowing the qualitative and quantitative description of benthic assemblages. Our research has focused on the sponge assemblages of the mesophotic zone of some Tyrrhenian and Ionian rocky bottoms as the Vercelli Seamount (North Tyrrhenian Sea) and several areas along the Calabrian coast. The term mesophotic or twilight zone refers to the bathymetric belt extended from 40 to 150 m depth, in the lower portion of the photic zone. The identification of deep sea sponges is virtually impossible if based only on videos and photographs, but ROVs may offer the opportunity to collect samples and to associate them to peculiar habitat. On the Vercelli seamount, characterised by a rich coralligenous community ranging from 60 m to 140 m depth, sponge distribution seems constrained by the hydrological conditions found along the two flanks of the ridge. While sponges of the genus Axinella are uniformly distributed along the two sides, *Tethya citrina*, a species generally recorded in habitats with high sedimentation rates, as coastal lagoons or fiords, is present only on the northern side of the ridge, characterised by downwelling conditions and high turbidity levels. On this side of the seamount, from 70 to 80 m depth, *T. citrina* shows a very dense population with a maximum of 8 specimens·m^{-2}. Also encrusting sponges show their highest per cent cover values (from 65 % to 75 %) on this northern side between 70 m and 100 m depth. On the gentle slope of rough detritus at the base of the Vercelli Seamount's coralligenous pinnacle (around 180-200 m depth) very few species form the benthic assemblage. Here it is common to encounter the blue encrusting sponge *Phorbas tenacior*, typically living on the rocks. *Oceanapia* sp. lives on the dead branches of the yellow scleractinian coral *Dendrophyllia cornigera*. In the Gulf of Santa Eufemia (Thyrrhenian Sea) we have explored a group of small rocky shoals arising from a muddy flat bottom to about 120 m depth. These shoals host a rich coral assemblage composed by sea fans and black corals. *Haliclona* sp. which displays white tubes, is the most characteristic sponge species of this area. On the sandy bottom around the shoals thrives a mixed assemblage of pennatulaceans and the stalked *sponge Rhizaxinella pyrifera*. The hard bottom assemblages of the Ionian side of the Calabrian coast show a scarce coral component whereas sponges are locally extremely abundant. Especially in the area, around 150 m depth, of the Amendolara Bank, fan shaped sponges, such the orange *Pachastrella monilifera* and the white *Poecillastra compressa*, recorded on a peculiar tabular beach rock, are the only living assemblages.

Posidonia oceanica meadows as sponge spicule traps

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Dense meadows of the sea-grass *Posidonia oceanica* are widely diffused in the Mediterranean Sea. It is known that the friction on the leaves slows water movement producing an intense deposition of the suspended sediment. The *Posidonia* plant faces the sedimentation by a continuous vertical growth of its rhizome with an average rate of about 1 cm·year^{-1}. Therefore *Posidonia* meadows represent a sort of natural sediment trap and their vertically growing mats are reservoirs of recent sediment whose age can be grossly determined. It is generally assumed that global warming trend of the last decades has deeply changed the structure of benthic communities. Nevertheless historical quantitative data about the composition of benthic communities are rare and confined to the last two-three decades. In this work we have conducted a quantitative study at Prelo Bay (Ligurian Sea) on the sponge spicules present in core samples taken from a 1.5 m thick living mat of *Posidonia*. All spicule fragments, present in a standard amount of sediment, at different levels of the core samples, were counted, measured and, when possible, attributed to a specific spicule type. The amount of spicules at the different levels of the core samples was assumed as directly related to the sponge abundance in the surrounding area of the meadow during the deposition period. Due to the low pH conditions of the sediment in the mats it is possible to suppose a good preservation of the biogenic silica. The number of recorded fragments ranged from about 7000 g^{-1} of sediment in the upper part of the core sample, close the surface of the mat to 18000 g^{-1} of sediment in the deepest portions. The silica volumes calculated at the same layers range from 3.5•106 to 2•107 µm^3/g of sediment with an increasing of more than five times. The data show a slow rising trend from the surface to 100-120 cm depth followed by a drastic increase in the deepest fractions. The comparison of the sediment granulometry at the different levels of the core samples does not show significant differences. This fact suggests that the higher number of spicules recorded at the deepest layers is actually due to an increased environmental supply rather than to a physical sinking through porous sediment. Since spicules present in the sediments are generally broken they are unuseful to identify the source species. Nevertheless, considering the percentage of the two main spicule kinds recognisable in the sediments, oxeas and tylostyles, it is possible to observe an inversion of their ratio from the sediment surface (40% oxeas, 60% tylostyles) to the deepest layer of the core sample (70% oxeas, 30% tylostyles) suggesting past differences in the species composition of the assemblage. On the base of this evidence it is possible to hypothesise that about 150 years AP the sponge assemblage of the Prelo Bay was significantly more abundant that nowadays. Further data from other *Posidonia* meadows close to Prelo Bay are necessary to test our finding.

The paleo sponge assemblage of the Mediterranean coralligenous

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Coralligenous is a Mediterranean hard substrate of biogenic origin mainly composed of calcareous algae growing in dim light conditions and sediment compacted. Coralligenous communities are the most important 'hot spots' of biodiversity in the Mediterranean together with *Posidonia oceanica* meadows. Until now few radiocarbon dating of the Mediterranean coralligenous are available but it is generally accepted that these concretions had their maximum development in a period comprised between 8000 and 5000 years ago. After that period, appreciable accumulation rates are recorded only for concretions situated in relatively shallow waters (10 to 35 meters depth), whereas the accumulation rates of coralligenous deeper than 50 meter are almost nil. Sponges are one of the most representative organisms of the coralligenous communities with more than 140 recorded species with different habits: branching, massive, encrusting, insinuating and boring. When boring and insinuating sponges die their siliceous spicules, remaining trapped inside the concretion, offer the unique possibility to describe the spongofauna of coralligenous during a very long time span, virtually extending to a large part of the holocenic period. Data here reported were obtained from two blocks of coralligenous collected by diving at 35 m depth from the Secche di Santo Stefano and the Portofino Promontory (Ligurian Sea). Twenty-five cm long cores, spanning through the entire thickness of the blocks, were divided into 1 cm long portions extracting then by hydrogen peroxide the sediment trapped in each portion. The recorded spicules show deep dissolution marks in form of circular holes on their surface or present an enlargement of the axial canal. However their original shape, generally intact, suggests the absence of mechanical injuries and may allow the identification of sponge species. The analysis of these old spicules and the comparison with those obtained from the sponges living on the block, allowed to recognise a "paleoassemblage" of boring and insinuating sponges composed by 28 recognisable species (16 in the block of Portofino, 21 in the block from Secche of Santo Stefano, 13 in common). The main bio-eroding agents recorded in the two blocks were *Cliona janitrix* and *Spiroxya heteroclita*. The most diffused insinuating species are *Stoeba plicata*, *Diplastrella bistellata*, *Jaspis johnstoni*, *Geodia* sp., *Timea* sp. Inside the conglomerate also spicules belonging to massive sponge were recorded showing differences from the two localities. In the block from Portofino, spicules of *Petrosia ficiformis* were widely recorded while, in the other conglomerate the achantostyles of *Agelas oroides* were abundant. The comparison of the paleo sponge assemblage reconstructed through the spicules trapped in the conglomerate with that recorded alive on its surface, indicate that the sponge community of Mediterranean coralligenous is very old and stable from a very long span of time.

Spongofauna diversity of the coralligenous in the Ligurian Sea (North Mediterranean Sea)

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Coralligenous concretions typically developing in dim light conditions represent the most important 'hot spots' of biodiversity in the Mediterranean Sea. Sponges are the group showing the higher number of species living on these biogenic structures. In addition to the conspicuous, relatively well known, species settled on the surface of the conglomerate there is a large number of small cryptic species, both insinuating and perforating, very poorly known. Aim of this work was to study the diversity of the coralligenous spongofauna in four sites of the Ligurian Sea. Samples from the coralligenous, were collected in the Marine Protected Area of Portofino (Genoa) (Punta del Faro, stations NW and S), Punta Manara (Genoa) on the eastern coast and Gallinara (Savona) (Falconara and Sciusciaù stations), Santo Stefano (Imperia) on the western coast, all between 30 and 40 m depth. Four blocks, with an average volume of about 20 l, were collected for each site. All the sponge species settled on the surface of these blocks were sampled and identified. Moreover two rocks per station were cut into slices about 2 cm thick to allow the identification of the endolithic species. A complete revision of the available literature indicates that, until now, 265 sponge species have been recorded from the Mediterranean coralligenous concretions. During the present study we have recorded 107 sponge species. At the MPA of Portofino 59 records were performed, 54 at the Gallinara Island, 53 at Punta Manara and 37 species at Santo Stefano. Among massive or incrusting species, eight (*Clathria* (*Microciona*) *armata*, *Clathria* (*M.*) *haplotoxa*, *Eurypon denisae*, *Forcepia* sp., *Hymedesmia* (*Hymedesmia*) *rissoi*, *Phorbas mercator*, *Haliclona* (*Gellius*) *marismedi* and *Haliclona* (*Reniera*) cf. *griessingeri*) represent new records for the coralligenous sponge fauna. Moreover *Clathria* (*M.*) *armata*, *Eurypon coronula*, *Eurypon denisae*, *Bubaris carcisis*, *Haliclona* (*G.*) *marismedi* and *Haliclona* (*R.*) cf. *griessingeri*) represent new findings for the Ligurian Sea. Among the endolithic spongofauna, 12 insinuating and perforating species *Stelletta mediterranea*, *Stelletta stellata*, *Pachastrissa* sp., *Stoeba* sp., *Triptolemma* sp., *Cliona burtoni*, *Paratimea oxeata*, *Acarnus souriei*, *Hymedesmia* (*Hymedesmia*) *baculifera*, *Plocamionida ambigua*, *Merlia normani* and *Haliclona* (*Gellius*) bioxeata are new discoveries for the coralligenous community.*S. mediterranea*, *S. stellata*, *C. burtoni*, *P. oxeata*, *A. souriei*, *M. normani* and *H.* (*G.*) *bioxeata* are new records for the Ligurian Sea. *Cliona viridis*, *Jaspis johnstoni* and *Stoeba plicata* are the species reaching the greatest depths inside the concretion penetrating up to 5 cm into the rock. Our data showed that in all the sites sponge abundance progressively increases from the exposed to the shady side of the concretion. Biodiversity indexes confirm a higher diversity on the shady side of the coralligenous concretions. The obtained data increase to 275 the species of the spongofauna associated with coralligenous concretions and confirm that this habitat is an extraordinary reservoir of biodiversity still largely unexplored, not only at the species level, but also regarding specific adaptations and life histories.

Variability of the chemical composition in *Lamellodysidea* sponges from Moorea.

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The tropical marine sponge *Lamellodysidea herbacea* is always found associated with the filamentous cyanobacterium *Oscillatoria spongeliae*, which occurs abundantly in the sponge mesohyl, and with numerous bacteria, both intracellular and extracellular. *Oscillatoria spongeliae* is suspected to be responsible for the production of polychlorinated peptides and polybrominated diphenyl ethers (PBDEs) in the sponges but these two families of halogenated compounds have never been found together in the same specimen. Hence marine sponge *L. herbacea* occurs in 2 chemotypes, one containing polychlorinated peptides and the other polybrominated diphenyl ethers. The variability of the chemical composition in *Lamellodysidea* sponges can be explained by the genetic variability of the sponges and its symbionts but also by the influences of environmental factors (ecological, geographical, seasonal factors…). In order to better understand the chemical variability, about seventy specimens of *L. herbacea* were collected all around Moorea Island (French Polynesia) in different locations, environments, seasons, and were analyzed by HPLC. All specimens contain 3,5-dibromo-2-(3',5'-dibromo-2'-methoxyphenoxy)phenol (4Br-OH-OMe-PBDE) as major compound. Concentrations of 4Br-OH-OMe-PBDE have been compared to concentrations of chlorophylle a (chloro a) to estimate if PBDE's rate can be related to the chlorophylle present in the symbiotic cyanobacterium. The correlation between the two concentrations is positive throughout the geographical locations, both varying significantly depending on the season and the environment. Few specimens of *L. herbacea* were found living overlapping the algae *Caulerpa racemosa*. These specimens show a different chemical fingerprint with a significantly different rate of 4Br-OH-OMe-PBDE and appearance of another PBDE, 4Br-H-OH-PBDE. The addition of a photosynthetic inhibitor (DCMU) on sponges transplanted in tanks, has induced, after a week, a degradation of the sponges and a decrease of levels of 4Br-OH-OMe-PBDE while dibromo and tribromo analogues appear. Our results suggest that variations in the levels of PBDEs and symbionts may be related. We also observe a chemical answer of the symbiotic association *L. herbacea* – *O. spongeliae* to biotic (competition with alga) and abiotic (pollutants) stresses, quantitatively (by overproduction or underproduction of 4Br-OH-OMe-PBDE) and qualitatively (by production of 4Br-OH-OMe-PBDE's analogues).

Study on bisilisification based on the recombinant silicatein of marine sponge *Hymeniacidon perlevis*

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Sponges (Porifera) are the oldest living metazoan in the world, among which most of them (Demospongia) can produce silicic skeleton from orthosilicic acid in the seawater under the natural environmental conditions. These biosilicic materials exhibit good mechanical and optical properties as well as good biocompatibility. During the biosilicification process of sponges, a protein, named as silicatein, plays an important role and has attracted great attention from biologist, chemists and material scientists. But till now, no direct proof had been report for the mechanism of the biosilicification catalyzed by silicatein from the view of protein structures. Now we try to use the recombinant silicatein of *Hymeniacidon perlevis* as model to give adirect information about the relationship between the structure and activity of it.

Disrupted spiculogenesis in deep-water *Geodiidae* (Porifera, Demospongiae) growing in shallow waters

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The environment has been shown to influence sponge morphology and distribution. Depth is particularly known to influence morphology through the synergistic action of light, temperature, silica concentration and/or wave action. Boreo-arctic *Geodiidae* (Demospongiae, Astrophorida) are mainly deep-water species growing on hard-bottoms. They are commonly collected and observed at depths ranging between 100 and 400 m. However, in fjords and along the Norwegian coast, *Geodia barretti* Bowerbank, 1858 and *Pachymatisma normani* Sollas, 1888 can occasionally be found at shallower depths. In this study, we examine morphological differences of shallow *G. barretti* and *P. normani* with respect to their deep-water counterparts. Shallow specimens had an identical cytochrome c oxidase subunit I (COI) gene with deep-water *G. barretti* and *P. normani*. We found phenotype differences with respect to 1) color, 2) external morphology, 3) cortex organization and above all 4) spicule morphology. Shallow specimens of both species had a softer, thinner and more flexible cortex. Scanning electron microscope observations of spicules showed that the spiculogenesis of microrhabds, sterrasters and triaenes was disrupted: these spicules are smaller, irregular and/or underdeveloped. Oxyasters and strongylasters are normal, albeit smaller. The actines of sterrasters are not fused and covered with silica microspheres or short spines. We conclude that *Geodia simplicissima* Burton, 1931 — originally collected at a depth of 10-75 m in Northern Norway, and never encountered since — is actually a shallow *G. barretti*, it is here therefore considered as an invalid species. We hypothesize that the lower silica concentration in shallow waters is responsible for the disruption of spiculogenesis in *G. barretti* and *P. normani*.

Towards a phylogeny of *Ancorinidae* (Demospongiae, Astrophorida)

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The order Astrophorida is characterized by possession of triaenes and oxeas as main megascleres, frequently in association with asterose and/or rhabdose microscleres. All of these may be absent, such that different combinations of these spicules and their morphologic spectrum allow recognition of many genera. The absence of triaenes is common in *Ancorinidae* for instance (*Asteropus*, *Holoxea*, *Jaspis* and *Melophlus*), which is by far the largest family in the order, and a natural target for a preliminary phylogenetic exercise aiming at establishing a framework of higher taxa relationships within the order, to be later contrasted to molecular evidence in search of corroboration. This exercise is also important to verify whether the old concept of *Coppatiidae* could be applied in case genera lacking triaenes would come out as monophyletic. Characters used in the analyses were chosen from the external and internal morphology of hypothetical OTUs representing the spectrum of variation of morphologic characters for all the 15 genera included in the family: (1) habit – A. no projections, B. with projections. (2) texture – A. slightly hispid, B. very hispid. (3) microscleres crust – A. present, B. absent. (4) Oxeas radially disposed – A. only at the periphery, B. all over the skeleton, C. absent. (5) Maximum size of oxeas – A. >1000µm, <2000µm; B. >2000µm; C. <1000µm. (6) triaenes – A. presence of ortho/plagiotriaenes, B. presence of anatriaenes, C. presence of dichotriaenes, D. absence of triaenes. (7) euasters – A. present, B. absence. (8) streptasters – A. present, B. absent. (9) microrhabs – A. present, B. absent. (10) trichodragmas – A. present, B. absent. The phylogenetic analyses were performed with PAUP 4.0*, using the "Branch and Bound" exact algorithm. Characters were treated as unordered and equally weighted, but subsequent weighting on their RC was applied. Two analyses were performed, the first one with Calthropellidae as outgroup and the other one with Geodiidae. These families were selected in view of their possession of euasters as microscleres, a trait shared with Ancorinidae, which contrasts the distinct nature of microscleres in Pachastrellidae and Thrombidae. The tree obtained with Geodiidae as the outgroup was more resolved and is transcribed here: (((((((((*Disyringa*, *Tethyopsis*) *Tribrachium*) *Psammastra*) *Ecionemia*) (*Holoxea*, *Melophlus*)) *Penares*) ((*Asteropus*, *Stryphnus*) *Ancorina*)) (*Jaspis*, *Rhabdastrella*)) *Cryptosyringa*) *Stelletta*). Important congruences were observed on both analyses though. Both majority-rule consensus hold that *Jaspis*/*Rhabdastrella*, *Asteropus*/*Stryphnus*, *Disyringa*/*Tethyopsis*, and *Holoxea*/*Melophlus* are sister pairs of taxa. *Ecionemia* is the sister of (((*Disyringa*, *Tethyopsis*) *Tribrachium*) *Psammastra*) and *Ancorina* the sister of (*Asteropus*, *Stryphnus*). In neither tree genera lacking triaenes formed a clade. Important incongruences are abundant too. Among these, the opposite allocation of *Stelletta* is the most remarkable: the basal most taxon when *Geodiidae* is the outgroup, and a rather derived placement when outgroup is *Calthropellidae*.

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Growth and mortality of early juveniles of the exotic sponge *Paraleucilla magna* (Porifera, Calcarea) from Brazil

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The sponge *Paraleucilla magna* is an exotic species in Brazil, although originally described from Rio de Janeiro. Since its introduction, probably in the nineties, it became very abundant. Nowadays, P. magna is find from Rio de Janeiro to Santa Catarina, in the south of Brazil (1,400 km apart), and in the Mediterranean Sea. In the present work, we analyzed the growth rate and mortality of early juveniles of *P. magna*. The study was performed in Arraial do Cabo (Brazil), from September to December 2009. Granite plates were used as substrate for the larvae. They were kept inside of boxes at 1 m of depth, protected from sunlight and sediment. A transparent grid was used to record the position of each juvenile on the plates. Weekly, measurements (base and height) of the sponges were taken with a ruler. When a sponge disappeared, it was considered dead. During the study, a total of 72 juveniles of *P. magna* were monitored. The first juveniles were found three weeks after the beginning of the experiment. Our results showed that mortality was higher during the first two weeks of life, as 43% (31 specimens) of the analyzed sponges died after one week and 21% (15 specimens) died after two weeks. The mortality rate decreased as the sponges became older, but only two specimens survived through the whole experiment, completing eight weeks of life. During the study, some specimens showed a reduction in their body volume. The growth rate increased with time, going from 0.59 (±61617; 0.49) mm^3.day^{-1} in the first week to 173.6 mm^3.day^{-1} in the last (8th) week. Volume dynamics (considering both growth and reduction) was also calculated for the two specimens that completed eight weeks of life. At the beginning, both presented the same volume (1.57 mm^3), but in the second week, one of them was 11.78 mm^3, while the other was 2.65 mm^3. This difference became higher along the weeks, but in the eighth week, the largest specimen reduced from 2,935.9 to 1,884.0 mm^3, while the smallest one increased from 2,245.1 to 3,460.3 mm^3. Studies focusing on the growth and mortality of calcareous sponges are very scarce, and growth rates of juveniles had never been calculated before. High mortality rates in the beginning of life had already been observed to *Demospongiae* species. To *P. magna* it was caused in part by grazing of sea urchins and competition with colonial ascidians and bryozoans.

Taxonomic revision of the genus *Leucascus* (Porifera, Calcarea) with revalidation of the genus *Ascoleucetta.*

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The genus *Leucascus* is composed of sponges with anastomosed tubes, cortical membrane and atrium delimited by pinacoderm. Recently, five species belonging to different genera were transferred to *Leucascus*, and several others were suggested but not formally included in this genus. In the present work, all species accepted or suggested as *Leucascus* were revised. For *Leucascus amitsbo* and *Leucascus soyo* taxonomic considerations were made based only on their original descriptions, as their type material was lost. To all the other species, spicule and histological slides were prepared according to standard procedures. Scanning electron microscopy was also applied to the analysis of the apical actine of the tetractines. According to our results, the following species should be excluded from the genus *Leucascus*: *L. amitsbo*, *L. compressa* and *L. ventricosa* (that should be transferred to the genus *Ascoleucetta*), *L. lobatus* (that should be included in a new genus), and *L. soyo* (that should be reallocated back in the genus *Leucettusa*). On the other hand, Leucetta leptoraphis should be included in Leucascus. Hence, the genus should be composed of seven species: *L. clavatus*, *L. leptoraphis*, *L. neocaledonicus*, *L. protogenes*, *L. roseus*, *L. simplex* (the type species) and *Leucascus* sp. nov. The presence of spines in the apical actine of the tetractines had never been observed before in *Leucascus*, although it showed to be a consistent character for the genus, as all species with tetractines presented spines. The revalidation of *Ascoleucetta* is being suggested, based on differences in the cortical skeleton composition and arrangement. Modifications were also made in the definition of both genera and of the family Leucascidae, which by our proposal should be composed of *Ascaltis*, *Leucascus*, and *Ascoleucetta*.

Solenoid: a new aquiferous system to the Phylum Porifera

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The aquiferous system is the main synapomorphy of the phylum Porifera, nonetheless, very few is known about its evolution. Calcarea is the class of sponges with the greatest variety of aquiferous systems: asconoid, syconoid, sylleibid, and leuconoid, and probably even others. Although no deep phylogenetic significance has been attributed to them, the aquiferous systems have great importance for the taxonomy of Calcarea. In the present work we describe a new type of aquiferous system found in the genera *Leucascus* and *Leucaltis*: the solenoid aquiferous system. Sponges belonging to these genera are composed of anastomosed choanocytary tubes and present a well-developed atrium without choanoderm. This organisation of the aquiferous system has already been considered as asconoid, syconoid, and leuconoid by different sponge taxonomists, showing that there is no consensus on this subject. However, we disagree with all these classifications. In the asconoid aquiferous system, all cavities of the sponge are lined by choanoderm, and in the sylleibid, syconoid and leuconoid aquiferous systems, choanocytes line chambers, and not anastomosed tubes. After analysing histological sections of *Leucascus* and *Leucaltis* species, we concluded that a new aquiferous system is present in those genera. The solenoid aquiferous system (from the greek solenis = tube) is defined by the presence of anastomosed choanocytary tubes and atrium lined by pinacoderm. As the others, this aquiferous system has possibly no phylogenetic significance, as *Leucascus* and *Leucaltis* are most likely distant correlated genera. Nevertheless, the description of the solenoid raises an important discussion on the causes of the variety of aquiferous systems observed in Calcarea.

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Advances towards the mechanisms of coral death in competitive interactions with *Cliona* excavating sponges

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Encrusting and excavating *Cliona* sponges spread laterally killing and displacing coral tissue through extension of excavating tissue fronts and filaments under coral polyps. Excavation is carried out by etching and dissolving the carbonate through acidic and enzymatic reactions. However, the sole erosion of the polypar skeletal support does not seem to be enough to produce polyp detachment and death. Although some excavating sponges produce allelopathic substances that kill coral external tissue upon experimentally forced contact, it is not clear if these are being used to kill polyps during excavation. To hypothesize if the acidic enzymatic reaction or if release of allelopathic substances may be responsible for polyp death when excavating tissue break into coral polyps, we used dissociated cell culture experiments as a model for close-range sponge-coral interactions, as follows: (1) pH changes and cell densities were monitored in combined cultures of coral and sponge cells; (2) coral cells were exposed to crude extracts, fractions and compounds from *Cliona* species. Additionally, to approximate antifouling chemical properties in *Cliona*, whole coral larvae were also exposed to the above extracts and compounds. Stained *Cliona delitrix* sponge- and *Siderastrea siderea* coral-enriched cell fractions were cultured separately and together for 12-24 h in 24-well tissue culture trays. Chemical extracts, fractions and compounds from three excavating sponges: *C. delitrix*, *C. tenuis* and *C. celata*, and the synthesized sponge compound epiaxinisamine, were added to dissociated *Siderastrea siderea* coral cells and *Favia fragum* whole coral larvae. When coral and sponge cells where cultured together, there was a significant decrease after 24 h in pH and in the number of *Siderastrea siderea* cells, as compared to controls. The crude extract, and the butanol and non-polar fractions of *C. delitrix*, and the pure compounds N-acetilhomoagmatine from *C. celata* and epiaxinysamine, produced greater cell death than controls. In contrast, Clionapyrrolidine A from *C. tenuis*, which was involved in coral tissue death upon external contact, did not elicit greater coral cell death as compared to controls. Epiaxinysamine, and clionapyrrolidine A paralyzed and killed coral larvae. The aqueous and butanol fractions of *C. delitrix* induced amorphous coral larvae metamorphosis. Although histocompatibility reactions between sponge and coral tissue at the zone of contact below polyps cannot be ruled out, we hypothesize that coral polyps can also be affected by local acidification and enzymatic reactions or by release in some cases of allelochemicals during the sponge excavating process. However, the lack of effect of clioapyrrolidine A against coral cells needs another explanation. Those chemical compounds toxic to coral larvae may be responsible for absence of coral recruitment on live sponges.

Preliminary observations of oogenesis in the Caribbean excavating sponge *Cliona delitrix*

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Excavating sponges of the genus *Cliona* are among the most destructive organisms of the calcium carbonate matrix in coral reefs. In the last three decades, abundance of these sponges has increased in Caribbean Sea reefs, particularly in areas affected by pollution, diseases, bleaching and hurricanes. Thus, to understand this recent expansion and its ecological and environmental implications, it is important to evaluate which specific conditions are causing excavating sponges to propagate. Therefore, a fundamental step in this process is to determine species reproductive biology, which remains poorly understood to date. To characterize the cycle of sexual reproduction in the abundant excavating sponge *Cliona delitrix*, we have collected five to ten tissue samples from different individuals one or twice a month from October 2009 to March 2010, as part of a study that is still in progress. All individuals belonged to a local population established on inner reefs, adjacent to Ft Lauderdale Florida (USA). Samples were fixed for histology in Bouin's fixative solution, decalcified, desilicified, dehydrated and embedded in paraffin. Paraffin blocks were cut into 4-μm-thick sections at three levels inside the tissue, then stained with Heidenhain's solution. To estimate densities of gametes over time, two pictures (100x) of each section per individual were taken. Tissue area per picture was estimated using the Matlab program, then standardized to 7mm^2. Oocytes were present in 80% of the individuals in October, but only in 20 to 40% of individuals from November to March. The average size (max. diameter) of oocytes in October and beginning of November (47±1.9, n=44) doubled that of the oocytes found during the other months (23.22±1.19, n=38). We estimate that oocyte maturation and release occur around October, but cannot yet determine whether release is a synchronous process limited only to October or it might have already started as an asynchronous process in previous months. So far, we failed to find spermatozoa and cannot decide whether the individuals are gonochoric or hermaphrodite. These preliminary results appear to indicate that oogenesis in this sponge is a long process that starts with early stage oocytes in the coldest months of the year, and that the process was not disrupted even during the particularly cold winter of 2010 (19.2-23°C).

Spicule formation during long-term cultivation of primmorphs from freshwater Baikal sponges *Lubomirskia baicalensis*

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Sponges (Porifera) are the most ancient Metazoa existing at present that use silicon for formation of the skeleton. Biomineralization processes in sponges are one of the most actual problems of their biology. The cell culture of primmorphs is the most suitable model for studying spiculogenesis. The main aims of this work was obtaining of long-term cultivated primmorphs culture from freshwater sponges *Lubomirskia baicalensis* (Pallas, 1776) in both natural (NBW) and artificial (ABW) Baikal water and studies of the silicate effect of various concentrations on formation and growth of spicules in primmorphs. The culture of Baikal sponge *L. baicalensis* primmorphs was obtained by means of mechanical dissociation of sponge cells. The first fine aggregates were formed in vitro 10 ~15 min after dissociation. Aggregates of ca. 200 ~ 350 μm of green colour were formed in 2 ~3 h, whereas young primmorphs ~ 1-2 days later. Primmorphs were cultivated in vitro for a long time in ABW the composition of which was similar to the Lake Baikal water adding silicate, as well as in NBW at 3-6 °C. Formation, growth and morphology of siliceous spicules in primmorphs were observed during long-term cultivation in different mediums by means of light and electron microscopy (SEM and TEM). The largest quantity of young spicules was formed in ABW with 70 M silicate, whereas in ABW with 120 M silicate the primmorphs died. In NBW spicules were formed without induction. Young spicules started their formation inside sclerocytes. Being of certain length (ca. on the third day), they were pushed from a cell into the extracellular space. On the10-11 day they continued to increase, reaching about 150 μm in length. At the beginning of their growth spicules looked smooth. In the middle of spicules, there was an expanded bubble. The silicon content and elemental composition of cultivated spicules were determined with a microanalyzer JXA-8200 (JEOL Ltd, Japan). Organic axial filament surrounded by concentric layers of silicon was formed inside the channel. High concentrations of O (41%) and C (36%) and insignificant concentration of Si (22%) were recorded in the spicules. The silicon concentration gradually increased during long-term cultivation making up 44-50 %. Long-term cultivation of Baikal sponge primmorphs is a good model for in vitro studies of spiculogenesis in siliceous sponges and will allow the identification of proteins involved in biomineralization and decoding of their genes.

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The spatial distribution of sponge spicule in sediments in the Taiwan Strait and the South China Sea

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Biogenic silica fragment in marine sediments is an important indicator to understand the distributions of living siliceous organisms and paleoproductivities. Diatoms, silicoflagellate, radiolarians and sponges are the organisms which have biogenic silica skeletons. Among these biogenic silica fragments, diatom frustules demineralise immediately after death, and hence recycle in the eutrophic zone. In contrast, large amount of sponge spicules are with low dissolution rate in sediments and likely to reduce the amount of dissolved silica through the well known biological pump. Here we investigated the distribution of siliceous fragments in surface sediments of Taiwan Strait and South China Sea (SCS) for understanding the possible environmental factors (e.g. water depth and grain sizes) which govern the distributions of the siliceous fragments. In this study, 23 surface sediments in depths of 37~1128 m from the SCS were examined. In the sediments, only biogenic silica fragments of sponge spicules were found, however, were relatively low in contents if compared to the other studies around SCS. The highest abundance of sponge spicules in this study was observed at site which is adjacent to coral reefs, suggesting that these relatively high amounts of sponge spicules may come from its adjacent coral reef biota. The amount of sponge spicule showed positive and negative correlations with the water depths and sediment grain sizes, respectively. The low abundances of spicule in the shallow areas could be a result of dilution by terrestrial sediment through riverine input close to our sampling sites. Our results showed that, in SCS, distribution of biogenic silica in the shallow surfaces sediments, especially in the regions which are influenced by terrestrial input, can not reflect the local productivity of siliceous organisms.

Glass sponge reefs are a silica sink on the eastern continental shelf of the Pacific Ocean

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The important role of diatoms in the cycling of silicon (Si) has fundamental links to global CO_2 sequestering in the world's oceans as they fix an estimated ~35% of the planet's atmospheric carbon. The entire biogenic component of the global silicon balance has been solely attributed to diatoms and ignored other siliceous animals, such as sponges. However, glass sponge reefs cover >700 km^2 on the continental shelf of western Canada and represent massive reservoirs of silica. Furthermore, the heavily silicified skeletons of glass sponges and their long-lived nature may potentially create a Si sink.

To address the role glass sponge reefs have on silica cycling, we established a Si balance for a reef. We estimated the Si locked into 3 reefs in the Strait of Georgia using Ekman grabs and high resolution maps of the sponge populations. We established vertical profiles of dissolved Si above the reefs and in adjacent areas without sponges. We determined the dissolution of spicules of the main forming reef sponge, Aphrocallistes vastus, in sea water sampled from the reefs using bottled experiments (1-8 months) and compared them to dissolution experiments with diatom frustules. A. vastus is 79% silica, with 63% found in the fused skeleton. Over the areas of Howe (19,325 m2), Fraser (20,720 m2) and Galiano reefs (53,231 m2), silica is locked into sponge biomass at 13.5 kg m-3, 15.5 kg m-3, 21.1 Si m-3; these represent total silica reservoirs of 141, 179, and 595 tonnes respectively. Vertical Si profiles indicated lower concentrations in surface waters (48 μM) compared to benthic waters over the sponges (52 μM). Lower concentrations were found in benthic waters over bare substrate (48 μM) compared to areas of sponge reef (52 μM). Our experiments showed no detectable dissolution of sponge spicules after 8 months, whereas diatom frustules dissolved substantially after only 1 month. SEM observations confirmed the absence of dissolution by spicules, compared to frustules. However, previously dead skeletons which had spicules covered in Fe and Mn oxides and already exposed to dissolution prior to sampling, showed a maximum dissolution of 10 μmol l-1 after our 8 month experiment and study by SEM revealed evidence of Si pitting on broken cross sections of spicules. Our calculations indicate that at the Fraser reef 2.98 x 106 mol Si is locked into sponge biomass. A conservative growth rate (1 cm year-1) indicates Si removal from sponge growth (3.5 x 104 > mol Si year-1) would exceed Si release from dissolution (1.3 x 103mol Si year-1), a 27-fold difference, suggesting a considerable Si sink at the Fraser reef. If half of the 700 km2 area of all known sponge reefs consists of live sponges, we estimate a considerable reservoir of 9.7 x 108 mol Si is locked in glass sponge biomass on the benthos creating a massive sink that removes 9.0 x 106 mol Si year-1and releases 7.9 x 105 mol Si year-1. Glass sponge reefs may significantly impact local silica cycling in regions of the eastern Pacific Ocean.

Bioactive Microorganisms Associated with Sponges and Mollusks

Concepcion GP

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Marine invertebrate organisms such as sponges and mollusks represent unique biological habitats that allow certain microorganisms to thrive in their tissues as symbionts or epibionts. Microorganisms possess high genetic plasticity that may confer an adaptive advantage to themselves and their host. Their natural functions in the host tissue, which may include defense and protection against predators and competitors, feeding and nutrition enhancement, and regulation of growth, development and settlement, serve as ecological leads for the discovery of bioactive molecules. Here we present an overview of our studies on sponge- and mollusk-associated microorganisms that produce bioactive compounds.

First record of a former SW Atlantic endemic, *Clathrina aurea* (Porifera, Calcarea), in the Southern Peruvian Coast revealed by morphological and molecular data: A review of the species complex *Clathrina* '*clathrus*'

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Species of the *Clathrina* '*clathrus*' complex has a yellow cormus formed by anastomosed tubes and non-organised skeleton comprising regular triactines. The actines are often cylindrical, undulated at their distal part and have a rounded, blunt or sharp tip. The species of this complex (type localities between brackets) are: *C. clathrus* (Lesina, Adriatic Sea), *C. aurea* (Arraial do Cabo, Brasil), *C. chrysea* (New Caledonia), *C. luteoculcitella* (Great Barrier Reef, Australia) and *Clathrina* sp. 1 (Abrolhos, Brasil). Recently, specimens morphologically similar to this species complex were collected in Ica (Southern Peru). The aim of this study is to verify the taxonomic status of these Peruvian specimens through morphological and molecular analyses. The morphological analysis was based on the observation of external morphologic characters (colour, anastomoses of the cormus and presence of collecting tubes) and skeletal architecture (composition and organization). To search for significant differences in the dimensions of the spicules, non-parametric tests were perfomed. The molecular analysis was based on the comparison of sequences of the DNAr ITSI-5.8S-ITS2. A phylogenetic tree was built using Neighbour-Joining and considering *Leucetta* species as outgroup. Similar external morphologic features were observed in *Clathrina* sp. (Peru) and *C. aurea*. Compared to *C. aurea*, the triactines of Clathrina sp. (Peru) were found to be slightly different, with actines less cylindrical and bearing blunt tips. The Kruskal Wallis test revealed a significant difference ($p<0.0001$) in the spicule size among the species of the complex. However, a posteriori Dunns test revealed no significant difference ($p>0.05$) between the spicule size of *Clathrina* sp (Peru) and *C. aurea*. The molecular analysis revealed low levels of genetic divergences (0-0.4%) when comparing *Clathrina* sp (Peru) and *Clathrina aurea*. In contrast, high levels of genetic divergence (5%) were found among *Clathrina* sp. (Peru) and the other species of the complex. These results suggest that the accepted distribution of *C. aurea* should be extended to Peru and the differences in the spicule shape should be attributed to intraspecific variation.

Phylogeny of *Mycalina* Hajdu, Van Soest & Hooper, 1994 (Poecilosclerida, Demospongiae): reassessment of the affinities of *Isodictya* Bowerbank, 1864

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Poecilosclerida is the largest and more diverse Order of Demospongiae, with four suborders recognized in current classification. Its Suborder *Mycalina* includes nine Families and 30 Genera, defined as Poecilosclerida with palmate chelae, smooth megascleres of a unique type and sigmas. The skeletal structure is mostly plumose or plumoreticulate; axial compression, as well as isodictyal-anisotropic reticulations may also be present. The internal relationships in *Mycalina* are unknown, and its taxonomic scope is not universally accepted. One point of divergence is the allocation of Isodictyidae, once postulated to belong in the *Haplosclerida*. The main underlying synapomorphy for *Poecilsoclerida* is usually taken to be the chelae, albeit the previous proposition that mycalostyles and the transformation series of sigmancistras and its derivatives should be seen as the sole underlying synapomorphies. The latter argument is focused in the observation that *Isodictya* Bowerbank, 1864 and Coelocarteria Burton, 1934 possess chelae, but share with *Haplosclerida* important aspects of their anatomy, both at the architectural as well as at the spicules' level. A strengthening point raised was the observation that there is not much opposition about the shared occurrence of sigmas and toxas by both orders. In this study, as part of an assay at reaching a hypothetical phylogenetic framework for family groups in the *Mycalina*, the best placement for *Isodictyidae* has been reevaluated, initially on the basis of morphologic characters. Fourty seven specimens originating from the Porifera Collection of Museu Nacional / Universidade Federal do Rio de Janeiro (MN/UFRJ) were examined. Samples comprised all currently accepted families of *Mycalina*, plus *Phellodermidae* (*Myxillina*), and some families of the *Haplosclerida*: *Phloeodictyidae*, *Niphatidae* and *Chalinidae*. Family *Geodiidae* (*Astrophorida*) was chosen to root the tree. Other taxa, such as *Halichondriidae*, end up in the ingroup due to parallelism in architectural traits, or have rather distinct 'bauplans', as is the case of all four keratose orders. A data matrix was constructed with a total of 14 characters (13 unordered and one ordered by dollo-up parsimony). The analysis was performed using PAUP* version 4.0b10 with the Branch and Bound exact algorithm, optimized through ACCTRANS and reweighted a posteriori based on characters' RC. The analysis resulted in two most parsimonious trees (CI - 0.89, RC - 0.85), of which the more fully resolved one is ((*Geodiidae*) (*Niphatidae* (*Chalinidae* (*Phloeodictyidae*, *Isodictyidae*))) (*Desmacellidae* (*Mycalidae* (*Esperiopsidae*, *Phellodermidae*) (*Cladorhizidae* (*Guitarridae* (*Hamacanthidae* (*Merliidae*, *Podospongiidae*))))))). These results point to haplosclerid affinity of *Isodictyidae*, left out of the *Poecilosclerida*, who is recognized on the basis of mycalostyles. *Isodictyidae* grouped with the haplosclerid families considered here, due to the shared occurrence of cigar-shaped oxeas. Curiously, *Phellodermidae* grouped with *Esperiopsidae*, rather than with *Cladorhizidae*, where *Abyssocladia* is currently classified. This genus has only recently been transferred from *Phellodermidae* to *Cladorhizidae*.

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16S rRNA gene-based analysis of bacterial diversity and composition in the freshwater sponge *Ephydatia fluviatilis*

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Research in the past few years has largely contributed to a better understanding of the diversity and complexity of microbial communities found in association with sponges (Porifera). However, our knowledge of the sponge-associated microbiome remains virtually restricted to marine ecosystems. Here, we report on the diversity and composition of bacterial communities in the freshwater sponge *Ephydatia fluviatilis* inhabiting the Vinkeveense Plassen lake, Utrecht, The Netherlands. PCR-DGGE fingerprints generated with "universal" (i.e., total bacterial communities) and group-specific (i.e. *Actinobacteria* and *Pseudomonas*) primer systems from total community environmental DNA (TC-DNA) revealed lower community diversity and enrichment of specific bands in *E. fluviatilis* TC-DNA in comparison with lake water samples. Multivariate analysis of PCR-DGGE profiles resulted in a separate clustering of *E. fluviatilis* samples collected in two consecutive years (2007 and 2008), whereas year-to-year variation in freshwater PCR-DGGE profiles was less pronounced. Bacterial 16S rRNA gene libraries were constructed from sponge and lake water TC-DNA samples from year 2008. These libraries were found to differ as to diversity and composition. *E. fluviatilis*-derived sequences (n = 154) were affiliated with six recognized phyla, i.e. *Proteobacteria* (32.5%), *Planctomycetes* (13%), *Actinobacteria* (10.4%), *Bacteroidetes* (9.7%), *Chlamydiae* (1.3%) and *Verrucomicrobia* (1.3%); one candidate division, i.e. TM7 (24%); and 12 highly similar (<0.5% sequence divergence) sequences (7.8%) that were not classifiable at phylum level. *Actinobacteria* (40.9%), *Proteobacteria* (32.9%), and *Bacteroidetes* (17.1%) were the dominant bacterial phyla in the freshwater clone library (n = 164), whereas sequences affiliated with *Planctomycetes* (2.4%), *Verrucomicrobia* (1.2%), *Acidobacteria* (1.2%) and the candidate phylum OP10 (3%) were registered wit lower frequency. Higher diversity of actinobacterial 16S rRNA gene sequences was detected in freshwater in comparison with sponge TC-DNA, complementing observations made for Actinobacteria-specific PCR-DGGE profiles. The majority (~90%) of the freshwater sequences resembled, with 98 to 100% nucleotide sequence similarity, uncultured bacterial clones previously registered in aquatic environments. Interestingly, this proportion dropped to about 63% in the *E. fluviatilis* clone library. Although the bulk of bacteria found in *E. fluviatilis* was not representative of highly specific sponge-bacterial associations, phylogenetic inference revealed a phylotype in the *Alphaproteobacteria* clade that is unique to *E. fluviatilis* and shares only 92% resemblance with its closest relatives. Altogether, our results indicate that the freshwater sponge *E. fluviatilis* holds a microbiota that differs from that found in its natural environment, what is suggestive of a selective process that is probably intrinsic to the organism´s metabolism and life style.

Diversity of sponges in bathyal coral reefs of a subsidiary canyon of Aviles Canyon complex (Cantabrian Sea)

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Submarine canyons are topographically complex seascapes, contain diverse bottom types and can be highly unstable environments due to active mass-wasting and high sediment loads. They act as conduits for the passage of material down the continental slope, and profoundly modify the hydrodynamic regime of continental margins. Benthic infaunal biomass and density in canyons can be higher, lower or similar to the adjacent slope. Whether macrofauna is more abundant and diverse inside canyons depends to some degree on the physical disturbance regime, bottom types and the rate and quantity of organic matter deposition. Although data on the abundance and biomass of the megabenthos in canyons are available for several sites worldwide, the diversity of the benthic assemblages is rarely documented and not known for the sponge fauna of canyons. Thus, our objective in this study within the Indemares project is to quantify levels of species richness of sponge assemblages in the Aviles Canyon. A Collection of sponges was collected during Indemares-Aviles 0410 expedition of the Spanish Institute of Oceanography in an unstudied subsidiary canyon of Aviles Canyon complex (Cantabrian Sea) named Las Yerbas Canyon of 16518 m length and 6152 wide. The first step to study this area was the cartography and identification of the main geomorphologic features of the study area. Information regarding the morphology of the seafloor of the Aviles Canyon was obtained with a Simrad EM-300 (30 kHz) multi-beam system on board R/V Vizconde de Eza of the SGM and the samples were collected by a rock dredge between 588 and 790 m deep. Despite presence of Scleractinian corals is well known in the Biscay Bay and particularly in the Cantabrian Sea, we demonstrate in this expedition, for the first time, the existence of a cold-water coral reef in the Spanish waters by means of a non-intrusive method based on a towed vehicle that collects continuous high definition video and periodic digital still imagery of the seabed along transects.

Ecology of the sponge fauna of El Cachucho (Le Danois Bank, Cantabrian Sea) the first off shore Marine Protected Area in Spain

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Sponge grounds (e.g., sponge dominated communities), are examples of areas (mega-habitats) which are topographical, hydrophysical or geological features (including fragile geologic structures) known to support vulnerable species, communities, or habitats. Vulnerability includes considerations of both the likelihood that a population, community, or habitat will experience substantial alteration from short-term or chronic disturbance, and the length of time required to recover to its prior state, after a disturbance. In this way, the main objective of the ECOMARG project was the integrated study of the benthic-demersal ecosystem of the singular Asturian marginal shelf (Le Danois Bank), a barely well-known area in spite of belonging to the Spanish EEZ and being subject to fishing activities. The study had a multidisciplinary strategy that includes the abiotic scenario and the benthic and demersal communities. Several samplers were used on each biological station in order to study the different compartments of the benthic fauna. Epibenthos and demersal species were studied using three different gears, a Porcupine baca trawl, a rock dredge and a beam trawl. The Ecomarg benthic project has been conducting an intensive study of the benthic fauna of the Le Danois Bank (El Cachucho, in the Spanish fishermen terminology). In this work more than 500 specimens of sponges were identified and assigned to classes *Hexactinellida* and *Demospongiae* (any representative of class Calcarea was collected). Differences in density, species richness, and diversity among habitats were analysed, as well as the relationships between these ecological parameters and depth range, slope. The effect of environmental variables on sponge distribution was studied using canonical ordination.

Redescription and ecological characterisation of *Artemisina transiens* Topsent 1890 collected by ROV techniques 120 years after original description

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In July 2008, Oceana Europe organized a cruise in Galicia and Cantabrian waters with the aim of to investigate about ecological significant areas to the amplification on marine protected spaces network. This expedition on board the ' Oceana Ranger ', a Ketch catamaran of 23.5 metres long, carried out 51 dives with a ROV (Remote Operated Vehicle) counted more than 88 effective filmed hours. The ROV employed was a Phamtom HD 2+2 of Deep Ocean Engineering providing a digital camera of 750 resolution lines. In ten different places in Galicia and Asturias was registered a sponge with a characteristic habitus named in the expedition "mushroom sponge". This species was found at different depths (sometimes in high densities) but preferably more than 50 m. To identify and study it, a special dive with ROV and divers were prepared to collect samples and analyze the specimens. As result of these dives, 10 specimens were collected in the Somos Llungo area in the west of Peñas Cape (Asturias) at 48-50 m deep. The specimens belong to *Artemisina transiens* Topsent 1890 species, a globular pedunculate sponge with apical oscules and even surface. The species is known only from the North West coast of Spain, and its status was questionable. In fact, there are only two records of this species after original descriptions. The genus *Artemisina* was erected by Vosmaer (1885) for the type species *Artemisina suberitoides*. The taxon has no real distinctive features, although it differs from other *Microcionidae* in lacking a distinctive choanosomal skeleton or definite sponging fibres, lacking echinating spicules and having a nearly radial ectosonal skeleton. For these reasons in this work we redescribe the species using modern techniques like Scanning Electron Microscope for the skeleton study and also provide many ecological data to characterize the habitat. The morphology of the seafloor of the Somos Llungo area was obtained with a Simrad EM-300 (30 kHz) multi-beam system on board R/V Vizconde de Eza of the SGM.

Chemical profiles of *Dysidea fragilis* from Portugal

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Sponges of the genus *Dysidea* (family *Dysideidae*, order Dictyoceratida) are widespread in marine environments and are a rich source of metabolites with promising biological activities to human health. The taxonomic identification based on morphological characters of species of the genus *Dysidea* is difficult due the plasticity of their morphological characteristics. Thus, the use of secondary metabolites as chemical markers may provide valuable clues for taxonomic classification (1). Previous studies on *Dysidea fragilis* (Montagu, 1818) from different geographic areas showed that this species has marked geographic variation on the metabolic patterns, which include sesquiterpenes, sesterpenes, polyhydroxylated sterols, polybrominated diphenyl ethers, diketopiperazines and azacyclo propene lipid derivatives (2). In this work we analyzed the chemical profile (by TLC, NMR, GC, and HPLC) of several organic extracts of different specimens of *Dysidea fragilis* collected in two different sites along the Portuguese Coast, Setúbal and Ria Formosa, in order to verify if there are metabolites that can be used as chemical makers of this species from these geographic areas. The results showed that the chemical profiles of *Dysidea fragilis* samples were similar: all samples are characterized by the presence of the same mixture of furanosesquiterpenes but in different quantities. Nakafurane-9, a know anti-feedant furanosesquiterpene first isolated from a pacific *Dysidea fragilis* specimen (3), was the main metabolite in the samples collected in Ria Formosa, whereas an unidentified furanosesquiterpene was the main metabolite of *Dysidea fragilis* samples from Setúbal. We are currently in the process of isolation of this high instable compound.

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Acanthostylotella, the forgotten genus. Affinities of the family Agelasidae (Porifera, Demospongiae, Agelasida): Chemical, molecular and morphological evidence

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Recently a few specimens were collected in shallow waters of Indonesia and Palau with an odd combination of characters, namely a regular reticulate skeleton with peculiar apically spined styles. Although, the specimens were first identified with aid of Systema Porifera as *Amphinomia* Hooper, 1991 (Raspailiidae, Poecilosclerida), closer examination of the literature revealed the genus, *Acanthostylotella* Burton & Rao, 1932 (Halichondriidae, Halichondrida). This mono-specific, poorly known genus, with the type locality in Indonesia (*Stylotella cornuta* (Topsent, 1897)), had evidently been overlooked in the recent classification of sponges. In this paper we re-evaluate this taxon and its position within the Demospongiae, based on morphological, chemical and molecular evidence. Morphological analyses revealed that all examined specimens of *Amphinomia* and *Acanthostylotella* share the regular reticulate skeleton cored with styles which may be smooth, apically and/or basally spined. In addition, all specimens possess coring acanthostyles and/or verticillated styles, although the number of this spicule type varies within species. The genus *Amphinomia* clearly revealed the same set of morphologically similar characteristics and this taxon is thought to be a junior synonym of *Acanthostylotella*; we therefore synonymise *Amphinomia* with *Acanthostylotella*. Sponges of the genus *Agelas* are chemically unique due to their content of brominated pyrrole alkaloids as well as numerous other metabolites that have been isolated and identified from *Agelas* spp. Chemical evidence showed that brominated pyrrole alkaloids were isolated from various *Acanthostylotella* spp and *Amphinomia sulphurea*, suggesting the affinities to the Agelasida. In addition, molecular analyses revealed that *Acanthostylotella* clearly clusters together with *Amphinomia* inside the Agelasida clade, and more distantly from Raspailiidae and Halichondrida. Thus, based on morphological, chemical and molecular evidence, we reassign the genus *Acanthostylotella* within the family Agelasidae (Agelasida). We therefore propose to amend the definition of both the order Agelasida and family Agelasidae.

Mitochondrial and nuclear gene sequences show conflicting patterns of divergence among putative cryptic species in the marine sponge *Callyspongia vaginalis*

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Cryptic species are likely prevalent in the Porifera due to their simple, plastic morphological characters. DNA sequence data from the mitochondrial cytochrome oxidase I (COI) gene of the coral reef sponge *Callyspongia vaginalis* sampled from seven Floridian locations, revealed three divergent mitochondrial haplotypes. These haplotypes are separated by an average of 12 mutational steps and have an average genetic (uncorrected p) distance of 2.2%. The ability to draw phylogeographic inferences from COI may be limited due to the slow nucleotide substitution rates of poriferan mitochondrial genes. We collected additional sequence data from the nuclear gene elongation factor-1α to test whether the three haplotype lineages from Florida are cryptic species and for phylogeographic concordance with the previous results from COI. In contrast to the results from the COI data, data from the nuclear gene revealed extensive allele sharing among the three mitochondrial clades. Results from a parametric bootstrap analysis allowed us to reject the hypothesis that the same neutral demographic process could produce the conflicting patterns observed in the mitochondrial and nuclear DNA datasets. These discordant patterns may be the result of non-neutral forces preventing introgression of genetically divergent mitochondrial haplotypes among individuals of the same species. Sequence data from additional nuclear markers, as well as broader geographic sampling, will allow us to further investigate the generality of the Floridian pattern at a Caribbean-wide scale.

Biodiversity of Porifera in the greater Caribbean—Status and emerging patterns

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The general perception of the greater Caribbean sponge diversity is that it is uniformly distributed, but there is no documentation of regional distribution patterns. This issue was assessed using previous reports of species in the region, geographic affiliations, and taxonomic analysis. A master list of species was gathered from recent faunistic surveys, taxonomic revisions, and grey literature. Validity of species and their classification was checked against the World Porifera Database. The information was gathered in a species-by-country matrix, with values of presence/absence. This matrix was analyzed with the biodiversity index Average Taxonomic Distinctness (AvTD) to evaluate regional and local diversity (for each country), and with cluster analysis (based on Sørensen's coefficient and taxonomic dissimilarities) to identify similar faunas in the region. The AvTD is a measure of the average degree to which individuals in an assemblage (or list, in this case) are related to each other, based on a taxonomic hierarchy; hence, a list of closely related species must be regarded as less 'biodiverse' than a list of the same number of more distantly related species. This index was used to test the null hypothesis that the AvTD for a particular country is within the expected range, considering the richness and the pool of species in the region (Taxonomic distinctness Test or TAXDTEST). As a result, a total of 760 species belonging to the three sponge classes, Demospongiae, Calcarea, and Hexactinellida, are listed. The best-represented group in the region is the Demospongiae, with 700 nominal species names. The Calcarea (33 species) is reported from most Caribbean sites with very low diversities (1-5 species per site). Hexactinellida (27 species) can be considered "unknown" since most species were reported with poor documentation and from only one or two locations. In any case, the number of sponge species reported for each country is highly variable (from 58 to 256), probably because of different sampling efforts. Nonetheless, and considering only Demospongiae, at least 11 countries appear to be "evenly examined", since more than 100 species have been reported from each. Still, all of them have values of AvTD significantly below the expected (TAXDTEST, 1000 simulations, $p<0.05$). On the other hand, the cluster analysis reveals five groups: 1, Venezuela, Panamá, Belize and Colombia; 2, Cuba, Bahamas and Jamaica; 3, Bonaire and Curaçao; 4, Florida and Gulf of México; 5, Dominican Republic and Cayman Islands, all having levels of similarities in species shared ranging 40-70%. The remaining six countries do not fit any group, probably because they are really different (deep-water material from Barbados), distant (Bermuda), or poorly studied (Puerto Rico, Virgin Islands, and Costa Rica). These results indicate that the high sponge diversity in the region is due to a sum of endemic faunas sub-regionally distributed. It is evident that the exploration of less known habitats and unsurveyed countries is still necessary for a proper understanding of sponge biodiversity in the Caribbean. Concerted international efforts to train and promote experts in the classes Calcarea and Hexactinellida are also crucial to this effort.

Bocas Del Toro, Panama: A Caribbean hotspot for Poriferan fauna

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The last published survey of sponge biodiversity from Bocas del Toro, Panama (Diaz, 2005) revealed a total of 123 species from mangrove and reef habitats after exploring by snorkeling a very small area in the region (12 sites within a radius 10 Km explored in nine days). This survey added approximately sixty species to a previously known 63 reported from an extensive evaluation of coral reef fauna (Guzman 2003). The large representation of Porifera biodiversity found in Bocas del Toro (123 from a total of 144 species reported from Panama) suggests that the region is a possible hotspot for marine biodiversity. In the past four years three specialized courses (Taxonomy and Ecology of Caribbean Sponges; 2005, 2006, 2007) and an expedition as part of the Porifera Tree of Life Project (August, 2009) allowed us to further explore the shallow (< 20 m depth) waters of the region, adding a survey of sub-rubble communities at one site (STRI Point), and five previously unexplored sites (Buoy 19, Sponge Gardens, Outer Hospital Point, Wild Cane Cay, and Long Bay Point). These surveys have steadily added approximately 100 different species to the known Panamanian sponge fauna. A recent revision of *Tetractinellida* from the region added five species, with two newly described ones (Cardenas et al. 2009). Here, we present 50 species previously unknown for Panama and describe five species belonging to the genera *Topsentia*, *Clathria*, *Aplysina*, and *Xestospongia*. As a result of further investigating the same shallow areas previously studied, and adding a few new sites within a 20 km range, we have increased the known sponge biodiversity of the region to more than 200 species. A similarity analysis of species composition among the three habitats surveyed reveals distinct affinities of mangrove, open reef and sub-rubble communities. It is expected that further exploration of sub-rubble habitats, deeper reef areas, and other sites of the region (i.e. the Principe Valiente region) will discover even more rare and undescribed species. This large concentration of sponge biodiversity within such a small geographic area supports the status of Bocas del Toro as a hotspot Caribbean marine biodiversity, with important implications for conservation ecology in this region.

A checklist from shallow and mesophotic marine sponges from Little Cayman Island.

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As part of the NOAA Ocean Exploration Project: "Cayman Islands Twilight Zone", two expeditions to Little Cayman Islands were carried out during 2007 and 2008. A multidisciplinary team explored marine communities from shallow reefs to 300 ft depths (mesophotic) using SCUBA, Nitrox and mixed gases diving technologies. The survey covered shallow fringing and patch reefs near shore to sheer walls with overhangs and small caves. 144 samples were collected, representing 110 species of marine sponges belonging to the classes Demospongiae and Calcarea (2 species). This is the first comprehensive survey of marine Porifera from the Cayman Island System. The diversity encountered was represented by approximately 38 genera from 29 families, and 14 orders of Demospongiae, and included 2 genera from 2 families and 2 orders of Calcarea. In the shallow reefs, the Demospongiae orders Agelasida, Haplosclerida, Verongida and Poecilosclerida exhibited the greatest diversity, comprising a classical signature of reef families such as Callyspongiidae, Niphatidae, Petrosiidae (Haplosclerida), Apysinidae (Verongida), Mycalidae, Microcidonidae (Poecilosclerida), Clionaidae (Hadromerida) and Agelasiidae (Agelasida). Half of the species inhabiting Mesophotic depths were restricted to those depths. Ten species restricted to mesophotic depths are considered undescribed and are presented here. They belong to the genera: *Ircinia*, *Aplysina*, *Topsentia*, *Jaspis*, *Spirastrella*, *Prosuberites*, *Xestospongia*, *Forcepia*, and *Theonella*, and are currently being described.

Recent advancements on the knowledge of marine sponge biodiversity from Venezuela

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Recent advancement on our knowledge of marine sponge biodiversity in Venezuela is here presented. Studies carried out since 2003, mostly kept as grey literature (agency reports, undergraduate and graduate thesis, etc), have doubled the number of species reported from continental and islands territories in Venezuela (from 76 to 150 species). This improvement in sponge Biodiversity knowledge is the result of the development of educational workshops (2003, 2009), and few explorations carried out locally by researchers and/ or students from public Universities (Universidad Central de Venezuela, Universidad de Oriente, Universidad de Carabobo, and Universidad Simón Bolivar), private institutions (Fundación los Roques, Museo Marino de Margarita) or in two instances projects supported by conservation or research agencies (Instituto Venezolano de Investigaciones científicas de Venezuela, Conservation international). An updated list of the sponge species and their distribution within the country is here presented, and local and regional geographic affinities are discussed. Sponge surveys in Los Roques National Park, Cubagua Island, La Restinga National Park, Isla Larga, La Tortuga and the continental shores of La Guaira are responsible for most of the additions to the sponge fauna. We estimate that we have only recorded a third of the potential number of species from shallow marine ecosystems in the country, considering that the current sponge biodiversity in the Caribbean is between 600-700 species (Diaz et al. this book) with a local maximum number of species reported from Cuba of approximately 250 species. Critical aspects to increase the knowledge and to preserve the biological integrity of this group are discussed. Funding support to national researchers is essential to allow the evaluation of unexplored regions, environments (Deeper areas), and to support the description of new species to science, and continue education on Sponge taxonomy. A National effort and official governmental support are required to establish and maintain a reference collection of species from the country.

Folk classification of sea sponges (Animalia, Porifera) by inhabitants of a traditional fishing community at Camamu Bay, Bahia State, Brazil.

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The sponges have been known and used since antiquity, however, systematic studies on folk perception, classification, and use of these marine invertebrates are still very rare and inexistent in Brazil. This study was carried out in the fishing community known as Ilha do Contrato, which is located at Camamu Bay, Bahia State, Brazilian Northeastern. The data were obtained by means of usual techniques of ethnographical recordings. A total of sixty six individuals have participated of the study, both men and women whose ages ranged from eighteen to seventy five years old. Six key-informers were identified based on their consolidated ethnobiological knowledge regarding sea sponges. The results show that different types of sea sponges are culturally recognized as like-plants organisms. Although there is a rich diversity in species, few of them receive local names and ethnosemantic analyses were performed. Sea sponges, as well as other marine organisms from different taxa, are all included in the ethnocategory 'Lime', and then the ethnotaxonomical complex lime has been proposed. This ethnobiological rank is equal to the Life Form level according to Berlin's principles of folk biology classification, which, by its turn, is inserted in the Unique Beginner locally labeled as 'Plant. Since there are scarce scientific knowledge on sea sponges from the region of Camamu Bay (just *Craniella quirimure* Peixinho, Cosme & Hajdu, 2005 is formally known in this locality), folk wisdom on them might help to indicate some strategies regarding the inventory of local biodiversity and more sustainable fishing practices and management of local resources.

Molecular phylogeny of glass sponges (Porifera, Hexactinellida): increased taxon sampling and inclusion of a mitochondrial protein-coding gene

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Despite their ecological, evolutionary, and geological significance, hexactinellids (glass sponges) are still one of the most understudied groups of sponges. Molecular approaches to reconstructing their phylogeny have only recently been applied, and were exclusively based on ribosomal RNA genes (rDNA). Although the rDNA phylogeny appears reliable as it agrees well with morphology-based systems, it has not been tested so far with protein-coding gene sequences. Also, taxon sampling for certain groups is still poor, hampering insights into their evolutionary history. Here, we have included further species in molecular phylogenetic analyses of Hexactinellida, especially improving taxon sampling of the dictyonal, sceptrule-bearing sponges (Sceptrulophora), and also established an ~1.3 kb region of the mitochondrial cytochrome c oxidase subunit I (COI) gene as an additional phylogenetic marker. Single-gene trees from the nucleotide and amino acid COI alignments were largely congruent with the rDNA phylogeny, but suffered from poor bootstrap support for many nodes. However, inclusion of the COI partition improved resolution of the combined-analysis phylogeny. Our results confirm monophyly of clavule-bearing farreids (Farreidae) with a typical farreid dictyonal framework. In contrast, a close relationship of Sarostegia to this clade is rejected; instead, our results are consistent with an earlier classification of this genus in Euretidae. We also found further support for monophyly of Tretodictyidae and reciprocal paraphyly of Aphrocallistes and Heterochone (Aphrocallistidae). Among Lyssacinosida, our results suggest non-monophyly of Lanuginellinae (Rossellidae) and reject the previously proposed affinity of *Clathrochone clathroclada* (*incertae sedis*) to Leucopsacidae.

Feeding in the calcareous sponge *Leuconia johnstoni* Carter 1871: trophic role of bacteria.

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In order to study bacterial associations of calcareous sponges we investigated the contribution of bacteria to the sponge diet in the temperate sponge *Leuconia johnstoni* Carter 1871 (Calcaronea, Baeriida) collected off Concarneau (Brittany, France). The ultrastructure of the sponge aquiferous system was observed via scanning electron microscopy of cryofractured fragments revealing the leucon organization of canals and extracellular bacteria in the choanocyte chambers and the mesohyl. Filtration efficiency and bacterial uptake by the sponge were measured during aquarium experiments, and compared for a strain of *Vibrio splendidus*, which are abundant coastal planktonic bacteria in the North Atlantic, and a strain of *Bacillus* sp. isolated from the sponge. Results show a similar filtration activity ranging from 5 to 10 mL/cm^3/h, and suggest a selective feeding on small-sized *Vibrio* (1 µm) rather than *Bacillus* (2 µm rods) bacteria as shown by higher values of retention rates respectively 70-75% for *Vibrio* and 22% for *Bacillus* . There was no induction of antimicrobial activity in sponges exposed to these bacteria, and the structure of the aquiferous system was maintained functional which indicates that these microorganisms were not killed by production of toxic metabolites but were rather ingested by the sponge. Bacterial fatty acids were used as markers of trophic association, via characterization of their contribution to the sponge fatty acid profile. Seasonal fluctuations of the proportions of bacterial markers, the branched fatty acids 15:0 iso, 17:0 iso,15:0 anteiso and17:0 anteiso, were evaluated over four successive years and linked to the annual cycle of somatic growth. Results indicate that in spring and in summer the sponge growth is strongly based on bacterial biomass. Lower levels of these bacterial markers were detected throughout the year which suggests the persistence of an association between the sponge-host and a small bacterial compartment. Its diversity, structure and role for the sponge are being investigated as part of other ongoing research projects.

Mapping impacts of disease on sponge communities using Geographic Information Systems (GIS)

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The diversity of species on coral reefs, combined with competition for limited resources, inevitably leads to complex community structure due to interactions between organisms. As a result, understanding coral reefs at a community level is difficult, especially with regard to quantifying organisms in a spatial context without oversimplifying community inter-relationships. Since some of the most abundant members of coral reef communities are sponges, we propose a method for studying sponge communities and the potential transmission of disease on Caribbean reefs using Geographic Information Systems (GIS). *Aplysina cauliformis* is an abundant and important member of the sponge community in the Exuma Cays, Bahamas. This species is susceptible to *Aplysina* Red Band Syndrome (ARBS), which results in a lesion, followed by tissue necrosis. Our goal was to identify patterns of distribution of healthy and diseased *A. cauliformis*, in order to test hypotheses regarding the transmission of ARBS among *A. cauliformis* and the ability to predict patterns of progression of the disease through a sponge population. Using GIS techniques, we mapped sponge communities in their spatial context, with respect to each other and to other sessile organisms on the reef. We then used GIS tools to conduct spatial analyses that are more complex than could otherwise be undertaken. Reef communities were mapped using photographic imagery. Each photograph encompassed an area of 1 m^2 and the individual photographs were combined into a photo-mosaic representing the entire community. Using data collected in the field, attributes such as size and health status of individual sponges were assigned to each sponge featured in the dataset. Using basic mapping principles and GIS software tools, we measured spatial dependencies, regressions and interactions in this community for these characteristics. By mapping the distribution of individual *A. cauliformis* infected with ARBS, we found that diseased sponges occur in clumped distributions, supporting our experimental data that ARBS is a contagious disease, largely transmitted by contact with infected individuals. Diseased sponges were nearly twice as large as healthy sponges, although there was no significant correlation between individual sponge size and number of ARBS lesions. The continued use of GIS techniques will enable us to conduct simulations and model the effects of changing variables within communities, and to predict the spread of diseases on reefs, greatly increasing our understanding of community-level interactions on coral reefs.

Isolation and identification of chitin from *Vauxia gracilenta* fossil sponge

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The Vauxiidae are among the best known taxa in the middle Cambrian from the Burgess Shale, although their interpretation is controversial. These sponges exhibit an apparently reticulate, fibrous skeleton. Assuming an aspicular organic skeleton, Rigby (1986) suggested an affiliation with the modern 'Keratosa'. Although this view has been widely adopted, their nature and relationships have not been fully resolved. Recent work has shown the occurrence of chitin within fibrous skeletons of recent "keratose" Verongida. Given the reasonable stability of chitin under geological conditions, these discoveries stimulated our attempts to locate this structural biopolymer in fossil sponges. The object of the current study was to test the hypothesis that chitin was an essential skeletal component of early sponges assigned to Verongida. *Vauxia gracilenta* (Walcott, 1920) is common in the Middle Cambrian of the Burgess Shale (British Columbia, Canada). We have studied Burgess Shale *Vauxia* samples because of the exceptional preservation of the fossils. The fossilized material consists of brownish anastomozing fibers (diameter ~100 µm) in an arrangement typical of *Vauxia*. The results of the structural and specroscopical analyses performed using NEXAFS, FTIR and CFW staining, and electron diffraction agreed that the demineralized fibrous material isolated from fossilized *V. gracilenta* consists of alpha-chitin. Chitinase digestion experiments again confirmed the chitinuous nature of the isolated *V. gracilenta* fibers. Additionally, results obtained using HPLC, HPSEC, HPCE and ESI-MS clearly indicate that the sample contains a species, which is highly similar in its properties to D-glucosamine. Our results confirm that *Vauxia* is a "keratose" demosponge. The Vauxiidae are therefore likely to be the most basal definitive demosponge group known, despite the abundance of protomonaxonid "demosponges" in the Cambrian fossil record.

Unique form of silica within skeletal fibres of Verongida sponges

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Spicules, skeletal frameworks and desmas are the main forms of the silica deposition in sponges. There are over 12 basic types of megasclere and 25 types of microsclere reported in Demospongiae, 20 basic types of megasclere, and 24 types of microsclere in Hexactinellida, besides a long list of variations of the basic types. The formation and templating of silica has also been studied in spicules of hexactinellids and demosponges. However, despite the recognition of Si in the outer compact layers of fibres of verongid sponge *Aplysina aerophoba*, determined using X-ray microanalysis by X. Turon previously, the synthesis of siliceous structures in Verongida has not been described up today. In other keratose demosponges, incorporated sediment and other foreign particles are the main source of inorganic skeletal material in spongin fibres. However, Verononglda species possess a skeleton without spicules or foreign detritus. Moreover, one of the criteria that differentiate the orders Verongida and Dictyoceratida is the presence of foreign particles only in the latter. Any silica present in verongid fibres must be synthesized by the sponge. To elucidate the nature of the stable fibre components under acid dissolution, we demineralized fibres of investigated verongids by treatment with HCl. Fibre skeletons were washed three times in distilled water, cut into 2x2.5 cm^2 pieces, and placed in a 10 ml glass vessel containing 8 ml of 3 M HCl solution. The vessel was covered to restrict evaporation and placed under thermostatic conditions (37 °C) with gentle agitation for 7 days. Immersion in HCl did not lead to an immediate loss of brown pigments from the fibres. The fibres became white and fragile only after 5 days of incubation in HCl. The white material obtained after acidic treatment of the sponge samples was washed with distilled water five times and finally dialyzed against deionized water. The treatment of native sponge fibres with 3M HCL solutions resulted in dissolution of the acid-soluble calcium carbonate mineral component. This yielded a perforated fibre surface. Their structure is visible in light, fluorescence as well as scanning electron microscopy. In this study we will report about unique aspiculated silica-chitin-based layered structures within skeletal fibres of Verongida sponges. Silicon concentrations were determined after alkali dissolution of isolated layers by the silicomolybdate method to be about 100-150 µg of Si per mg of dried skeletal fibre. Possible implications of this discovery for systematic and phylogeny of Verongida sponges will be discussed.

Temperature effects in reproduction of the Mediterranean sponges

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Potential effects of global change act from the physiological level to the community and can lead to diversity loss and affect ecosystem functioning. Baselines on reproduction cycles of marine invertebrates are needed to better assess the possible effect of the environmental changes at the population level. Sponges represent one of the most ubiquitous groups of metazoans within benthic communities. Consequently, these organisms have a special set of adaptations, including life and reproduction strategies. With the programme PHENOMED "Climate change, phenology and reproduction: Mediterranean sponges as models", we revisited the successive reproductive phases of some Mediterranean sponges (Demospongiae, Homoscleromorpha and Hexactinellida) with distinct reproductive strategies and sensitivity to environmental changes. Samples of each species were collected monthly over several years in a site of the NW Mediterranean equipped with a permanent temperature recorder. In spite of different reproductive cycles, all investigated species exhibited responses to change in seawater temperature. It is too early to predict potential effects of the global warming on Mediterranean populations, but these first findings fully justify the monitoring of life cycles and reproductive effort of benthic organisms.

First detection of miniature transposable elements in Porifera

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Transposable elements play a vital role in genome evolution and may have been important for the formation of the early metazoan genome, but only little is known about transposons at this interface between unicellular opisthokonts and Metazoa. Here we report on the detection of the first miniature transposable elements (MITEs, Queen1 and Queen2) in sponges. Queen1 and Queen2 derive from two novel Tc1/mariner MITE families and are present in more than 6,000 resp. 7,000 copies in the *Amphimedon queenslandica* genome.

Assessing diversity and evolution of Lake Tanganyika's sponges using DNA barcoding

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The diversity, systematics and ecology of sessile invertebrates in ancient lakes, particularly sponges (Porifera), are poorly understood despite their likely pivotal ecological function as filter feeders. Since these animals require careful hand collection underwater, are often delicate to transport, and are perceived by some to be less charismatic than, for example, cichlid fish, there has been a dearth of work on sponges. This is compounded by challenges in fresh water sponge taxonomy for the non-expert due to the depauperate suite of diagnostic morphological characters. We have sought to redress this by a series of underwater surveys of sponge occurrence in the Kigoma region of Lake Tanganyika, coupled with collections for systematic study. We used a first pass morphological approach to categorize morphospecies and worked to link this with the established taxonomy. However, a means of molecular identification such as DNA barcodes will accelerate the process of unambiguous species identification and reveal cryptic species. Here, we present preliminary results on molecular barcoding approaches on freshwater sponges of Lake Tanganyika, which provides first insight into lineage diversity and morphological plasticity of this group. This is part of the Sponge Barcoding Project (www.spongebarcoding.org), which is the first worldwide barcoding project on any non-bilaterian taxa, and covers the complete taxonomic range of Porifera. We also demonstrate that the barcoding approach not only facilitates the assessment of biodiversity, but also sheds light on evolution and radiation of sessile invertebrates in Lake Tanganyika.

Evolution, distribution and chemotaxonomy of Dysideidae with anti-plasmodial metabolites

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Sponges of the family Dysideidae (Dictyoceratida) are renowned for their diversity of secondary metabolites. Recent biochemical profiling revealed an anti-plasmodial activity in Dysideidae of the Pacific, particularly in Lamellodysidea. Here we present a comparative chemotaxonomic and molecular analysis of selected Dysideidae from the Pacific and the Indian Ocean (New Caledonia, Great Barrier Reef, Fiji, Solomon Islands, Mayotte, Guam, Palau) based on the internal transcribed ribosomal spacer (ITS). We aim to gain insight into the phylogenetic relationships between the bioactive (and other) species in order to attempt a reconstruction of the anti-plasmodial metabolite biosynthesis evolution.

Progress and workflow of the Sponge Barcoding Project

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The Sponge Barcoding Project (www.spongebarcoding.org) is the first worldwide barcoding project on any non-bilaterian taxon, and aims to cover the complete taxonomic range of Porifera. The Sponge Barcoding Database is the prime access point for DNA signature sequences together with information on conventional morphological taxonomic characters of sponges. This unique combination of sponge-specific conventional taxonomic information and their DNA signature sequences is crucial for the use on sponges and the distinguishing feature to all other DNA databases. In its initial phase, the Sponge Barcoding Project currently constructs its backbone framework of DNA signature sequences primarily from specimens of the Queensland Museum (Brisbane, Australia), which hosts the largest sponge collection of the Southern Hemisphere. We present our DNA extraction-, PCR-, sequencing-, and data analysis pipeline for sponges as developed by our lab and report on the current status, progress and taxonomic coverage of the Sponge Barcoding Project.

NUMTs in the demosponge genome reveal conserved transposition mechanisms in metazoans

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We provide evidence for mitochondrial genetic material being transposed to the nucleus (NUMTs) in sponges. In the *Amphimedon queenslandica* nuclear genome NUMTs represent nearly all regions of the mitochondrial genome. As previously observed in eumetazoans, sponge NUMTS are small and do not encode functional protein or RNAs. They appear to be transposed via a DNA intermediate but are inserted into nuclear regions of high gene density. This is in contrast to observations in bilaterians, where NUMTs are typically in areas of few genes. We infer from the shared characteristics of sponge and bilaterian NUMTs, the last common ancestor to all extant metazoans used DNA-based mechanisms to transpose mtDNA fragments into the nuclear genome. These processes may have contributed to the initial compression of the crown metazoan mitochondrial genome, although it is unclear the role they have played in the differential evolution of the genome in sponge and eumetazoan lineages.

A new *Symbiodinium* sp. lineage in Atlanto-Mediterranean *Cliona viridis*

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Marine sponges in the Clionaidae family are cryptic species known for their boring activity. Some of the most competitive and destructive bioeroding sponges belong to the '*Cliona viridis* complex' and contain associated zooxanthellae. The presence of these microorganisms has shown to improve the host's growth, survival rate and boring activity. Although excavating sponges are important structure builders in coral reefs, studies on their association with dinoflagellates are scarce and recent. In this context, a PCR based strategy was applied to detect and identify zooxanthellae associated with these sponges. Specimens of *Cliona viridis* (Schmidt, 1862) were collected by scuba diving from multiple sites in the Portuguese archipelagos of Azores and Madeira and along the coast of Blanes (Spain), between 2005 and 2006. Using a newly designed dinoflagellate-specific primer (Dino18SF) and a eukaryotic universal primer (NL4), we amplified and sequenced a 500 bp fragment of the ITS+28S rDNA region. The sequences obtained are highly homologous among them (around 98% homology) but show low identity when compared with sequences available in public databases (GenBank). Maximum identity (around 86%) was observed with potentially free-living *Symbiodinium* spp. sequences isolated from Japanese coral reef sand. Since few studies on associated zooxanthellae have focused on the eastern Atlantic, these results point to the existence of a different lineage of *Symbiodinium* living in association with clionaid sponges inhabiting temperate waters.

Taxonomic revision and phylogeny of *Crambe* Vosmaer, 1880 and *Monanchora* Carter, 1883 (Crambeidae, Poecilosclerida, Demospongiae)

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Sponges belonging to *Crambe* and *Monanchora* have a global distribution and constitute an important source of guanidine polycyclic alkaloids with antibacterial, antineoplasic and antiviral activities. These genera are currently defined according to an Aristotelian rationale, where *Crambe* is characterized by the presence of desmas and *Monanchora* by the lack of these microscleres. Their strong chemical similarity brings them even closer. Our objectives were to revise the taxonomy, re-describe known species, describe new species and reconstruct the phylogeny of *Crambe* and *Monanchora*, in order to verify their monophyly. Approximately 200 specimens were examined, including type and/or additional material of both genera, originating from important collections around the world. Collection and in situ photos were made by several researchers along the Brazilian coast, in the Caribbean, and in the south-eastern Pacific. The phylogeny of *Crambe* and *Monanchora* was reconstructed using *Discorhabdella* spp. (Crambeidae) as outgroup. Phylogenetic analyses resulted initially in 3894 most parsimonious trees (mpts) with 85 steps, reduced to 15 mpts with 18.6 steps (Consistency index = 0.6226) after successively weighting characters. The Strict consensus revealed a monophyletic *Monanchora*, characterized by the loss of desmas, with *Crambe* paraphyletic to *Monanchora*. Hence, in a morphologic phylogenetic perspective, *Monanchora* should be abandoned, with its species transferred to Crambe, the oldest name. In order to follow current practice in Demosponge classification, we propose keeping both genera as subgenera of *Crambe*. Following is a list of the taxonomic decisions taken. (1) *Monanchora dianchora* and *M. unguiculata* are synonyms of *C.* (*M.*) *viridis*. (2) *Monanchora enigmatica* belongs to *Echinochalina* (*E.*). (3) *Monanchora laminachela* and *M. pulchra* were transferred to *Stelodoryx*. (4) *Monanchora* quadrangulata belongs to *Protosuberites* (Suberitidae, Hadromerida). (5) *Clathria* (*Isociella*) *selachia* is a valid species of *C.* (*M.*). (6) *Crambe acuata* comprised a complex of four species, three of which belong to *C.* (*C.*): *C.* (*C.*) *acuata*, *C.* (*C.*) *chelastra* (revalidated in the present study), *C.* (*C.*) sp. nov.; and, in part, to *C.* (*M.*) *viridis*. (7) *Monanchora arbuscula* comprised a complex of several species, four of which belonging to *C.* (*M.*): *C.* (*M.*) *arbuscula* and three new species. (8) *Monanchora clathrata* comprised a complex of three species, two belonging to *Crambe*: C. (M.) *clathrata* and a new species. (9) *Monanchora alaskensis* was *transfered* to *Amorphoclada* (*Myxillidae*), revalidated in the present study. (10) A total of eleven new species of *C.* (*M.*) (five from the Atlantic, six from the Indo-west Pacific) and one of *C.* (*C.*) (W Indian Ocean) were described. Accordingly, 12 species are now recognized in C. (C.) and 18 in C. (M.): C. (C.) *acuata*; *C.* (*C.*) *amarilla*; *C.* (*C.*) *chelastra*; *C.* (*C.*) *chilensis*; *C.* (*C.*) *crambe*; *C.* (*C.*) *erecta*; *C.* (*C.*) *maldonadoi*; *C.* (*C.*) *oamaruensis*; *C.* (*C.*) *panamensis*; *C.* (*C.*) *tailliezi*; *C.* (*C.*) *tuberosa*; *C.* (C.) sp. nov.; *C.* (*M.*) *arbuscula*; *C.* (*M.*) *clathrata*; *C.* (*M.*) *laevissima*; *C.* (*M.*) *lipochela*; *C.* (*M.*) *selachia*; *C.* (*M.*) *stocki*; *C.* (*M.*) *viridis*; and *C.* (*M.*) spp. nov. 1-11.

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The UK canal system may have affected freshwater sponge distribution

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Using sponges as model organisms, this study examines how changing waterway connectivity has affected freshwater populations. Historically, many UK freshwater systems were discrete entities. However, the construction of canal waterways over 200 years ago potentially had an important impact by first increasing connectivity between systems and then during the mid-twentieth century as many canals became heavily polluted and/or fell into disrepair, returning to a fragmented situation. Recent restoration of many waterways again raises the potential for increased connectivity among water bodies. Thus, the network of natural freshwater systems and anthropogenic canal habitats represents a complex matrix of historical isolation and more recent connectivity. This changing landscape has altered environmental conditions and availability of sponge habitat. From 2008-10, sponge sampling was conducted at >60 sites in the Shropshire Union Canal system measuring differences in environmental factors such as salinity and total dissolved solids. Our taxonomic and ecological findings suggest that freshwater sponges have a wider distribution in the UK than previously reported. Using morphological techniques, two putative sponge species, *Spongilla lacustris* Linneaus, and *Ephydatia fluviatilis* Linneaus have been identified from this canal system. Our data suggest that the canal system provides a conduit for sponge dispersal and further work to determine phylogenetic links between sponge populations is in progress.

Revision of Israeli calcareous sponges (Porifera, Calcarea) using molecular and morphological characters

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Calcareous sponges are usually small and inconspicuous. Hence, they are often neglected by marine biologists. Little is known about the calcareous sponges of the Mediterranean coast of Israel. The first and only evaluation of the Israeli diversity was conducted by Tsurnamal in 1962-1966, who recognized ten species in coastal waters down to 7m depth. Since this work, the taxonomy of calcareous sponges has been extensively modified and some of the species described by Tsurnamal are no longer valid. In the present study, calcareous sponges were sampled from 7 different shallow (1-5m deep) habitats and 3 deep (30m) sites along the Israeli cost. Histological sections were prepared in order to analyze the organization of the skeleton. Spicule preparations were photographed and measured. For the reconstruction of phylogenetic relationships, the 18S rDNA gene and the nuclear gene coding for ALG-11 were sequenced. From the sub-class Calcinea, Tsurnamal identified four species. Two of them could be collected during this work: *Clathrina contorta*, which was very abundant in shaded holes in the rocky substrate and *Clathrina coriacea*. The other two species, *Clathrina rubra* and *Clathrina clathrus* were also recorded before the onset of this research, but they were not found during this survey, although *C. clathrus* was described as abundant in Tsurnamal work. A fifth Calcinea species formed large colonies (ca. 5-8 cm in diameter) on the upper, well illuminated, part of the artificial pillars of the coal dock in Hadera, unlike all other Calcarea specimens that were sampled in crevices, notches or under rocks. Few points suggest that this species might be a new invasive species in Israel. First, it clustered in molecular analyses with *Leucascus* sp. (with low support), from a pacific origin. Second, another exotic species, the gorgonian coral *Acabaria erythraea* was reported from this site. Third, it was not observed in other locations. Additional work is needed to confirm that this is indeed an invasive species. Regarding the sub-class Calcaronea, Tsurnamal identified five species and described a sixth new species *Leuconia belemnifera*. In the present study we could only identify three of Tsurnamal's species: *Scypha raphanus*, *Leuconia globosa* and *Leuconia solida*. Today these two last species are considered to be synonyms of *Leucetta solida*, and to belong to Calcinea. Our molecular results suggest that individuals called *Leuconia globosa* and *Leuconia solida* by Tsurnamal are indeed two different Calcaronea species and thus not *Leucetta*. Further morphological work is thus needed to determine the taxonomical identity of these individuals. In addition, a unique specimen collected in 30m depth clustered with *Sycettusa* sp. and *Vosmaeropsis* sp. with high bootstrap support, based on the analyses of the 18S rDNA. Although this species was not described by Tsurnamal, this is probably not a recent migrant. It might have been missed in previous study that sampled only shallow waters. Since the opening of the Suez Canal, Red Sea migrants have profoundly modified the Levant fauna. Our results thus suggest that unlike other animal groups the Calcarea fauna did not change much in the last 50 years.

Three new species of *Tetilla* (Spirophorida, Tetillidae) from the coast of Bahia, Northeastern Brazil

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Sigmaspiras and triaenes are the main features shared by sponges of the family Tetillidae Sollas, 1886, as well as oxeas, auxiliary spicules (anfitrianios, calthrops, etc.) and radial architecture (Van Soest & Rützler, 2002). This family is the most representative of the Order Spirophorida Bergquist & Hogg, 1969, with eight genera and 150 species (Van Soest & Hooper, 2002). The genus *Tetilla* Schmidt, 1868 is defined by not having porocalices, cortical specialization and accessories megascleres. Most species have sigmaspiras, but seven species lack microscleres, including the typespecies of the genus, *Tetilla euplocamus* Schmidt, 1868. The other species devoid of microscleras are *Tetilla enoi* Brändsted, 1934; *T. globosa* (Bear 1906); *T. pedifera* Sollas, 1886; *T. radiata* Selenka 1879; *T. schulze* Kieschnick, 1898 and *T. truncata* Topsent, 1890. The genus *Craniella* Schmidt, 1870 is close to *Tetilla* by having globular sponges without porocalices, differing only by the presence of cortex. There were previously 45 valid species of *Tetilla*, with 10 recorded from the Atlantic and only two from Brazil: *T. euplocamus* Schmidt, 1868 and *T. radiata* Selenka, 1879. This paper describes three new species of the genus *Tetilla* for the State of Bahia, from three different localities, at shallow water and soft bottom. *Tetilla pentatriaena* n. sp. occurs on the northern coast of Bahia, off Camaçari, between 23 and 28m depth, in Salvador (depths < 20m) and inside of Camamu Bay, south coast of Bahia (depths < 16m). *Tetilla muricyana* n. sp. and *Tetilla rodriguezi* n. sp. were both collected in the mangroves of Camamu Bay, at less than 5m depth. *Tetilla pentatriaena* n. sp. is the only species of the genus with five categories of triaenes: two of protriaenes and three of anatriaenes. *Tetilla muricyana* n. sp. and *Tetilla rodriguezi* n. sp. are the only species of the genus occurring in mangrove and is distinguished from most species of *Tetilla* for failing microscleres. *T. muricyana* n. sp. differs from all species by having a secondary skeleton of oxeas organized in a reticulate pattern. *T. rodriguezi* n. sp. differs from species without sigmaspires by the presence of three sizes of oxeas, two of trichoidal protriaenes and one of rhizoidal anatriaenes. Adding up these three new species with one of *Craniella* already registered for the area, *Craniella quirimure* Peixinho et al., 2005, the Camamu Bay has now four records of Demospongiae. Moreover, *T. muricyanna* and *T. rodriguezi* represent the first report of the genus for mangroves. Our results show that these environments seem to have high potential for research of new records and new species of marine sponges in Brazil. The genus *Tetilla* has a general broad distribution pattern, occurring in all oceans, with distinct tropical and subtropical centres of diversity and only one species recorded from cold water: *Tetilla geniculata* Marenzeller, 1886 (at Jan Mayen and Savalbard Islands, in the Norwegian Arctic territory).

Microbial Diversity Associated with the Giant Barrel Sponge, *Xestospongia muta*

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The giant barrel sponge, *Xestospongia muta*, is one of the most prominent coral reef sponges in the Caribbean; its large size and high abundance provides habitat for a large number of macrofauna. Many sponges, including *X. muta*, have also been documented to harbor various microbes, which can influence sponge metabolism and the cycling of inorganic nitrogen, by nitrogen fixation, nitrification and denitrification. Cyanobacteria, in particular, have been increasingly studied, as they are known to participate in carbon and nitrogen cycling, the latter via nitrogen fixation. Nitrogen is a limiting nutrient in tropical waters and the process of nitrogen fixation adds biologically available nitrogen to the system. While it is known that *X. muta* contains cyanobacteria, it is not known if these or other bacteria in *X. muta* are capable of nitrogen fixation. This study characterized the taxonomic and functional diversity of the bacterial community of *X. muta*, focusing on the cyanobacterial population but including the nitrifying and denitrifying communities. Sponge samples from reefs near Little Cayman Island were collected from depths of approximately 9 – 91 m and the microbial community was examined using cyanobacterial specific primers for 16s rDNA and primers for nifH to examine if nitrogen fixing bacteria were present. Cyanobacteria including *Candidatus Synechococcus spongiarum*, *Synechococcus* sp., *Chroococcales* sp. and *Xenococcus* sp. were isolated primarily from the pinacoderm of *X. muta* and were observed over the entire depth gradient, while *Candidatus Synechococcus spongiarum* may be more common at the deepest depths. The nifH gene was amplified from *X. muta* pinacoderm, as were the genes for nitrification (amoB), denitrification (nirS) and ANAMMOX (amx) from the mesohyl. The results generated so far indicate a diverse cyanobacterial community that may differ in shallow (9 m) verses deep water (91 m). There may also be mirohabitat variation within each sponge that allow for taxonomic and functional differences of bacterial symbionts in *X. muta*. There appears to be potential for nitrogen fixation, which may contribute 'new' nitrogen to the sponge and to the reef, the extent of which is not known, but may be important given the large size of the sponge and high concentration of cyanobacteria. Future work will include investigating complex nitrogen cycling in *X. muta*, which will improve our understanding of nutrient cycling on Caribbean coral reefs.

Sox genes in the calcareous sponge *Sycon cilliatum*

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Sox genes encode transcription factors involved in development of animals, playing a variety of functions as diverse as sex determination and cell differentiation. Recent studies demonstrated that genes belonging to the Sox family are also present in the most basal metazoans, the sponges. For example, in the fully sequenced genome of the demosponge *Amphimedon queenslandica*, four Sox genes have been found and could be placed in three of the five major invertebrate Sox classes: SoxB, SoxC and SoxF. To gain insight into evolution of the Sox family of genes, we are investigating these genes in the calcareous sponge, *Sycon ciliatum*. This species is commonly found in Norwegian fjords and has been chosen as a new model system by our research group for developmental biology studies. Up to date, we have identified two Sox genes within an EST dataset generated from adult, larval and juvenile samples. Based on the domain structure and HMG box sequences, our phylogenetic analysis of *S. ciliatum* Sox genes suggests that they belong to two classes, SoxB and SoxE. To better understand the role of Sox genes in sponge development, we are currently studying the expression patterns of the identified genes using in situ hybridization. SoxB is dynamically expressed during *Sycon* development, beginning in early embryonic stages and continuing in differentiating cells in the juvenile sponges. Determining the developmental expression patterns of *S. ciliatum* Sox genes and the phylogenetic relationships with their sponge and basal metazoan homologues will provide insights into evolution and function of these important developmental regulators.

Qualitative variation in colour morphotypes of *Ianthella basta* (Porifera: Verongida)

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In recent years, sponges have dominated marine natural products research producing a large variety of structurally diverse secondary metabolites many of which possess interesting biological activities. Sponge secondary metabolites represent over 30% of isolated metabolites and are implicit in over 75% of antitumor patent registrations [1, 2]. Sponges harbour dense and diverse populations of microorganisms and numerous studies have attempted to ascertain the role microbes play in the production of these secondary metabolites. In addition to this high chemical and microbial diversity, sponges also exhibit plasticity of taxonomically relevant morphological characters. The genus *Ianthella* (order Verongida), predominately produces bromotyrosine derived bioactive metabolites including Bastadin-5, a probe for understanding the role of immunophilins in regulating sarcoplasmic reticulum Ca2+ transport [3]. *Ianthella* sp. found throughout the Indo-pacific present a number of colour morphotypes, most commonly yellow and purple. Specimens nominally identified as *Ianthella basta* were collected from Orpheus Island on the Great Barrier Reef and analysed using genetic (cox 1 mitochondrial gene and ITS-2 region), chemical (mass spectrometry) and microbial (denaturing gradient gel electrophoresis and 16S rRNA clone library analysis) techniques. Analysis of both genetic markers confirmed the taxonomy as *I. basta*, and was found to be consistent between the yellow and purple colour morphotypes with no significant divergence observed. The two colour morphotypes did, however, correspond to two distinct mass spectral profiles; the yellow containing predominately bastadins and the purple containing both bastadins and araplysillins [4]. Whilst the microbial community within the yellow colour morphotype of *I. basta* is highly conserved over a wide geographic range [5], DGGE analysis revealed distinct differences between the yellow and purple colour morphotypes. Further comparison of the microbial communities by 16S rRNA gene sequencing revealed some small differences in the microbial communities between the two colour morphotypes. Whilst there was a complete dominance of Gamma and Alphaproteobacteria in the yellow colour morphotype, the purple morphotype also contained representatives of the Deltaproteobacteria, Cyanobacteria and Nitrospira. Future research will investigate the possible role of these microbial species in the putative production of both the bastadin and araplysillin compound classes. These results have potentially important implications to ecological investigations of sponges whose morphology is fundamentally dynamic, stressing the need for precise taxonomic, chemical and microbial descriptions.

Inputs of symbiont-derived nutrition differ across sponge species: not all sponge-microbe interactions are equal

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Abundant and diverse microbial communities are present within some sponge species. These microbes may allow the sponge access to products of novel metabolic pathways, and thus increase growth and competitive ability in oligotrophic waters. Although several studies suggest that symbiotic cyanobacteria may provide supplemental nutrition to host sponges, the underlying questions of whether nutrients are actually exchanged and how important this input is to the host sponge remain unanswered. We addressed these questions by combining two methods that had previously been utilized only in isolation: 1) field-based shading experiments that reduce the input of photosynthetically fixed carbon to the sponge host and 2) stable isotope analyses of sponge and symbiont cell fractions to trace inputs of symbiont-derived nutrition to the host sponge. By combining these methods, we investigated how inputs of symbiont-derived nutrition change under shaded conditions. We conducted experiments using four sponges that show a range of interactions with microbial symbionts: *Aplysina fulva*, *A. cauliformis*, *Neopetrosia subtriangularis*, and *Niphates erecta*. The sponge *N. erecta* lacks cyanobacterial symbionts and had a higher growth rate under shaded conditions, with no difference in chlorophyll a concentration across treatments. Isotope values suggest that this sponge obtains the dominant part of its nutrition from particulate organic matter in the water column. Sponges hosting cyanobacterial symbionts (*Aplysina* spp. and *Neopetrosia*), however, underwent a drastic reduction in growth rates and chlorophyll a concentrations under shaded conditions, suggesting that inputs of photosynthetically derived nutrition are important to these sponges. Isotope analyses on sponge and bacterial fractions of these three species provide evidence that these sponges obtain nutrition from their microbial symbionts, but that inputs of carbon and nitrogen differ across these species. For instance, while *A. cauliformis* displays delta-15N and delta-13C values that are suggestive of a transfer of both carbon and nitrogen from symbionts to host cells, values for N. subtriangularis suggest that this sponge receives only carbon from its symbionts. Further, isotope values for *A. fulva* suggest that this species receives only nitrogen from its symbionts, with little evidence for a transfer of carbon. Changes in these interactions under shaded conditions vary across sponge species, indicating that some sponge-microbe interactions may be more flexible under differing environmental conditions. Together, manipulative shading experiments and stable isotope analyses provide strong evidence for the input of symbiont-derived nutrition to some host sponges. Our results also suggest that sponge-cyanobacteria interactions are not functionally equivalent across host species.

Paraleucilla magna in western Mediterranean: growth dynamics, reproduction, and ultrastructure features.

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Paraleucilla magna is an alloctonous species of unknown origin, which has proliferated in the western Mediterranean in the last decade. Knowledge on the *P. magna* biology in the newly colonized areas appears necessary to asses its invasive potential. Here, we describe the life cycle and the main reproduction features of *P. magna* in the western Mediterranean. An area of 20 x 20 cm^2 was scraped from the substrate (5 replicates) and the early recruits of calcareous species (from 1.4 mm in diametre on) were carefully separated under a stereo microscope. Potential *P. magna* recruits were confirmed by amplification of nine species-specific microsatellite markers, until the developement of the species characteristics allowed us to identify it morphologicaly. The species life cycle was unambiguously annual at the study populations. It showed a clear seasonal pattern with complete disaparition of the adults in July-early August, after larval release in June, and a re-colonization (or not) of the area by new recruits. Monthly growth rate showed a very high inter-individual variation. Size increased from 3mm ±1.3mm in September to 15.2mm ±10mm in March of the following year, when reproduction started. In March, the first occyctes, 15-20 µm in size, were noticed. They increased in size with time by feeding on modified maternal cells (trophocytes). Spermatic cysts were observed in a sole individual. Their cell origin stays obscure but their size (ca. 15 µm in diameter) excludes the transformation of a whole choanocyte chamber into a spermatic cyst, as reported for demosponges. From May on, embryo and larvae in different developmental stages were observed. Cleavage was asynchronous and occurred inside a membrane formed by cells and surrounded by collagen. Stomoblastulae, 30-40 µm in size showed macromeres and micromeres clearly differentiated. The macromeres had a large central nucleolated nucleus; the micromeres a small apical nucleus. Micromere cilia were compacted at the inner larval zone. Charriant-like cells with spermatic material were observed surrounding the stomoblastula. They contained a dense to electrons, irregular body with an acrosome-like zone. Ciliated amphiblastula, ca. 50 µm in size, ovoid and hollow, were surrounded by a three–layer membrane, even when the cilia were clearly faced outwards. This suggests that the whole process of inversion happenned inside the so-called "placental" membrane, instead of moving into a choanocyte chamber during inversion, as often described for Calcaronea. Four cross cells, placed at the four cardinal directions of the larval ecuator, were packed with three types of reserve materials. Maternal microgranular cells were often visible at the inner, hollow space. Collagen was profusivelly formed in cell vacuoles and excreted to the inner larval cavity. The annual life cycle of the species, an stochastic recruitment success, a patchy proliferation, and a highly variable growth rate, altogether allow to predict a moderate impact of *P. magna* on the native assemblages of western Mediterranean.

Taxonomic survey of the Hawaiian shallow-water sponge fauna with 21 new records

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The Hawaiian Archipelago is among the most isolated land masses on Earth. This high geographic isolation has strongly restricted colonization from the Indo-Pacific, and lead to relatively poorly diversified assemblages with high rates of local endemism. About 20% of its marine invertebrate and 24% of its fish species occur only in the Hawaiian Archipelago, and have probably evolved from ancestral organisms of Indo-Pacific origins. The aim of this study was to survey the shallow-water sponge fauna (1-25 m) of the Hawaiian Archipelago. For this, specimens were collected during several surveys conducted between 2006-2008. The sponge fauna of the archipelago is not well known, with the most recent taxonomic studies being conducted in the 1970s. We identified 50 species from our collections, most of them having Indo-Pacific distributions. Of these, 21 species represent new records for the archipelago: *Spheciospongia solida*, *Protosuberites epiphytum*, *Pseudosuberites andrewsi*, *Batzella aurantiaca*, *Lissodendoryx* (*Lissodendoryx*) *similis*, *Monanchora clathrata*, *Monanchora quadrangulata*, *Iotrochota baculifera*, *Iotrochota purpurea*, *Strongylamma wilsoni*, *Mycale* (*Aegogropila*) cfr. *lilianae*, 1998, *Mycale* (*Carmia*) *toxifera*, *Mycale* (*Mycale*) *phyllophila*, *Julavis levis*, *Dragmacidon* cfr. *halichondroides*, *Topsentia halichondrioides*, *Cladocroce burapha*, *Haliclona* (*Haliclona*) cfr. *fibrosa*, *Dactylospongia metachromia*, *Darwinella australiensis*, and *Dictyodendrilla* cfr. *dendyi*. Many of the sampling locations of this study were previously surveyed by others, and therefore it is likely that several of these new records may represent recent introductions. Additionally, several sponge taxa identified during this study, such as (*Tedania* (*Tedania*) *ignis*, *Biemna fistulosa*, *Mycale* (*Zygomycale*) *parishii*, *Gelliodes fibrosa* and *Haliclona* (*Soestella*) *caerulea* have already been considered recently introduced or cryptogenic species (Carlton and Eldredge 2009). Our study suggests that the shallow-water Hawaiian sponge fauna has strong links with the Indo-Pacific, particularly the Western Pacific, the Indo-Malay region and Australia (Bergquist, 1967, 1977). Furthermore, this survey detected the presence of probable, several alien species of Atlantic-Caribbean origin within the fouling community, and highlights the need for further taxonomic surveys.

Polyphyly of the genus *Axinella* and of the family *Axinellidae* (Porifera: Demospongiae)

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The phylogeny of Demospongiae has been revisited recently by molecular phylogeny. They are now considered to be composed of four groups (G1 to G4). Among G4, several orders seem to be not monophyletic like Halichondrida. Among this order, the genus *Axinella* is difficult to define on the basis of morphological characters and includes a heterogeneous assemblage of species. Several previous authors have suspected the polyphyly of both this genus and the family Axinellidae. To clarify the phylogeny of *Axinellidae* and *Axinella*, we propose a new hypothesis based on the phylogenetic study of two molecular markers (18S and 28s rRNA) and used secondary structures information. In our analyses, Axinellidae and *Axinella* are polyphyletic assemblages. The 15 species of Axinellidae in our dataset belong to five clades and the nine species of Axinellato three clades. One contains the type species of the genus: *A. polypoides*, plus *A. aruensis*, *A. dissimilis*, *A. infundibuliformis*, and *A. vaceleti* that can be named *Axinella*. Together with *Dragmacidon* species, they form the Axinellidae that share specific bulge in their secondary structure. A new clade *Cymbaxinella* is named, following the PhyloCode, and including *C. damicornis*, *C. verrucosa*, *C. corrugata* and *C. cantharella*. The clades *Agelas* and *Cymbaxinella* constitute a new clade: Agelasida supported by the presence of specific secondary metabolites (pyrroles) and bulge. *Axinella cannabina* is reallocated to *Acanthella*, they share the presence of flexuous spicules and specific bulge. Except for *Acanthella cannabina*, which shares clear morphological characters with *Acanthella acuta*, no morphological synapomorphies can be found for the *Cymbaxinella* or Axinellidae clades. It is thus urgently necessary to find other data sets of characters. A search for other characters based on cytology, embryology and secondary metabolites would be helpful as a basis for future progress. This work is an attempt to build a framework for the phylogeny of taxa allocated to *Axinella* and Axinellidae in the traditional classification.

Impacts of nutrient enrichment and disease on the caribbean sponge *Aplysina cauliformis* and its cyanobacterial symbionts

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Recent declines in the health and condition of coral reefs worldwide have been attributed to both natural and anthropogenic sources, but most likely result from interactions among multiple types of stressors. Coral reefs thrive in oligotrophic waters, however, elevated levels of nutrients from runoff of fertilizers or sewage can lead to potentially stressful eutrophic conditions. In addition, emerging diseases of marine organisms are considered major causes of the accelerating destruction of coral reefs, particularly in the Caribbean. Although relatively little is known about the dynamics of diseases on reefs, it has been suggested that their prevalence is due to increases in the ranges or virulence of pathogens and/or to reduced resistance in their hosts, likely due to changing environmental conditions. In addition to the more familiar coral diseases, sponge diseases have recently emerged as potential structuring forces on reefs across the globe. *Aplysina cauliformis* is an abundant member of the Caribbean sponge community, and is susceptible to *Aplysina* Red Band Syndrome (ARBS), a disease that causes reduced growth and survival in A. *cauliformis* on Caribbean reefs. To assess the interacting effects of nutrients and disease on *A. cauliformis*, we provided nutrient enrichment to healthy and diseased sponges in a factorial experiment. Impacts on ARBS virulence (rate of lesion growth) and host response (growth and physiology of both the sponge and its cyanobacterial symbionts, *Synechococcus spongiarum*) were assessed. Overall, disease had a much greater impact on the growth and physiology of *A. cauliformis* and its cyanobacterial symbionts than did nutrient enrichment. ARBS lesions on diseases sponges increased in size rapidly regardless of nutrient levels. Diseased sponges were equally likely to develop additional lesions whether they were exposed to elevated nutrient levels or not. Healthy sponges grew faster and had higher concentrations of soluble protein, a measure of overall sponge condition, than did diseased sponges. Cyanobacterial symbiont density, chlorophyll a concentrations, and fluorescent yield were not significantly impacted by nutrient enrichment. There was an overall increase in symbiont density and fluorescent yield over the duration of the experiment, which may be related to overall enhanced metabolic rate as summer progressed. However, ARBS infection did appear to inhibit proliferation of symbiotic *Synechococcus* cells in diseased sponges. The rapid proliferation of symbionts in healthy sponges was associated with a decline in chlorophyll a concentration over time in healthy sponges, suggestive of a physiological trade-off between symbiont cell proliferation and photosystem development. Our study of interactions between two different types of environmental stressors highlights the importance of disease as a factor structuring sponge populations.

Hexactinellida from the ANT XXIV/2 (SYSTCO)-expedition, including the rediscovery and detailed description of the poorly known genus *Lonchiphora* Ijima, 1927

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We present the hexactinellid sponges sampled in the Weddell-Sea during ANT 24-2 SYSTCO expedition (30.10.2007 – 31.01.2008) of RV Polarstern (Janussen 2010). These include *Bathydorus spinosus* Schulze, 1886, *Caulophacus* (Oxydiscus) *weddelli* Janussen et al. 2004, six *Rossella* species: *Rossella antarctica* Carter, 1872, *Rossella fibulata* Schulze & Kirkpatrick, 1910, *Rossella levis* (Kirkpatrick, 1907), *Rossella nuda* Topsent, 1901, *Rossella racovitzae* Topsent, 1901, *Rossella vanhoeffeni* (Schulze & Kirkpatrick, 1910), and a new species of genus *Lonchiphora* Ijima, 1927. The rediscovery of *Lonchiphora* in the Antarctic Ocean was a big surprise, as it was formerly known only from the Sagami Bay, Japan. This distribution may be explained by a concept on the evolution of Cenozoic seaways by Lawver and Gahagan (2003). According to this hypothesis, a circumglobular current existed throughout almost the entire Cenozoic, bringing equatorial waters from regions near Japan southwards through a passage east of Africa into the Antarctic area of the Weddell-Sea and then northward towards the Equator. Thus, there was a strong inflow from northerly seas towards the Weddell-Sea throughout a prolonged period of time, and it is likely that *Lonchiphora* followed this current. According to our hypothesis, the Antarctic species is probably the younger, derived from a northern species that migrated southwards through the deep-sea basins surrounding Japan. The recent records of *Lonchiphora* may represent relics of formerly widespread occurrences, which were decimated due to changes of environmental factors on smaller and bigger scale, important for hexactinellid sponge societies (Gutt and Koltun 1995). *Lonchiphora* is characterized by a specific spicule, the lonchiole, which is unique for the genus. It was described, but not illustrated, by Ijima (1927) and was therefore a kind of mystery to sponge scientists until now. We documented in detail the lonchiole and some derivates, which we will present and discuss. Ecological analysis of hexactinellid assemblages show that three distinct associations of glass sponges exist, replacing each other with increasing depth. These are characterised by the following genera: *Rossella* spp. on the shelf, *Bathydorus* spp. on the continental slope and *Caulophacus* spp. in the abyssal.

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Morphologic variability and distribution of Clathria species from the Southern Gulf of Mexico.

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This study presents the morphologic variability of eight Clathria species from the Southern Gulf of Mexico in light of the slight similarities between them. Specimens of *Clathria* (*Clathria*) *foliacea*, *C.* (*C.*) *prolifera*, *C.* (*Microciona*) *calla*, *C.* (*M.*) *spinosa*, *C.* (*Thalysias*) *venosa*, and *C.* (*T.*) *virgultosa* were collected by scuba diving and dredging from different points of the Southern Gulf of Mexico (Veracruz, Campeche, Yucatan and Quintana Roo), from shoreline to 60 m depth. Identification was achieved by analysis of external morphology, skeletal architecture, spicule measurements and scanning electronic microscopic photographs. Besides, *C. foliacea* Topsent, which has not been formally examined since the original species description, is described for the first time, along with *C. spinosa* in the Mexican coasts. Differences versus similarities of the species in question are illustrated as well as a tentative identification key is provided. The distributional pattern of species is enlarged.

Cloning and expression analysis of the vasa-related gene in fresh-water sponge *Spongilla lacustris* (Demospongiae, Haplosclerida, Spongillidae)

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Vasa-related genes encode the members of DEAD box protein family. Protein Vasa is a ATP-dependent RNA helicase which is involved in germ cells specification in metazoan animals. The universal occurrence of *vas*-related genes and their germ-line expression has been demonstrated in a number of studies. The proteins of DEAD box family are involved in splicing processes, RNA editing, translational regulation and mRNA degradation. Vasa protein and sometimes *vasa* mRNA forms part of ribonucleoprotein particles that are putative participants of processing, localization and regulation of germ-line mRNA. Morphologically these particles have been identified as mitochondrial clouds or "nuage". In the lower invertebrates, such as *Hydra* and *Planaria*, this ribonucleoprotein particles and *vasa* expression occur both in multipotent stem cells and germ-line cells (i-cells in Cnidaria and neoblasts in Planaria). As it is assumed, the germ cells are a subpopulation of a multipotent stem cells in these animals. According to literature data, in different groups of sponges the germ cells derive from multipotent stem cells – archeocytes or somatic cells – choanocytes. In some sponges, including *Spongilla lacustris*, it was reported that the oocytes are derived from archeocytes, and spermatocytes are derived from choanocytes, suggesting that in one sponge both archeocytes and choanocytes can maintain their pluripotency. To clarify the origin of male and female gametes the 598 base pair fragment of *vasa*-related gene in fresh-water sponge *S. lacustris* was cloned. The *S. lacustris vasa* sequence showed high similarity with *vasa*-related gene of sponge *Ephydatia fluviatilis Povas1* (Mochizuki et al, 2000). The analysis of deduced amino acid sequence revealed four well conserved motifs, including DEAD. In order to investigate the expression of *S. lacustris vasa* gene, *in situ* hybridization in paraffin sections was carried out. Paraffin section was made using sandwich method, where one section was used for *in situ* hybridization and the other – for histological analysis. Till now only two stages of *S. lacustris* life cycle were studied: the sponges in a gemmulation process and in nonreproductive ones. We found *S. lacustris vasa* expression products in the subpopulation of pluripotent cells – archaeocytes. Expression products of this gene were not detected in the choanocytes. Our data indicate the heterogeneity of the archaeocyte population and do not confirmed pluripotency of choanocytes in this species.

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Sponge biodiversity of the United Kingdom

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During 2005 a survey of Rathlin Island, Northern Ireland found 128 sponge species including 29 species new to science, 3 new to the UK and 3 new to Northern Ireland. The sponge fauna of the UK is still poorly known, partly due to a lack of sampling effort in circalittoral depths and it is believed that there may be similar findings to be made elsewhere. The diving methodology used on Rathlin enabled the sampling of encrusting sponges and circalittoral habitats which have been under-sampled historically. This project used the Rathlin methodology to study the sponges of the Firth of Lorn (Scotland), Pembrokeshire (Wales), Sark (Channel Islands), Plymouth (England) and the Isles of Scilly (England). There were significant differences in regional faunas with some species showing a clear northern or southern bias in distribution. Some species recorded from the UK for the first time during the Rathlin project, including *Antho (Antho) brattegardi* and *Hexadella racovitzai*, were shown to be widespread and common in some areas. Several species new to science and one genus new to the UK (*Trachytedania*) were recorded. Here we discuss the survey methodology and preliminary findings on the biogeography of the UK sponge fauna.

Sponge biodiversity of the Falkland Islands

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Sponges have been recognised as one of the most important components of the Falkland Islands sublittoral fauna. However, there is little information available on species composition or abundance, with the only two previous detailed studies having been undertaken in the late 19th and early 20th centuries from dredged material. This diving survey of the sponge biodiversity of areas around Stanley, the Jason Islands (West Falkland), Sealion Island and Beauchêne (SE Falkland) provides new information, for many species *in situ* appearance is recorded for the first time. Twelve species new to science were recorded from the Jason Islands: as for other invertebrate groups many of these may be endemic as a result of the geographical isolation of the archipelago. There was a significant difference between the fauna of the Jason Islands which are influenced by the warmer waters of the Argentine Drift and Beauchêne which is influence by Antarctic upwelling. Here we discuss the survey methodology and preliminary findings.

Cytotoxicity of extracts from marine sponges from S. Miguel (Azores)

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Sponges are at present the most promising source of novel molecules with pharmacological potential. Our knowledge of the ecological roles of these compounds continues to grow as new techniques are developed to understand the complex relationships between sponges and their predators, competitors, microorganisms, and associated invertebrates (Valerie et al., 2006). In their long evolutionary history, sponges and their symbionts have evolved pathways which synthesize defence compounds which are both extremely potent and unique. These molecules are often promising leads for new chemotherapy agents. In this context, we screened four sponge species for cytotoxicity against HeLa tumour cell line. *Axinyssa aurantiaca*, *Haliclona cinerea*, *Hymeniacidon sanguinea* and *Cliona viridis* were collected by scuba diving near the coast of S. Miguel island (Azores). Methanol, dichloromethane and n-hexane extracts from sponge tissue were evaporated to dryness, dissolved in dymethylsulfoxide and tested for their antitumor properties, using the MTT colorimetric assay of Mosmann (Mosmann, 1983), with some modifications. *Hymeniacidon sanguinea* did not present cytotoxicity in the concentrations tested. The other species were active against HeLa cells but in different levels. The methanol extract of *Haliclona cinerea* and the n-hexane extract of *Cliona viridis* were two most active extracts, with EC50=7,45 μg/ml and EC50=27,8 μg/ml respectively. Further investigation into the cytotoxic mechanisms of the two more active extracts was carried out with fluorescence microscopy, which revealed that, in the concentrations tested, *Haliclona cinerea* induced death by necrosis, whilst *Cliona viridis* activated cell death by apoptosis.

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Population genetics of the introduced, calcareous sponge *Paraleucilla magna* in the western Mediterranean. Structure variation across years of three close populations.

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Foreign species are proliferating in the sublitoral areas of the Mediterranean in the last decades mainly due to man associated introductions. Foreign introduced species interact in several ways and directions with the autochthonous species (with non-predictable outcomes). Thus, knowledge on the capacity of proliferation of these species is mandatory for management decisions in Conservation policies. The introduced calcareous sponge *Paraleucilla magna*, has proliferated along the western Mediterranean during the last decade. The species distribution is highly patchy with dense populations in zones close to seafarms and slightly contaminated marinas. Biological studies showed that large individuals of the species disappeared after larval release (in early summer), although in some particular habitats such as mussel farms, the species has been recorded the whole year round. To gain knowledge about the proliferative capacity of the species once it has colonised a new area, and on the possible origin of the yearly recruits (whether philopatric or allopatric), we studied the genetic structure of three close populations of *P. magna* in the NE of the Iberian Peninsula (Blanes littoral) in three successive years. We sampled the "same" populations (i.e. the populations established in the same area) in 2006, 2007, and 2008 and genotyped them for nine microsatellite markers previously designed for the species (authors unpublished results). The nine microsatellites proved to be polymorphic in other populations previously analysed. Allele frequencies, heterozygosity, genetic variation, and the extent of asexual reproduction were estimated. The results point to a complete disappearance of the adults after larval release, and the establishement of new populations with an important contribution of recruits resulting from foreing larvae, which contrasts with the expected philopatry of those extremely short-life larvae (reported for calcareous sponges in general). These results also suggest a high vulnerability of the populations, as they seem to greatly depend on stochastic events such as weather conditions that may cause recruitment failure. Thus a relatively low negative impact of the species in native communities is expected.

Antimicrobial and anti-feeding defences of two Axinellid sponges

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The diverse natural products found within sponges mediate many of their ecological interactions such as feeding deterrence and defence against fouling and microbial pathogens. Sponges of the genus *Axinella* are especially known for their bromopyrrole compounds, which are often involved in feeding deterrence. Here we studied the chemical defence of the two co-occurring Mediterranean sponges *Axinella polypoides* (Schmidt 1862) and *A. verrucosa* (Esper 1794) collected from the Gulf of Naples, Italy, against microbial fouling by field and laboratory assays as well as the feeding deterrence against the generalist shrimp *Palaemon elegans* (Rathke 1837). Surprisingly, in these assays no activity was detected in the extracts of *A. polypoides*, while the butanolic part of the *A. verrucosa* extract showed activity in all assays. Using bioassay-guided isolation procedure we purified two antibacterial compounds from the latter extract: debromo-carteramine A, a new derivative, and the main compound of the extract hymenidin, which also showed activity in the feeding deterrence assay. In conclusion our results indicate different defence strategies in the two examined congeneric and sympatric sponge species and multiple roles of hymenidin in the defence of *A. verrocusa*.

'Sparkling Aurantoside': the mystery of some sciophilous sponges in the Indo-West Pacific

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A monumental marine biodiversity survey of macrobenthos on the Great Barrier Reef (GBR), Australia (http://www.reef.crc.org.au/resprogram/programC/seabed/index.htm), has unearthed an enigmatic, yet ubiquitous, component of the sponge fauna. From 2003 to 2006, the seabed was documented and sampled across more than 1,400 sites along the length and breadth of the GBR, with 72% of these sites yielding an unusual and distinctive new species of sponge. This species, which comprised more than 1/4 of the total invertebrate biomass collected, is made conspicuous by its characteristic eosinic yellow dye and sciophilous habit. Although it is the most common sponge in inter-reef habitats, the taxonomic affinities of this new species were obscure. Identification of this species creates more questions than it provides answers. Morphologically, it is most similar to pachastrellids; the presence of microrhabd-like sanidasters and the gross habit of encrusting and agglutinating benthic rubble are in broad agreement with *Dercitus* Gray and *Stoeba* Sollas. This new species is now described within *Dercitus*; although calthrops were found in a small proportion of the sample, warranting placement in *Dercitus*, the species lacks the consistent presence of tetraxons, making this classification slightly unsatisfying. Chemically, the sponge contains aurantosides. These complex sponge metabolites have been isolated previously only from species of the theonellid genera *Manihinea* Pulitzer-Finali, *Siliquariaspongia* Hoshino and *Theonella* Gray. The presence of aurantosides in the new species of *Dercitus* is strong evidence that it is a theonellid, not a pachastrellid, yet the absence of any rigid skeletal components contradicts such classification. Genetically, the new species is closest to theonellids. Analysis of DNA sequences (~1,200 bp of COI mtDNA and ~800 bp of 28S rDNA) supports a clear and very close relationship between this species and several other species of *Theonella*, and in turn, of this group to other theonellid genera such as *Discodermia* du Bocage. Again, the absence of desmas makes classification within Theonellidae awkward. Existing nomenclature does not fully accommodate this species. Morphological data support pachastrellid affinities, but disagree with chemical and molecular datasets. We have since found additional genotypes, displaying subtle morphological distinctions, but which share the yellow colouration (signalling the likelihood of aurantosides) and sciophilous habit of the new species of *Dercitus*. These types almost certainly represent additional species, suggesting there may be a complex of these enigmatic sponges in Indo-West Pacific waters. Our phylogenetic estimates establish convincingly the position of Theonellidae within Astrophorida, as a sister to clades containing geodiids and pachastrellids; this is in broad agreement with previous DNA-based studies. Our findings open wider questions about the relationships and allocation of theonellids within demosponges, and about the boundaries and diagnosis of pachastrellids within astrophorids. Here, we hypothesise an evolutionary scenario to explain the mystery of these sciophilous sponges, which have pachastrellid morphology but theonellid chemistry and genetics. Like all good stories, it is a tale of conflict (of data), loss (of skeletal components) and (niche) exploitation, of families united and families ripped apart, but ultimately, it is a tale of redemption, via a new natural classification.

Taxonomic position of *Eunapius subterraneus* (Porifera, Spongillidae) inferred from molecular data - a revised classification needed?

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The freshwater sponge *Eunapius subterraneus* was described in 1984 on the basis of its morphology and unique ecological features. It inhabits caves in the Ogulin karst area as the only known stygobitic sponge, and an endangered karst species. We used three genetic markers with different evolutionary rates in phylogenetic analyses of *E. subterraneus*. All of the markers exclude this sponge from the genus *Eunapius*. Based on our results, we emphasize the need for revision of the taxonomic classification of *E. subterraneus* as well as the need for a thorough re-evaluation of freshwater sponge systematics.

COI-barcoding reveals two cryptic *Tethya* species from the North Atlantic and the North-East Pacific

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Within the family Tethyidae (Demospongiae, Hadromerida) the genus *Tethya* is the most species rich. The 82 valid species represent two-thirds of all valid species of the family. The genus is widespread and common in nearly all marine waters. Globally, the highest diversity of the genus, with more than one third of the species, is found in the South-West Pacific along the temperate coasts of Australia and New Zealand. In contrast, until recently only three *Tethya* species for the Mediterranean Sea and the North Atlantic and one for the North-American Pacific coast were described. Interestingly, most of these and also other *Tethya* species are represented by extremely wide-spread sample locations (e.g. *T. citrina* and *T. californiana*) or are/have been regarded as cosmopolite (e.g. *T. aurantium, T. seychellensis, T. japonica* and *T. robusta*). Taking into account the general lack of valuable taxonomic characters in sponges, it seems likely that some reports on these species actually represent un-identified cryptic species. The aim of our study was to revise *Tethya* samples from underrepresented geographic regions with a holistic approach to characterize the species with molecular genetics and morphological techniques. We sequenced a part of the cytochrome oxidase subunit I (COI; Folmer fragment) which is located on the mitochondrial genome. Morphological species discrimination was performed on spicule and skeleton preparations, imaged by SEM and light microscopy. In addition, we established synchrotron radiation based x-ray microtomography (SR-µCT) and 3D reconstruction as a tool to visualize and analyse skeletal architecture and virtual spicule preparations and measurements in silico. COI turned out to be a suitable marker for species discrimination within the genus *Tethya* and also to reveal cryptic species. COI analysis provided evidence for two new species. Those were subsequently studied morphologically and morphometrically. We were able to confirm significant morphological characters which allowed us to describe *Tethya hibernica* for Northern Ireland and *Tethya leysae* for Barkley Sound, Vancouver Island, Canada. In the latter, SR-µCT 3D image data was included into a species description for the first time. The genetic distances between members of the genus *Tethya* as revealed by COI-barcoding seems to be larger than in most other sponge genera. The question arises whether *Tethya* represents an old genus or if its COI faced higher nucleotide substitution rates than other poriferans. Generally and similar to the situation in the Cnidaria, sponges display a low nucleotide substitution rate. Therefore, we assume that the radiation of the genus *Tethya* is a result of a very long evolutionary history. For a better understanding of the phylogeny of the genus *Tethya* and the relations among the other genera of the Tethyidae, more barcode sequences from members of the Tethyidae are needed and a revision of parts of taxa is desirable.

Stability of symbiotic microbial community structure during development and metamorphosis of the Caribbean fire sponge, *Tedania ignis*

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Although marine sponges can host diverse communities of symbiotic microbes, the process of symbiont acquisition remains unclear for most species. Symbionts can potentially be obtained directly from the parent (vertical transmission) or acquired from the surrounding environment (horizontal transmission). Our study examined the symbiotic microbial community associated with the Caribbean fire sponge, *Tedania ignis*. We sampled adult sponges, newly released larvae, and larvae held in isolation through their first 72 hours of development. Microbial community composition was measured using terminal restriction fragment length polymorphisms (TRFLPs). Adult sponges and newly released larvae showed a similar diversity of bacterial terminal restriction fragments (TRFs). Within 72 hours of release, all larvae had settled and metamorphosed. These new metamorphs displayed lower TRF diversity than the newly released larvae. A subset of TRFs dominated the symbiotic bacterial communities and were found in all three life stages, suggesting that these taxa are vertically transmitted. Clone libraries constructed from adults and larvae revealed three dominant phylotypes representing a clade of betaproteobacteria and two clades of Chloroflexi (green non-sulfur bacteria). Our observations suggest that the symbiotic microbial communities associated with *Tedania* are a combination of dominant, vertically transmitted symbionts and less abundant, environmentally acquired microbes. The horizontally transmitted taxa could result from either host feeding or microbial colonization of host surfaces and tissues. Future work will focus on DNA sequence analyses and localization of specific phylotypes using fluorescent *in situ* hybridization.

Phylogeny of the Hexactinellida: Phylogenetic reconstruction of the subclass Hexasterophora based on morphological characters

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As an old and probably most conservative taxon, the Hexactinellida is promising for the understanding of early Metazoan evolution (Mehl-Janussen 1999). However, since the Cenozoic the hexactinellid sponges are almost exclusively restricted to deep water environments, and therefore this Porifera class is largely under-investigated. So far, only a very preliminary model based on morphological characters exists (Mehl 1992) and a comprehensive molecular phylogenetic analysis of the Hexactinellida was published (Dohrmann et al. 2008). Within this project we investigate the phylogenetic relations between the families and partly also genera of the Hexactinellida, here beginning with the biggest subclass, the Hexasterophora. The resulting phylogenetic trees, based on molecular and morphological characters, will be compared with each other and with the classic systematics. So far, 145 generic characters of the taxon Hexasterophora have been compiled into a matrix by presence/absence data, and first attempts towards the phylogenetic analysis have been done. A main problem is the expression of the different character stages, which are very pronounced and taxonomically important within the Hexactinellida. For phylogenetic trees reconstruction, presence/absence data are most appropriate, whereas a complicated matrix with many character stages is unsuitable. However, this simplification of the character stages does not reflect accurately the evolutionary steps and may result in misleading phylogenetic models, this is true also for the necessary à priori decisions concerning homology versus homoplasy. Our analysis of selected hexasteroporan families resulted in good conformity with the existing systematics in most cases, especially in those taxa with high differentiation of diagnostic characters (e.g. the Euplectellidae), whereas the resolution within some other families is low. Based on the reconstructed phylogeny, we aim to gain further insight into hexactinellid evolution, e.g. geological age of the main taxa, paleogeography and evolutionary ecology of the Hexactinellida.

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Celtodoryx ciocalyptoides: the first documented record of a sponge species transferred from one world ocean into another by human activity

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According to the Delivering Alien Invasive Species Inventories for Europe (DAISIE), currently more than 1000 marine exotic species have been recorded in Europe with no records of sponges so far. Nevertheless, 15 sponge invaders have been described recently for the Dutch inshore waters (Van Soest et al., 2007), including a sponge that was firstly recorded in a well studied area in the Ria of Etel, Brittany, France in 1996. This sponge was later described as a new species and genus, *Celtodoryx girardae* by Perez et al. (2006), who concluded that it is probably an invasive species. Within several years C. girardae was found to occur successively in the Gulf of Morbihan, France, and Oosterschelde estuary, Netherlands. It is characterized by an extensive spatial spreading, and therewith it rates today among the dominant benthic megafauna in the shallow waters of the Gulf of Morbihan and Dutch inshore waters. During our recent survey of the Chinese Yellow Sea sponge fauna, we found an abundant species with close morphological similarities to *C. girardae*. The assumption that both specimens are of the same species was confirmed by molecular analysis. Further taxonomic studies have revealed that both the Chinese and European sponges are in fact conspecific with *Cornulum ciocalyptoides* described by Burton (1935) from Posiet Bay, Sea of Japan. His record was later confirmed by further authors from other localities of the NW Pacific. Several characteristics indicate the invasive nature of *C. ciocalyptoides*. First of all, *C. ciocalyptoides* is new to the NE Atlantic, whereas its original distribution is strictly localized. According to the findings by Perez et al. (2006), the dispersal followed a chronological order from the first evidence in 1996 in the Ria of Etel. The sponge strongly proliferates within populated biotopes, i.e. competes successfully for space with various other marine invertebrates, such as Octocorallia and other poriferan species. Our findings confirm the hypothesis that aquaculture of the Pacific oyster *C. gigas* along the French and the Dutch coasts, may be the source of the invasion of *C. ciocalyptoides*. Taking all these facts into account, we conclude that the NE Atlantic populations of *C. ciocalyptoides* originated from the NW Pacific. To our knowledge, *C. ciocalyptoides* is the first verified "non-cosmopolitan" sponge species that has been transferred from one world ocean into another by human activity.

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Cladorhizid sponges from hydrothermal vents and cold seeps in the northernmost Atlantic and Arctic Oceans

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The deep-sea demosponge family Cladorhizidae is a group of sponges that has attracted attention due to the discovery of an atypical carnivorous feeding-mode based on the capture and subsequent digestion of small invertebrates. Additionally, methanotrophic symbionts have been described from a species associated with a mud volcano off Barbados. While normally not counted among the true vent fauna, cladorhizids are known to appear in greater numbers in the general enrichment zones around vent and seep systems. Here we present the cladorhizid sponge fauna found in a range of reducing habitats in the northernmost east Atlantic- and the Arctic Oceans. Our survey comprises the Nyegga cold seeps at 700 m depth on the Norwegian shelf, the Haakon Mosby mud volcano at 1300 m depth at the western flank of the Barents Sea, the Jan Mayen white smoker vent fields at 5-700 m depth north of Iceland, as well as the astonishing "Loki Castle" new black smoker field at 2400 m depth at the Arctic mid ocean ridge. All together nine species were encountered. Some of these were found on active chimneys, suggesting that they directly or indirectly benefit from gases in the fluids. Presence of the *pmoA* gene and isotope values suggests that some of the species have methane oxidizing symbionts. The phylogenetic relationships within the Cladorhizidae and the position of the family within the Mycalina have been debated. Based on this new material and other Atlantic as well as Pacific cladorhizids we hereby present a molecular phylogeny supported by morphological data to further elucidate the evolutionary history of this highly specialized group of sponges.

Phylogenetic analyses of poriferan families using highly conserved housekeeping gene sequences: a status report

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Here we report on an on-going, collaborative, large-scale research effort to assess the monophyly of, and resolve the branching order among, the major sponge lineages of Demospongiae, Hexactinellida, and Calcarea (AToL: PorToL – The Porifera Tree of Life Project). Despite their importance in benthic habitats worldwide, phylogenetic relationships within Porifera are not well resolved. Our team seeks to establish a robust phylogeny of sponges by integrating traditional morphological systematics with multi-tiered analyses of gene sequences. To achieve the goal of refining our understanding of higher-level sponge relationships (i.e., Class, Order, Family), we have adopted three primary strategies. The first involves exploring mitochondrial genome sequences and gene orders (to be presented by D. Lavrov elsewhere). The second involves examination of nuclear ribosomal DNA. The third strategy, presented here, involves sequencing 7 nuclear, protein encoding genes (aldolase (ALD), ATP synthase beta chain (ATPB), catalase (CAT), elongation factor 1-alpha (EF1a), methionine adenosyltransferase (MAT), phosphofructokinase (PFK), and triose-phosphate isomerase (TPI)) from the 127 families of sponges identified in Systema Porifera. Representatives of more than 30 different Poriferan families have been collected primarily from Floridia and Panama. Most of the families stem from shallow-water specimens, though several taxa were obtained from deeper water habitats by Harbor Branch Oceanographic Institute's Johnson Sea Link submersibles. Each sample has been processed for high quality mRNA for the purpose of generating cDNA libraries that will be used to obtain the protein encoding genes mentioned above. Combined with previous work in this area, we have results for nearly 40 families surveyed to date. We present phylogenies created with these data, and discuss branching orders in the context of recent hypotheses of evolutionary history within Porifera. To continue to build this phylogeny, we are eager to identify partners and collaborators who have access to fresh material from which high quality mRNA can be extracted. The collaborative and comprehensive approach that we have adopted to sequence mtDNA and nuclear genes supports efforts to understand relationships among the major clades of the sponge phylogenetic tree.

The complexity of the moons: on the phylogeny and phylogeography of Tetillidae in Indonesian waters.

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Moon sponges include the genera *Paratetilla* and *Cinachyrella*. These globular sponges resemble the lunar surface with their numerous porocalices. It has long been considered that the two species *Paratetilla bacca* (Selenka 1867) and *Cinachyrella australiensis* (Carter 1886) are widespread and abundant with a geographic range from the Red Sea to the Central Pacific in a variety of habitats. However, recent morphological work (see abstract Santodomingo et al.) has clearly shown that *Cinachyrella australiensis* represents a complex of at least six species, and *Paratetilla bacca* of at least two species. Our aim was to establish whether these species are validated by molecular markers and determine the phylogeny of the *Cinachyrella* spp. from a wide range of marine environment (anchialine lakes, coral reefs, rocky shores and coastal mangroves) from various locations within Indonesia, and over a greater spatial scale in the Indo-Pacific region. Preliminary phylogenetic analyses using COI mtDNA showed that the *Paratetilla* spp. formed a monophyletic group distinct from the *Cinachyrella* spp., which thereby confirms the distinction between genera. Preliminary results of the COI phylogenies of *Cinachyrella* spp. will be presented. In addition we found strong intra-specific variance in a number of the *Cinachyrella* spp. that may be related to variation in habitats. In this poster we will discuss the possibility of a *Cinachyrella australiensis* species complex and the role of populations isolated in anchialine lakes as stepping stones in their evolution.

Characterization of melanin from marine sponges

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Various types of melanin have already been found in the majority of organisms, being this biopolymer considered as one of the major pigments present in nature. The presence of this pigment in marine sponges (Phylum Porifera, one of the simplest multicellular organisms) was postulated, but this type of melanin was never characterized. Thus, our work consists in the extraction and characterization of a dark pigment observed in five different sponges collected in four different places. Characterization of the extracted pigment was done using solid state analytical techniques, due to the characteristic non-solubility of these pigments. Therefore characterization techniques like SEM-EDS, IR, UV-Vis, CHN analysis and X-ray were used to identify the polymer. The results showed that the extracted material was obtained in high purity, being identified as melanin. The results also emphasize the huge structure variability present in this pigment, showing different structure arrangements and compositions depending on its source, which influences the UV behaviour. The structural characterization of this class of pigments is fundamental, allowing a better understanding of melanin properties.

Chitosan of poriferan origin

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The chitin isomorphs isolated so far from arthropods and mollusks occur in the form of granules, sheets, or powders. However, fibrous skeletal frameworks of verongid sponges represent unique three-dimensional scaffolds made of alpha-chitin. Chitin is a linear polymer consisting mainly of β-(1→4)-linked 2-acetamido-2-deoxy- β-D-glucopyranose units and partially of β-(1→4)-linked 2-amino-2-deoxy- β-D-glucopyranose. In this form, chitin is insoluble in water and common organic solvents. When the degree of N-acetylation (defined as the average number of N-acetyl-D-glucosamine units per 100 monomers expressed in percent) is less than 50%, chitin becomes soluble in aqueous acidic solutions ($pH < 6.0$) and is then denoted as chitosan. That means, chitosan denotes partially (> 50 %) or fully deacetylated chitin. So far, the transformation of the recently discovered sponge chitin into chitosan has not been reported. The main goal of the present study was the development of an effective method for chitosan production from chitinous skeletal scaffolds isolated from the marine sponges *lanthella basta* and *Aplysina cauliformis*. Industrially, chitin is converted into the more readily applicable chitosan by partial or complete deacetylation under alkaline conditions or by enzymatic hydrolysis. We have used chitinous scaffolds from *I. basta* and *A. cauliformis* and NaOH solutions with different concentrations (25% - 45%) at 95°C and adjusted the degree of acetylation (DA) by the treatment time. FTIR (Golden-Gate-ATR-technology), FT-Raman and solid state NMR-spectroscopy have been used for the identification of chitosan. DA was measured by the established FTIR–method. It was shown for the first time that chitosan can be obtained from the chitinous scaffolds of sponge origin. Possible applications of chitosan/chitin based two- and three-dimensional constructs in biomedicine, bioengineering, and materials science will be discussed.

Study of the biodiversity of deep-water sponges of Lake Baikal by molecular methods

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Lake Baikal is the deepest and the most ancient lake in the world, it's age estimated at 30 million years. Due to oxygen exchange Baikal is inhabitable at all depths up to the deepest point of 1637 m. An unusually big number of sponge species live in Lake Baikal in comparison to other lakes. According to present classifications 13 species and one subspecies from the family Lubomirskiidae, and five species from the family Spongillidae, are known from the Lake Baikal. Though sponges form the dominating biomass of the benthos of Baikal and play an important role in the ecology of the lake, the taxonomy of the Baikal sponges is complicated. The existing classification of baikalian sponges was developed mainly on the basis of samples collected by scuba diving from depths up to 40 meters. A small quantity of deep-water samples collected earlier by dredging allowed the description of new species of the Lubomirskiidae but the deep-water fauna of Baikal sponges remains poorly studied. In 2008-2009, expeditions in Baikal with use of the deep-water manned vehicles Mir-1 and Mir-2 were carried out. During dives between 120 - 1450 m unique samples of deep-water sponges were collected and their habitats studied. Species identifications were performed via morphological and molecular methods and their distribution to depths over 40 meters was studied. Scanning electron microscopy (SEM) identified collected samples as *Baikalospongia intermedia*, *B. intermedia profundalis*, *B. bacllifera*, *B. fungiformis*, *B. martinsoni*, *Swartschewskia papyracea* all from the family Lubomirskiidae. According to spicule morphology some specimens are representatives of undescribed Lubomirskiidae species. Several specimens were assigned to the family Spongillidae however species identification was not possible via SEM. Analysis of ribosomal internal transcribed spacer regions (ITS1 and ITS2) was also used to perform a species identification of 15 deep-water specimens and to study their phylogenetic relationships. All deep-water specimens were shown to belong to the Lubomirskiidae including those assigned to the family Spongillidae via morphology. Furthermore, nucleotide substitutions distinguish newly collected deep-water specimens from described species of Lubomirskiidae thereby supporting the existence of new undescribed species. Study of the deep-water fauna of the Baikal sponges will improve the systematics of freshwater sponges and will provide new data for understanding the origin and evolution of the endemic family Lubomirskiidae.

Molecular taxonomy and phylogeny of freshwater sponges from Lake Kinneret: family Malawispongiidae (Porifera; Spongillina) is polyphyletic

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Freshwater sponges (Porifera; Haplosclerida; Spongillina) currently comprise six extant families: Spongillidae, Lubomirskiidae, Malawispongiidae, Metaniidae, Metschnikowiidae, and Potamolepidae. The phylogeny of this group is poorly understood. Molecular analyses of freshwater sponges have revealed that their taxonomy should be reconsidered. This, in particular, was shown for endemic genera without gemmules and microscleres inhabiting various ancient lakes (e.g. Baikal, Tanganyika, Malawi). Family Malawispongiidae Manconi & Pronzato 2002 includes five genera: *Cortispongilla* Annandale, 1918, *Malawispongia* Brien, 1972, *Spinospongilla* Brien, 1974, *Ochridaspongia* Arndt, 1937, *Pachydictyum* Weltner, 1901 found in ancient lakes: Kinneret (Middle-East), Malawi (African Rift Valley), Ohrid (Europe) and Poso (Central Sulawesi). Previous taxonomic studies on Lake Kinneret identified two to five sponge species and hypothesized their close phylogenetic affinity with other Malawispongiidae. The latter is doubtful due to large geographical distance between Lake Kinneret and other locations. The molecular systematics of endemic and cosmopolitan sponge species (families Malawispongiidae and Spongillidae, respectively) in Lake Kinneret, were analyzed using nuclear and mitochondrial markers which possess different rates of evolution. Analyses of mtCOI gene data showed that endemic Malawispongiidae of Lake Kinneret is clustered together with cosmopolitan genera *Ephydatia* and endemic *Ochridaspongia* (Lake Ohrid), but separated from *Pachydictyum globosum* (Lake Sulawesi). This supports the previously reported polyphyly of the family Malawispongiidae. Moreover, analyses of the ITS region of rDNA suggests that in Lake Kinneret four described earlier endemic species (*Cortispongilla barolsy*, *Nudospongilla aster*, *N. mappa*, *N. reversa*) can be assigned as *Ephydatia fluviatilis*. Our results indicate that probability of occurrence of endemic sponge species in ancient lakes increases with lake isolation time.

Metabolic fingerprinting as an indicator of biodiversity: towards understanding inter-specific relationships among Homoscleromorpha sponges

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Sponges are an important source of secondary metabolites showing a great diversity of structures and biological activities. Secondary metabolites can display specificity on different taxonomic levels, from species to phylum, which can make them good taxonomic characters. However, the knowledge available on the metabolome of non-model organisms is often poor. In this study, we demonstrate that sponge chemical diversity may be useful for fundamental issues in systematics or evolutionary biology, by using metabolic fingerprints as indicators of metabolomic diversity in order to assess interspecific relationships. The sponge clade Homoscleromorpha is particularly challenging because its chemistry has been little studied and its phylogeny is still debated. Identification at species level is often troublesome, especially for the highly diversified *Oscarella* genus which lacks the fundamental characters of sponge taxonomy. An HPLC-DAD-ELSD-MS metabolic fingerprinting approach was developed and applied to 10 Mediterranean Homoscleromorpha species as a rapid assessment of their chemical diversity. A first validation of our approach was to measure intraspecific variability, which was found significantly lower than interspecific variability obtained between two *Oscarella* sister-species. Interspecific relationships among Homoscleromorpha species were then inferred from the alignment of their metabolic fingerprints. The resulting classification is congruent with phylogenetic trees obtained for a mitochondrial DNA marker (COI) and demonstrates the existence of two distinct groups within Homoscleromorpha. Metabolic fingerprinting proves a useful complementary tool in sponge systematics. Our case study calls for a revision of Homoscleromorpha with further phylogenetic studies and identification of additional chemical synapomorphic characters.

A molecular model of biocalcification in the coralline sponge *Astrosclera willeyana*

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Understanding the forces that both permitted and drove the diversification of all multicellular life remains one of the great challenges to evolutionary biologists. The metazoan origins of the ability to biocalcify is significant because the appearance of this trait within diverse lineages coincides with the appearance of many disparate body plans in the fossil record. However, the degree of evolutionary conservation in the biosynthetic pathways that guide the deposition of metazoan CaCO3 skeletons is largely unknown. Here we show that *Astrosclera willeyana*, a living representative of the now largely extinct stromatoporid sponges (a polyphyletic grade of poriferan bauplan), has apparently bypassed the requirement to evolve mineral-regulating matrix proteins by using the degraded remains of bacteria to seed CaCO3 crystal growth. Furthermore we have identified a gene that was apparently transferred from a delta-proteobacterium into the genome of an ancestral demosponge via a horizontal gene transfer (HGT) event, and that this gene is now likely to be involved in the process of spherulite formation. In combination with previous work where we characterised the action of an alpha carbonic anhydrase and its role in spherulite formation, we are now developing a model of skeleton formation in this early branching metazoan. This model suggests that it was a combination of highly conserved genes inherited from the last common ancestor of the Metazoa, and lineage specific events (HGT and bacterial seeding of spherulite formation) that supported the evolution of skeleton formation in this so called 'living fossil'.

Bathydorus sp. nov. and *Docosaccus sp. nov.* (Lyssacinosida, Hexactinellida): Two new species of glass sponges from the abyssal northeast Pacific

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Two new species of glass sponges are described from Station M, a long-term study site in the abyssal plain of the northeast Pacific Ocean. Though similar in their plate-like gross morphology, analysis of spicules from these two species places them in two different families within Order Lyssacinosida. *Bathydorus* sp. nov. (Family Rossellidae) is supported by evenly-spaced lophophytous spicules that project into the soft sediments of the abyssal plain. Its perimeter is lined with long spicules, giving it a hairy appearance. The species is white to cream color and has no visible oscula on the upward-facing atrial surface. Spicules include long, blunt-tipped diactins, smooth pentactins, spiny stauractins and hexactins. Microscleres include hexasters, hexactins, and oxyhexasters. Within the genus, *Bathydorus* species does not match any of the existing eight species based on its biogeography and the spicules that comprise its skeleton. *Docosaccus* sp. nov. (Family Euplectellidae) also has long lophophytous spicules that anchor it into soft sediments, but they project in tufts from the bottom of the sponge and are not present along its perimeter. Visible oscula on the atrial surface give individuals a spotted appearance. Spicule analysis places the sponge into Family Euplectellidae and its lophophytous method of attachment places it in Subfamily Euplectellinae. Megascleres include hexactins, pentactins, and diactins; microscleres include oxyhexasters, hemioxyhexasters, and floricomes. The presence of extremely large hexactins eliminates most genera within Subfamily Euplectellinae, leaving the monospecific genus *Docosaccus* Topsent 1910. Descriptions of these new species are necessary as studies will soon emerge establishing sequences of their mitochondrial genomes and studying population-level changes using time-series data from Station M.

Bacterial communities associated with the freshwater sponges of Lake Baikal and identification of polyketide synthase genes from the sponge metagenome and cultivated strains

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Phylum Porifera consists of approximately 15,000 species inhabiting marine and freshwater environments. Although most sponges are found in the marine environment, an estimated 150 species populate freshwater habitats. Since sponges are filter feeders they make associations with great amount of microorganisms. Molecular ecological studies have suggested that a majority of sponge-associated microbes are uncultured. Investigation of cultured microorganisms derived from sponge communities is also important because they can produce a novel bioactive metabolites. In recent years, a numerous studies have concerned to estimation of microbial diversity and to detection of the ability of sponge-associated bacteria to produce different bioactive compounds, large number of which are polyketides. In contrast to marine sponges, freshwater-sponge microbial communities are poorly studied. In present work for the first time the uncultivated bacterial diversity associated with the endemic baikalian sponge *Lubomirskia baicalensis* was described employing 16S rRNA gene library construction, restriction fragment length polymorphism and phylogenetic analysis. It was found *that L. baicalensis*-associated bacterial community consist of Actinobacteria, alpha-, beta-Proteobacteria, Verrucomicrobia, Bacteroidetes and Cyanobacteria where Actinobacteria was predominant group (about 36% of all phylotipes obtained). The potential of sponge microbiota to produce bioactive metabolites was also analyzed by PCR-screening of PKS genes with degenerate primers DK-F and DK-R (Ehrenreich et al., 2005) using metagenome DNA as template. Amplicones obtained from the clone library were sequenced resulted in 18 different sequences (with 35-99 % identity). A BLASTX analysis revealed that all sequences were related to KS-domains of PKS genes isolated from alpha-, beta-, delta-Proteobacteria (Myxococcales), Verrucomicrobia, Cyanobacteria, uncultured bacteria (including symbionts of sponge *Discodermia dissoluta*) and eucariotic Chlorophyta. All PKS gene fragments showed varying similarities (47-82% identity) to their closest matches in GenBank. Several KS-domains obtained in present study were related to genes involved in byosynthesis of known metabolites: curacin A, (CurI; antitubulin natural product), melithiazol (MelF; electron transport inhibitor), stigmatellin (StiB, StiC; electron transport inhibitor), cryptophycin (CrpB; anticancer agent), myxalamid (antibiotic). Cultivation of bacteria associated with baikalian sponges (*L. baicalensis* and *Baikalospongia bacillifera*) resulted in isolation of 85 strains, 30 of which were identified by microbiological and molecular (16S rRNA) methods. Most of the strains analyzed were belong to phyla alpha-, gamma-Proteobacteria, Firmicutes and Actinobacteria. Strain *Pseudomonas fluorescens* 28Bb-06 was chosen for molecular screening to determine the presence of PKS genes. Four obtained KS-domain sequences shared 40-53% identity to each other at amino-acid level and 51-66% identity with PKS genes of different bacteria including producers of yersiniabactin (Ybt; siderophore, virulence factor), rhizoxin (RhiA, antimitotic agent with anti-tumor activity), epothilone (EpoE; cytotoxic agent) and disorazol (DszA; inhibitor of tubulin polymerization). Our results revealed the high diversity of Lake Baikal sponge microbial community and potential ability of microorganisms associated with baikalian sponge to produce the novel bioactive compounds.

A set of ribosomal RNA gene copies in a single individual of *Ephydatia fluviatilis*

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A number of internal transcribed spacer (ITS) sequences for the freshwater sponge *Ephydatia fluviatilis* from various localities all over the world can be found in databases. However, only one distinct ITS sequence has been published per a sample of *E. fluviatilis*, though typically up to several hundreds of copies of ribosomal RNA genes are found per eukaryotic genome. The tandem repeats of the rRNA genes are considered to be subjected to a concerted evolution. Still, the rRNA genes outside the tandem repeats (e.g. separated by more than 1Mb or even located on different chromosomes) evolve independently which may lead to a certain heterogeneity of the rRNA gene pool. For *E. fluviatilis* it has also been shown that ribosomal genes are located on at least two different chromosomes (Ishijima et al 2008, Zoolog Sci 25(5):480-6). Thus very likely detection of rRNA gene pool heterogeneity for a single individual is possible. The pool of rRNA genes instead of a single sequence could be considered to be characteristic of a particular individual. We used the amplification of ITS region from *E. fluviatilis* with primers specific for 18S and 28S rDNA conserved positions in freshwater sponges. The obtained amplicon was a heterogenous product with several single nucleotide polymorphisms (including nucleotide substitutions and deletions/insertions). This heterogeneity was resolved by cloning the product and sequencing the obtained clones. We conclude that the direct sequencing of the ITS region together with a semiquantitave analysis of the rRNA gene pool is sufficient for the molecular characterization of a sponge individual. Our approach would help to find out the relationships between individuality, coloniality and definition of strains of *E. fluviatilis*.

Geographic variation of microbial diversity of symbionts in the coralline sponge *Astrosclera willeyana* from the Red Sea (Egypt) and the Great Barrier Reef (Australia)

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Distantly related sponges from different oceans host similar microbial communities. However, despite of intensive research on symbiotic microorganisms in sponges, our knowledge is still not comprehensive. The microbial diversity in coralline sponges, unique members of Phylum Porifera, was never investigated using culture-independent molecular methods. The coralline demosponge *Astrosclera willeyana*, thought to be a living representative of reef-building stromatoporoids of the Mesozoic and the Palaeozoic, is widely distributed throughout the Indo-Pacific, from the Red Sea to the Society Islands. The aim of the study was to examine the variability in microbial community composition of *Astrosclera* from separated geographical regions. The phylogenetic affiliation of sponge-associated bacteria was assessed by random sequencing of two 16S rDNA clone libraries constructed from single specimens of *Astrosclera willeyana* from the northern Red Sea (Egypt) and the Great Barrier Reef (GBR) (Australia). A single archaeal 16S rDNA genotype, affiliated to the phylum Crenarcheota, was found in the specimen from the GBR, but none in the one from the Red Sea yet. In total 322 and 347 clones were sequenced from the GBR and the Red Sea specimens, respectively. Phylogenetic analysis revealed complex microbial communities in both samples, sharing significant features. The phylogenetic distribution of sequences in both sponges was relatively uniform, characteristic for microbial communities with high diversity. *A. willeyana* from the Great Barrier Reef showed slightly higher diversity, where Poribacteria and Archaea were additionally found (each 1%). In both samples, the green non-sulfur bacteria – Thermomicrobia/Chloroflexi (20% Egypt and GBR) were the most abundant, as well as the Gammaproteobacteria (17% Egypt, 14% GBR), and the Actinobacteria (10% Egypt, 14% GBR). The Deltaproteobacteria and Nitrospirae were more abundant in the Red Sea specimen (12%, 8% Egypt and 10%, 6% GBR, respectively). The Acidobacteria and Alphaproteobacteria (about 9% each), as well as the Gemmatimonadetes, Nitrospina, and Spirochaetes (in total 8%) were found in both samples with similar abundance. Over 90% of sequences obtained from *Astrosclera willeyana* from both geographically separated areas were closely related to other sponge-derived sequences, highlighting the uniqueness of the microbial consortia in sponges. The remaining sequences were similar to sequences from uncultivated organisms, particularly from marine environments. None of the sequences were closely related to validly described microorganisms. In both *Astrosclera* samples 8% of the sequences was affiliated to the group of "uncultured bacteria", which enclosed unidentified bacteria diverse from all hitherto characterised and described microorganisms.

Phylogenetic diversity of the microbial symbionts in the coralline sponge *Vaceletia crypta*

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Recent advances in culture-independent molecular techniques have greatly improved our knowledge of microbial diversity in sponges, which harbour rich and various microbial communities in their tissues. However, a clear picture remains still afar. This is especially true for coralline sponges, also called sclerosponges, which are unique members of Phylum Porifera because they build a solid ('hypercalcified') calcareous skeleton, similar in appearance to some reef building corals. During long periods of the Earth's history coralline sponges were dominant reef-building organisms and are regarded as 'relict taxa' or 'living fossils'. *Vaceletia* is the only recent member of so-called 'sphinctozoan-type' sponges, which contributed to reef-building in the Palaeozoic and Mesozoic. Thought to be extinct, *Vaceletia crypta* was rediscovered in the 1970's. Here we present results about the microbial diversity of *Vaceletia crypta* from the Great Barrier Reef (Australia), obtained by sequencing of a bacterial 16S rDNA clone library. 396 clones were sequenced and used for subsequent phylogenetic analysis. Archaeal 16S rRNA was PCR-amplified, but not yet successfully sequenced. The microbial community of *V. crypta* showed a high microbial diversity and a complex composition with a relatively homogeneous phylogenetic distribution. The most abundant were members of Thermomicrobia/Chloroflexi (18%). Over 50% of the microbial community consisted of the Gammaproteobacteria, Gemmatimonadetes, Actinobacteria, Nitrospira, Acidobacteria with nearly equal participation. Less abundant members of the microbial community belonged to the Deltaproteobacteria (6%), Nitrospina (6%), as well as the Alphaproteobacteria, Poribacteria, Betaproteobacteria, Cyanobacteria, Spirochaetes, and Deinococcus-Thermus (in total 10%). 9% of the sequences from *V. crypta* were assigned to the group of "uncultured bacteria" of undefined affiliations, which indicate new unspecified microorganisms with unknown physiological potential. 95% of the sequences clustered together with other sponge-derived sequences, what underscores that the 'living fossil' coralline sponge *Vaceletia* shares features of its microbial community with other sponges.

Genotypic and phenotypic characterization of fluorescent *Pseudomonas* spp. isolated from the freshwater sponge *Ephydatia fluviatilis*

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Bacteria are believed to play an important role in the complex biochemistry of sponges (Porifera). Cosmopolitan and metabolically-versatile *Pseudomonas* species are known to produce a variety of secondary metabolites with putative biotechnological applications. These organisms also display an extraordinary capability to colonize a broad range of eukaryotic hosts, but knowledge of their occurrence and activity in sponges is still rudimentary. In this study, *Pseudomonas* ssp. isolated from the freshwater sponge *Ephydatia fluviatilis* were analysed as to their diversity and antagonistic capabilities against a range of model environmental organisms. Culturable fluorescent pseudomonads were numerically enriched in *E. fluviatilis*, surpassing counts obtained for lake water samples by 3-4 orders of magnitude. The genotypic diversity of 90 fluorescent isolates retrieved from *E. fluviatilis* was determined by BOX-PCR fingerprinting and shown to substantially vary among different sponge individuals. Sixty-two *Pseudomonas* isolates were tentatively identified by 16S rRNA gene sequencing to species level with ≥ 97.8% probability, whereby 7 different *Pseudomonas* species were found: *P. mandelii*, *P. migulae*, *P. jessenii*, *P. corrugata*, *P. gessardii*, *P. putida* and *P. fluorescens*. Forty-nine percent of the isolates displayed *in vitro* antagonistic activity towards the Gram-positive bacterium *Bacillus subtilis*; 34% towards the oomycete *Phytium ultimum*; and 19% towards pathogenic fungi. Moreover, 35% of the isolates showed remarkable resistance against predation exerted by the ciliate *Colpoda steinii* in microcosms. Selected cell extracts of resistant pseudomonads caused either complete immobility or lysis of active *C. steinii* cells *in vitro*. Multivariate analysis revealed a clear allocation of the antagonistic *Pseudomonas* isolates in two groups. One group displayed pronounced activity against the tested fungi and the oomycete *P. ultimum*, whereas the second group was characterized by its activity against *B. subtilis* and resistance towards *C. steinii*. Both antagonism and predation resistance were positively correlated with the ability of the strains to produce biofilms in static microcosms. Among the isolates displaying the most pronounced antagonistic features were those tentatively identified as *Pseudomonas fluorescens*, *P. jessenii* and *P. mandelii*. Our results indicate that the freshwater sponge *Ephydatia fluviatilis* harbours diverse and bioactive *Pseudomonas* spp. of potential use in applied biotechnology.

Spermatozoids or asexual propagules in Calcarea?

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During one year we have accompanied a population of the sponge *Clathrina aurea* (Porifera, Calcarea, Calcinea) in Cabo Frio, Rio de Janeiro state, Brazil, to study its reproduction. A total of 10 specimens of *C. aurea* were monthly collected during one year, and fixed for standard histology (Bouin fixative) and SEM analyses. For the SEM, specimens were fixed in 2.5% glutaraldehyde, 0.2 M sodium cacodylate buffer (pH 7.0) in filtered seawater (1:4:5), decalcified (5% EDTA pH 7.0 for 48 - 72 hours), dehydrated, criofractured, coated with gold, and observed in a JEOL JSN 5310 SEM. Oocytes, embryos or larvae were not found after one year of study, however, we observed in some specimens flagellate cells budding from choanocytes. While several specimens showed a normal choanoderm, with choanocytes presenting collar and flagellum, others presented a disorganised choanoderm with flagellated cells, without a collar, budding from the region inside the choanocytes' collar. In some cases, in the same specimen, we could observe different stages of budding, including free flagellated cells. While the choanocytes measure 3.4 μm (± 0.2 μm) in diameter, those cells are rounded and measure 2.1 μm (± 0.2 μm). Additionally, they do not have a collar but present a flagellum with the same thickness as the choanocyte's flagellum (0.3 μm). The existence of choanocytes without collar in calcareous sponges has already been reported by a few authors in different species. However, this change has usually been attributed to factors other than reproduction, for example, as a result of anoxia. Gatenby was the only author that suggested that choanocytes without collar could be related to reproduction. Although the origin of the missing collar may be related to factors other than reproduction in other calcarean species, in *C. aurea* we believe that these flagellate cells without collar are spermatozoids or asexual propagules. The budding of cells from choanocytes, the presence of different budding stages and the disorganization of the entire choanoderm support our hypothesis that the free flagellate cells observed in the lumen of *C. aurea* are probably spermatozoids. On the other hand, the absence of oocytes and larvae after one year of investigation suggests that these flagellate cells might be asexual propagules. If that is the case, then, asexual reproduction would be the most important strategy in maintaining populations of *C. aurea* in Cabo Frio. Further studies will have to be performed until we can determine the function of these differentiated cells.

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Temporal variability in sponge assemblages inhabiting an altered lagoon system in a remote Pacific atoll

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Remote atolls are some of the last strongholds for near-pristine reef systems. Knowing the potential threats to these reefs is critical to their future conservation. Palmyra Atoll is one of the most remote reef systems in the world, located 1,930 km south of Hawaii. The atoll remained essentially uninhabited until the U.S. military developed the lagoon system and islets during WWII. The alterations included extensive lagoon dredging, land reclamation and the creation of a shipping channel into the lagoon. The reefs represent a largely pristine oceanic environment with few sponge species (excluding bioeroding spp.), yet the dominant fauna in the degraded lagoons are sponges, including at least two introduced non-indigenous species. We aimed to examine the temporal stability of sponge assemblages in the lagoon to determine whether they were stable or increasing/changing, thereby representing a potential future threat to the adjacent reefs. In July and October 2008 and 2009 we recorded sponge species density and area coverage relative to available boulder surface area *in situ* (upper surfaces examined only) using 15 x $1m^2$ permanent quadrats at 3 sites. We found that boulders moved between seasons, but despite this, sponge area cover remained relatively stable, whereas species densities fluctuated significantly between seasons. Although we observed seasonal changes in the nature of sponge assemblages, they were relatively stable between years. We suggest the seasonal variation in sponge densities may be due to seasonal fragmentation of the sponges or occurs as a result of boulder turnover. Our results indicate that the lagoon sponges at Palmyra are relatively stable despite exhibiting seasonal fluctuations in species densities. Our study highlights the important of examining temporal changes in sponges at both seasonal and inter-annual scales.

Environmental variability drives differences in sponge assemblages at a remote Pacific atoll

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Habitat modifications will inevitably alter the way an ecosystem functions. Palmyra Atoll National Wildlife Refuge, located 1,930 km South of Hawaii, was occupied by the US military during WWII. During this time, Palmyra was heavily altered through dredging, land reclamation, extensive building and the creation of a shipping channel through the reef. The sponges at Palmyra (excluding bioeroding spp.) are scarce on the near-pristine reef but are the dominant fauna within the atoll's lagoons. In 2008 we conducted a baseline survey at 12 sites across the 4 lagoons at Palmyra to assess how environmental variability influenced sponge species diversity, density, percentage cover and assemblage structure. We also measured a suite of environmental variables at each site including temperature, chlorophyll-a, water flow, total particulate matter and percentage organic matter of settled sediment. In this presentation, we will discuss the association between sponge assemblage patterns and environmental variables and identify indicator species characterizing the different environmental regimes. Our data suggests that sedimentation and flow rates are key proximal factors structuring sponge assemblage patterns. Of the species found at Palmyra, two are of particular interest as they have been identified as non-indigenous (NIS) with the possibility of other NIS potentially originating from Hawaii. The functional significance of the lagoon sponges at Palmyra remains unknown. However, the lagoon sponges have the potential to directly influence other species through spatial competition or indirectly through filtering the lagoon water, which subsequently flows over the adjacent reef. Our surveys demonstrate that most of the sponges at Palmyra are restricted to the lagoons, including the NIS and are therefore unlikely to spread to the reefs unless there is a decline in environmental quality.

Isolation and identification of chitin within fibrous skeletons of Verongida sponges.

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Because sponges, with a fossil record dating back to the Precambrian, are probably the earliest branching animals, the description of chitin forms in this group is of evolutionary and phylogenetic interest. The goal of the present study was to carry out systematic screening for the presence of chitin-based scaffolds within different representatives of the order Verongida. Verongida sponges are characterized by rigid fibers which are strongly and regularly anastomosed thus forming skeletal networks. Fragments of the sponges were treated in distilled water, acetic acid, sodium hydroxide and optionally hydrogen peroxide. The residual (skeletal) fraction was dialyzed, air dried and analyzed by various methods (SEM, 13C solid-state NMR spectroscopy, NEXAFS spectroscopy, IR and Raman spectroscopy, fluorescence microscopy) as well as biochemical approaches (chitinase digestion and test). The present systematic study of representatives from the Verongida order revealed the presence of a chitin-based scaffold closely resembling the shape and morphology of the original sponge for all species under study. The particular chitin biosynthesis pathways in verongid sponges remain a target for future research. The issues relative to the potential applicability of three-dimensional chitin-based scaffolds in tissue engineering as well as for technical application are discussed.

Structural features of *Aspidoscopulia* sp. (Hexactinellida) skeleton with respect to isolation of organic matrix

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Aspidoscopules have discoidal head and spines that protrude from a single marginal whorl of the head. Microscleres usually hexasterous with oxyoidal, discoidal and onychoidal outer ends – to be eliminated at all. The genus *Aspidoscupulia* in most characters is similar to the genus *Farrea*, both are representatives of Farreidae. Earlier, we reported the presence of chitin within skeletal frameworks of *Farrea occa*. However, there is no information about the nature and origin of organic matrix in *Aspidoscopulia* species. Organic matrix differs is close genera of Amphidiscophora: *Monorhaphis* (Monorhaphididae) and Sericolophus (Pheronematidae), close genera and families, have different organic matrix in their silica spicules: collagen and chitin correspondingly. Here, we present a detailed study of structural and physico-chemical properties of skeletal fragment of *Aspidoscopulia* sp. We applied both HF- and alkali-based approaches for demineralization of glass sponge skeletal fragments. By investigating of these materials using a wide array of techniques (FTIR, SEM, TEM and electron diffraction, Calcofluor White Staining and fluorescence microscopy), we show that chitin is a component of the siliceous skeleton of *Aspidoscopulia* sp. similar to *Farrea occa*.

Structural diversity of silica-based formations in freshwater sponge *Drulia brownii* (Metaniidae: Demospongiae)

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There are three strategies within Porifera to strengthen their skeletons: biomineralization of organic templates, use of the foreign material (mineral microparticles, spicule debris) and use of self producing spicular material. It is well established that when the sponge spicules are formed, the largest of them are often aggregated in bundles embedded in a fibrous sheath (e.g. spiculated spongin fibres). This kind of strategy has been observed by us also in poorly investigated freshwater Amazonian sponge *Drulia brownii*. This sponge can easily be seen and collected during the low water season, dried out, encrusting the trunks, branches, twigs and leaves of the flooded forest. Besides the megascleres, the gemmules of this species are covered by small siliceous shield-shaped (scuteliform) gemmoscleres. The present study deals with structural diversity of silica-based formations, which has been observed in *D. brownii* using stereo-, light- and fluorescence microscopy as well as using SEM and TEM. Detailed physico-chemical and biochemical investigations of organic material that glues spicules of *D. brownii* showed no presence of chitin. In this communication will be discussed about the possible nature and origin of the pigmented organic glue.

Comparative investigations of nature and origin of skeletal organic matrices of *Aplysina aerophoba* (Verongida) and *Dysidea avara* (Dendroceratida)

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According to the literature, in the orders Verongida, Dictyoceratida, and Dendroceratida, jointly referred to as 'keratose demosponges', the skeleton does not contain siliceous spicules but only spongin fibres. This shared trait has encouraged placement of these orders together within Demospongiae, although their relationships remain uncertain. Here, we will report about comparative study with respect to identification of organic matrix within skeletal fibres of Dendroceratida sponge *Dysidea avara* and of Verongida sponge *Aplysina aerophoba*, which has been collected from the same place of the sea bottom and at the same time in Kotor Bay in Montenegro. Different structural (SEM, TEM, LSM, FTIR, Raman) and biochemical analyses of the chemically treated and untreated skeletal fibres of both sponge species were performed. It was shown that chitin and silica-chitin-aragonite biocomposite are present only in *A. aerophoba*, however *D. avara* possesses only collagen-like (spongin) organic matrix with incorporated foreign microparticles (quartz, basalt) and spicular debris. Our results confirm previously reported data based on 18S, and the C1, D1, and C2 domains of 28S rRNA, which suggested that the verongids to be not related to the 'keratose' orders Dictyoceratida and Dendroceratida. Because, chitin system has been recently suggested as more ancient as collagen system, we can speculate that Verongida are older than Dendroceratida. Here, we propose for the first time that the key to the orders as one of the criteria that differentiate Verongida from Dictyoceratida and Dendroceratida could be the possibility of having chitin.

Sponge 2-5A synthetases form the distinct subgroup of the 2-5A synthetase family

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2',5'-oligoadenylate synthetases (2-5A synthetases; OAS) catalyze the polymerization of ATP into 2',5'-linked oligoadenylates (2-5A). The only known function of 2-5A in higher vertebrates is to activate latent ribonuclease (RNase L) that degrades single-stranded RNA of viral and cellular origin. Being important components of the antiviral action of interferon, 2-5A synthetases have been suggested to also be involved in other cellular processes, such as regulation of cell growth, differentiation, pre-mRNA splicing, tumorigenesis and apoptosis. We have demonstrated the presence of OAS in the evolutionarily most basal multicellular animals, sponges, where no interferon system is present. Our studies have revealed significant differences in the enzymatic properties of the enzymes from sponges and vertebrates. Differently from higher animals, the OAS activity in sponges does not need external dsRNA and the enzymatic activity cannot be destroyed by RNase treatment. However, when expressed in *E. coli*, the recombinant OAS from *Geodia cydonium* binds to bacterial RNA with high affinity. The homology of sponge OAS proteins compared to those from vertebrates is rather weak. Moreover, the OAS gene structures (i.e. exon/intron patterns) of the marine sponges *G. cydonium* and *Amphimedon queenslandica* are completely different from the conserved OAS gene structures of vertebrates. Sponge OAS genes do not form a single contiguous cluster but are grouped in separate loci containing one or two tandemly arranged genes. Thus, sponge OASs form a distinct subgroup of 2-5A synthetases. Some evidence exists, that sponge OASs are also involved in innate immune system of these animals. In contrast to mammals, the exact signalling pathways leading to the corresponding response (stress response, including probably antiviral response) are still unknown in sponges. Further studies of these ancient enzymes might be helpful for unravelling mechanisms of the other, less well known signalling pathways involving mammalian OASs.

Reproductive dynamics of *Paraleucilla magna* (Porifera, Calcarea) and its relationship with some environmental parameters in Rio de Janeiro (Brazil)

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The reproduction of several groups of marine invertebrates is usually associated to changes in environmental parameters. However, the influence of such parameters is still obscure to several groups of sponges (phylum Porifera). In this work, we try to elucidate how changes in the environment affect the reproductive dynamics of *Paraleucilla magna* (Calcaronea, Amphoriscidae), an alien species in Rio de Janeiro (Brazil). Ten specimens of *P. magna* were sampled monthly from September 2006 to March 2008 and fixed (Bouin) to histological analysis. Densities of reproductive elements (ripe oocytes, embryos, and larvae) were calculated counting them in a square with 4 mm^2 area on the sections. The environmental parameters analyzed were: photoperiod, rainfall, day and night tide height, superficial seawater temperature, salinity, primary productivity (chlorophyll a and pheophytin), and bacterioplankton abundance. Although *Paraleucilla magna* was reproductive throughout the whole studied period, the quantification of the reproductive elements allowed us to observe two major periods of a stronger reproductive effort: January to June 2007 with peak in April [16.9 (±12.1) reproductive elements/mm^2], and November 2007 to March 2008 with peak in January [12.6 (±6.6) reproductive elements/mm^2]. This result differs from a previous work with this species, when *P. magna* was reproductive only during the summer. It was not found any seasonal pattern for almost all the environmental parameters studied, except for the daily tide height (higher mean values were observed in the summer) and the night tide height (higher mean values occurred in the winter). Ripe oocytes were found mainly during the summer, although their maximum peak was in April 2007 [8.0 (±9.3) oocytes/mm^2]. The variation of the densities of oocytes during the studied period was significant ($p<0.0001$, Kruskal-Wallis test [KW]), and was not correlated with any environmental parameter. Embryogenesis (all the stages from the 1st cleavage to stomoblastulae) occurred only after January 2007, with peaks in the summer. Their densities significantly varied among the months ($p<0.0001$, KW) and could be correlated with the densities of oocytes ($p<0.0001$, Pearson's correlation [PC]) and with the water temperature ($p=0.401$, PC). Larvae were observed in almost all the studied period with peaks during the summer and April 2007. Larvae densities were also significantly different among the studied months ($p<0.0001$, KW) and this variation was correlated with pheophytin ($p=0.014$, PC). The reproductive effort (mean value of the sum of the means of the three reproductive elements of each month) was significantly different among the months ($p<0.0001$, KW) and was correlated with pheophytin ($p=0.04$, PC) and bacterioplankton ($p=0.025$, PC). Although oocytes, larvae, and the "reproductive effort" were not significantly correlated with the temperature, plotting the reproductive elements vs. the temperature indicated a possible role of this parameter in the reproduction of *P. magna*.

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Life cycle and the reproductive dynamics of *Sycettusa hastifera* (Calcarea: Calcaronea: Heteropiidae) in the Southwestern Atlantic

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In this work, we describe some aspects of the life cycle and reproductive dynamics of *Sycettusa hastifera* Row, 1909 (Calcaronea, Heteropiidae), a tubular syconoid sponge with osculum-basis polarity and radial symmetry. From September 2008 to November 2009, ten specimens of *S. hastifera* were monthly collected in Arraial do Cabo (Rio de Janeiro), Brazil (22°58'S and 42°00'W). Specimens were fixed in Bouin for standard histological techniques. Densities of reproductive elements were calculated after digital imaging of tissue's sections of each individual. We found that the population of *Sycettusa hastifera* was reproductive in Arraial do Cabo throughout the studied period, with at least four of ten specimens showing some stage of reproduction in each month, except for July 2009 when none specimen was reproductive. To form the oogonia, choanocytes divide transversally, one of the daughter cells is still in the choanoderm and the other is modified to form the oogonium. The first division gives rise to the oocyte I and a nurse cell. After the second division, the oocyte starts its growth period and is aided by two or three nurse cells, which are never phagocyted. The mature oocyte measures ca. 50 µm, is rounded, with a large nucleolated nucleus. Oocytes were observed in the choanosome in all studied months and reached a maximum density of 19.7 oocytes/mm^2 (December 2008). Spermatic cysts (sc) arise from functional choanocyte chambers and the spermatozoids seem to be derived from choanocytes. Ripe spermatozoids are small triangular and flagellated cells. Spermatic cysts were also observed in all studied months and reached a maximum of 12.0 sc/mm^2 in November 2008. Embryogenesis, from the 1st cleavage to the stomoblastula, was similar to what was observed in other calcaronean sponges. Embryos were absent in two months (November 2008 and April 2009), and reached a maximum density of 9.2 embryos/mm^2 in December 2008. To form the amphiblastula larvae, the embryos invert directly into the lumen of the choanocyte chambers and are, probably, released soon after their inversion. This is a possible explanation to the fact that larvae were the less constant reproductive elements of the sponge (larvae were not observed in several months of the studied period). However, when present in the choanosome, larvae reached a high density (maximum of 16.5 larvae/mm^2 in January 2009), one of the largest densities ever documented in the phylum Porifera. The reproductive effort of *Sycettusa hastifera* was significantly different among the months of the studied period ($p< 0.0001$, Kruskall-Wallis test). The reproductive effort was positively correlated to the wet weight of the specimens ($p<0.001$, $r= 0.467$, Spearman's correlation), but it could not be correlated with the superficial seawater temperature ($p= 0.796$, Pearson's correlation).

Ultrastructure of the amphiblastula larva and its "placental membrane" in *Paraleucilla magna* (Calcarea, Calcaronea)

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The process of inversion/excurvation observed in all studied Calcaronea during their embryological development is a unique event in all the Metazoa, and a synapomorphy of the species of this subclass. In this process, the stomoblastula turns inside out, giving the amphiblastula larva. Despite the importance of this phenomenon, very few studies have focused in the ultrastructure of this larva and its accessory membrane, the so-called "placental membrane". In this study, we describe the ultrastructure of the amphiblastula larvae in the choanosome of the sponge *Paraleucilla magna*. From September 2006 to March 2008, sponges were monthly collected and fixed in a solution of glutaraldehyde 2.5%, sodium cacodylate (0.2M, pH 7.0) and filtered seawater. Specimens were post fixed in osmium tetroxide, embedded in Epon resine and cut in an Ultramicrotome (60-90 nm). The immature larva, soon after its inversion, has three different cell types (micromeres, macromeres, and cross cells) and some maternally derived amoebocytes in its cavity. After consuming these amoebocytes, the amphiblastula starts the formation of the placental membrane, which seems to be derived from the macromeres. After several cell divisions the membrane surrounds the entire larva, which becomes mature. The placental membrane is composed of flattened cells, with electrondense inclusions, large vacuoles and parallel junctions. It seems to protect and nourish the amphiblastula, since its cells are in close contact and they are packed with different inclusions that seem to be transferred to the larva. The mature amphiblastula has now a fourth cell type, the mesoblast, which seems to be derived from the macromeres and can be found inside the cavity of the larva. Mesoblasts have few electrondense cytoplasm filled with rounded homogeneous inclusions and are consumed during larval maturation. The macromeres of the mature larva are globular and large. They have a central, rounded, large, and nucleolated nucleus, and their cytoplasm is filled with different types of inclusions. They seem to be responsible for the nutritional reserve of the larva. Different macromeres are kept together by parallel junctions. The micromeres are the most common cells in the mature larvae. These cells are smaller than the macromeres and are flagellated. Micromeres can be considered to be polarized since they have an apical nucleus and a basis filled with inclusions. In the apical region of the cell it is also possible to observe the cilium basal body closely related to the nucleus. At this region, micromeres are also connected by parallel junctions. In *P. magna* there are four cross cells in the larval equator. Cross cells are elongated as the micromeres, but lack a cilium and their structures are completely different. The nucleus of the cross cells is irregular and localized close to the centre of the cell. The cytoplasm has different inclusions and several glycogen rosettes.

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The γ-aminobutyric acid (GABA) modulates feeding activity in sponges

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In this study we report the presence of the γ-aminobutyric acid (GABA)-ergic system in the calcisponges *Leucandra aspera* and *Clathrina clathrus* and examine the topographic localization of the components of this system, including GABA-like receptors, using immunofluorescence and confocal microscopy. We found that both GABAB R1 and R2 subunits were present in the choanocytes of both sponges as well as in the eso- and endopinacocytes of *L. aspera*. The aptitude to synthesize GABA from glutamate and to transport it into vesicles was supported by the presence of glutamate decarboxylase (GAD) and of the vesicular GABA transporter (vGAT), respectively. The functional role of GABA in the feeding behaviour of these sponges was tested. The involvement of GABA receptors in the uptake processes was demonstrated in *L. aspera* using dextran conjugated to Texas Red and in *C. clathrus* using fluorescent latex particles, and by treating isolated sponge cells with a GABAB receptor agonist and antagonist. The amount of dextran and latex particles that was ingested by the sponges increased in dissociated cells when the GABAB receptor agonist baclofen was used, and this stimulatory effect was prevented by treatment with the GABAB receptor antagonist phaclofen. Additionally, stimulatory effects of baclofen could be blocked by treatment with pertussis toxin, indicating a role for G proteins in food uptake in *L. aspera* and *C. clathrus*. These results suggest that the modulation of feeding behaviour by GABA receptors is an ancient characteristic of the animal kingdom and that the pharmacological properties of these receptors have been highly conserved through the evolution.

Filtering activity of two demosponges in experimental sponge culture

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As far integrated sponge culture has been proposed for bioremediation in coastal areas we report about farming experiments on the demosponges *Ircinia variabilis* and *Agelas oroides* performed in a harbour within the Ligurian Sea (Rapallo Harbour). The farming site is characterized by high level of pollution mainly due to marine activities, wastewater and urban runoff. The sponge explants were cultured following USAMA® methodologies (nylon ropes) and *in situ* clearance rate tests were performed on cultured *I. variabilis* and *A. oroides* to detect their filtering ability in stressing conditions. Water analyses were conducted on particulate organic matter by gravimetric method and on bacterial density by epifluorescence microscopy counting. The same farming and filtering experiments were performed in two other sites located within the Portofino Marine Protected Area (Ligurian Sea) and the Capo Caccia – Isola Piana MPA (Sardinian Sea) to compare culture performances and clearance rates among polluted/unpolluted sites. Farming trials revealed high growth rates for both studied species during six months of culture from winter to summer. At the polluted site a rapid increase of sponge explants mortality occurred, as a consequence of extreme variations of environmental conditions (salinity, sedimentation, temperature) at the beginning of summer. Despite the death of the explants, survival and growth rates within the harbour until spring ending were similar to the values observed in the two unpolluted sites. Filtering experiments in spring revealed high retention and clearance rates of *I. variabilis* and *A. oroides* in all the sites with the higher rates from the polluted site further reflecting the optimal metabolic performances of explants in the first phase of culture in stressing conditions. Our results highlight the ability of *I. variabilis* and *A. oroides* in clearing significant volume of water to deploy organic and bacterial loads also in a polluted site.

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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

A new extraordinary species of *Chondrocladia* from off northern California, USA

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Two specimens of a large, fan-shaped species of *Chondrocladia* were collected from ca 3300 m in the Escanaba Trough, off N. California by ROV 'Tiburon' in 2000 and 2005. Review of the designated holotype shows it anchored in sediment by central basal rhizoids arising from a thickened central area supporting 2 tapering horizontal stolons; these extend laterally, at an angle of 110 degrees for 26-37 cm on either side of the sponge; a 3rd horizontal stolon branches from one main stolon at an angle of 90 degrees. Each stolon supports a series of respectively 17-32 evenly-spaced vertical branches analogous to a comb; the tallest branches (26 cm) are central and height diminishes distally to mere stubs at the stolon growth tips. Four rows of thin filaments (1.5-2.5 mm long, spaced at 1-1.5mm intervals) line the edges of the branches. The filaments are supported by parallel bundles of subtylostyles while rhizoids, stolons and branches are supported by parallel bundles of styles. All surfaces (rhizoids, stolons, branches, filaments) are covered by a thick mat of tridentate isochelae forming a substantial Velcro-like ectosome. Vertical branches have irregularly occurring small discrete swellings along their length and larger balloon-like spherical swellings on their upper tip; the latter, shrunk about 50% in preserved specimens. Specialized functions are highly regionalized. What are likely choanocyte chambers occur only in horizontal stolons and branch swellings. Spermiogenesis occurs only in terminal balls. Long cylindrical filamentous sperm, probably lacking flagella, are packed in spermatocysts, 10-20 of which are grouped into spermatocyst packets bristling with small sigmas. The packets appear to be released from terminal balls and caught on filaments by Velcro-like interaction between sigmas and chelas. Prey capture of small crustaceans occurs on all other surfaces but has not been observed on filaments. The designated paratype has approximately the same form and structure as the designated holotype. The form of the two specimens was compared with the 34 *Chondrocladia* species listed in the World Porifera Database. Only 3 species have a form even remotely like that of the two NE Pacific specimens. Only one of these 3 species, *Chondrocladia nicolae* has isochelae which are tridentate. *C. nicolae* was described from a fragment which could be interpreted as comparable to the base of the NE Pacific specimens. However, it has an undulating stolon and a significantly smaller size range of styles. It is concluded that the NE Pacific specimens represent a new species, perhaps, most closely allied to *C. nicolae*.

Brachyury expression during development of the calcareous sponge *Sycon ciliatum* sheds light on gastrulation in sponges.

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Sponges represent one of the evolutionarily oldest metazoan phyla. Although they are often considered not to possess true tissues or organs, their embryonic development and metamorphosis from larvae into adult animals are characterized by morphogenetic movements and cell differentiation processes similar to these in Eumetazoans. Recently, more tools for molecular analyses become available, and knowledge on molecular mechanisms of development is increasing for the highly diverse phylum of the Porifera. One of especially important questions is whether a gastrulation-like process – which is present in all other metazoans – occurs during sponge development. While many authors argue there is no gastrulation in sponges, others speculate it happens either during embryonic development or later during metamorphosis. In order to shed light on this issue we cloned a brachyury homologue from the calcareous sponge *Sycon ciliatum* (SciBra). Brachyury is a member of the T-box transcription factor family. It is known to be a conserved player in the majority of animals being specifically expressed during gastrulation. Remarkably, a clear brachyury ortholog is absent in *Amphimedon queenslandica,* the only sponge for which the complete genome has been sequenced. However, Brachyury genes have been reported from other Demosponges, as well as Calcareous sponges and Hexactinellids. We analysed the spatial and temporal expression of SciBra in the course of embryonic development and metamorphosis of *S. ciliatum* using *in situ* hybridisation. SciBra expression is not detectable in adult tissue or at any embryonic stage, but it is strongly expressed in settling larvae and during metamorphosis, and decreasing in young juveniles. The observed expression patterns are highly dynamic, with different cell types expressing Brachyury transcripts as metamorphosis progresses. In settling larvae, expression can be detected exclusively in the ciliated micromeres at the anterior pole. These cells will later constitute the inner cell mass of the postlarva and give rise to choanocytes of the juvenile sponge. In the first phases of the transition from the larva to the juvenile and differentiation of specific cell types, a fraction of pinacocytes, the former granular cells of the larva, begin to express SciBra, together with the majority of internal cells. This pattern changes with the formation of the inner cavity and the differentiation of choanocytes, as predominantly pinacocytes display a high expression level of SciBra. With growth of the young sponge and enlargement of the choanocyte chamber SciBra expression changes again with choanocytes being the only cells in which SciBra transcripts can be detected, albeit at low levels. Therefore, given that Braychyury genes are employed in gastrulation processes in an overwhelming majority of metazoans, our results suggest that gastrulation is present in calcareous sponges rather in the course of metamorphosis than during embryonic development. Further studies utilizing additional pan-gastrulation markers, as well as addressing questions of homologies of structures and processes during development among metazoa will be needed to resolve the issue of gastrulation in sponges.

Disruption of polarity in a sponge: evidence for an ancient origin of the organizer

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When the first multicellular animals evolved, polarity would have been a key feature in organizing the process of feeding. The aquiferous system of sponges, the most ancient muticellular animals, is polarized such that water enters via the ostia and exits via the osculum. However it is unclear whether the establishment of this polarized physiology is under the same molecular developmental control as in other metazoans. We hypothesized that the aquiferous system, and in particular the osculum, polarizes the sponge, and that its formation is established by *Wnt* pathway which are responsible for setting up polarity in many metazoans. We tested the effect of agents known to disrupt the polarity of a wide variety of animals on sponge organization during development. In other animals, these drugs have been shown to block GSK-3b, a negative regulator of the *Wnt* pathway, such that the *Wnt* pathway becomes "overexpressed". Treatment of sponges hatched from gemmules with lithium chloride (LiCl) or alsterpaullone (AP) disrupted the aquiferous system and generated phenotypes with multiple oscula, reduced number of ostia, and choanocyte chamber malformation. We isolated and determined the temporal expression of a *Wnt* homolog. *Wnt* expression is upregulated at hatching and maintained during development of the aquiferous system; expression levels differed in treated animals. In other animals *Wnt* expression gives a region of the developing embryo the ability to induce polarity elsewhere and is referred to as the 'organizer'. To determine whether the osculum has organizer abilities we transplanted fluorescently labeled oscula from newly developed sponges to the dermal tissues of other individuals. Transplanted oscula attached to the host sponge and within 24 hours had connected to and taken over the role as primary osculum. Thus the aquiferous system in sponges can be polarized by expression of signals from the osculum. Together our data suggest that *Wnt* signaling is involved in formation of the aquiferous system in sponges, but further characterization of *Wnt* expression in larvae and metamorphosing juvenile stages is required to clarify its precise role in setting up polarity in the sponge.

Glass sponge reef structure: sexual vs asexual reproduction

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Glass sponge reefs are biogenic structures that cover tens of kilometers of seafloor on Canada's Pacific continental shelf. The reefs are formed by three species of hexactinellid sponge which are able to fuse the skeleton to form a robust scaffold that remains after the tissue has died. Due to the deep habitat (~160m) little is known about the reproduction and recruitment of reef forming glass sponges and therefore the extent to which individual reefs consist of genetically distinct populations. To determine when reef forming sponges reproduce we have studied tissue samples collected at different times of the year for evidence of embryogenesis and larval development, and have examined settlement plates and skeletons of dead animals for evidence of recruitment of juveniles. Glass sponge larvae were only found in one specimen collected in November, but early stages of developing embryos were found throughout tissues of samples collected in late September and October of two separate years. Tiny juvenile cloud sponges were abundant on the skeletons of dead animals collected in summer months from the Alberni fjord (Barkley Sound), but only one juvenile was found in a survey of dead skeleton collected from the Fraser ridge reef. We analyzed video transects carried out by remote operated vehicle for evidence of buds or new recruits and found many examples of small sponges attached to dead skeletons on the reefs, but evidence of sponge budding was equally prevalent. To determine extent to which reefs are formed by asexual budding or by recruitment of new individuals we carried out a tissue compatibility experiment with pieces of sponges collected from adjacent clumps within the same reef and pieces collected from different reefs. Initial results suggested that tissue fusion might be possible between sponges collected from a single region of a reef and even between two sponges collected several hundred meters apart within a reef, but experimental conditions and the method of evaluation of tissue fusion were imperfect. We therefore carried out a second experiment under controlled temperature and light conditions and pieces that remained in contact after 24 hours were fixed and processed for study of tissue contact by electron microscopy. We found that whereas the tissues of pieces taken from the same individual (controls) fused to form a single syncytium, no fusion occurred between pieces taken from different individuals. Together our results suggest that despite evidence for extensive budding in reef forming sponges, sexual reproduction and recruitment is common in pacific coast glass sponge populations, and that reefs are formed by a combination of asexual budding and recruitment of larvae. We are currently developing microsatellite markers to determine the population structure of glass sponges in Pacific coastal waters.

In situ and in tank experiments show glass sponges take advantage of current-induced flow

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Flow through sponges (Porifera) is thought to be enhanced by ambient current due to Bernoulli's principal, pressure differential, viscosity entrainments, or a combination of the three mechanisms. Vogel's test of this phenomenon suggested that an increase in ambient flow increased the flow through sponges that had been killed by immersion in freshwater, but the results were less clear for live sponges. Glass sponges are an ideal subject with which to re-examine the hypothesis because individuals are large (up to 1m tall), vase-shaped with a body wall less than 1cm thick that houses the flagellated chambers. Hexactinellids are also the only sponges that can instantly arrest their feeding current; such exact control over the feeding current seems counter-intuitive to a system based on passive flow. We used Accoustic Doppler Velocimeters (ADVs) to measure flow velocities from glass sponges and ambient water at 165m depth at the Fraser Ridge sponge reef in the Strait of Georgia, and carried out in tank experiments using ADVs in a flow flume, located at Bamfield Marine Sciences Centre, using specimens collected from Barkley Sound. Estimated flow through the sponge filtration system was calculated from measurements made from scanning electron micrographs. Week-long in situ ADV records illustrated that excurrent flow corresponded to local tidal rhythms (measured with a nearby current profiler). Typical pumping rates were 1cm/s or less, but at ambient velocities above 25cm/s excurrent velocity was approximately 5cm/s. We brought animals into flow tanks and measured excurrent velocities in live and killed sponges. We found that *Aphrocallistes vastus* pumped at 1-5cm/s and that an ambient flow of 25cm/s generated a small increase in flow out of the sponge. Both in situ and in the tanks, the sponge pumping pattern occasionally deviated from the ambient flow suggesting it is able to control pumping irrespective of ambient flow. The effective velocity of water across the collar microvilli was estimated to be 0.6μm/s at an axial velocity of 1cm/s. Our results show that while glass sponges can control their excurrent flow velocities by arresting their pumps, they can and do also take advantage of induced current. Given the extensive surface area of the sponge filtration apparatus, even a small increase in excurrent velocity translates into substantial increase in water processed by the sponge.

Biodiversity of shallow-water sponges (Porifera) in Singapore

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In spite of rapid and extensive coastal development, Singapore still has a very high diversity of marine sponges. Some 250 species in 81 genera from 50 families were documented from extensive sampling of both natural and artificial habitats between 2003 and 2010. Of these, 40 sponge species are new records for Singapore, with six new species currently being described. Intertidal habitats contained about 100 sponge species whilst 150 species were recorded subtidally. Interestingly, only about 30% of sponges reported from artificial substrata were found in the natural habitat. This figure is very low, considering that fouling sponges are generally known to be shallow-water sponges from adjacent natural habitats. Not unexpectedly, sponges in Singapore appear to have biogeographic affinities with both Indian and Pacific Ocean species.

Phylogenetic analysis of the sponge genus *Mycale* (Demospongiae; Poecilosclerida)

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Sponge systematics is largely based on morphological characteristics, specifically the skeletal elements such as spicule and spongin architecture. This presents a challenge as sponges are very plastic morphologically, and change in response to environmental or biotic influences. Molecular studies of sponge taxa have revealed that certain groups were not properly classified and needed to be revised. This study is the first phylogenetic analysis of the genus *Mycale* (Demospongiae; Poecilosclerida) using genetic data. *Mycale* is defined by the presence of particular spicules, the palmate anisochela microsclere, which occurs together with a single type of megasclere, usually subtylostyles. As the genus contains approximately 200 species, it is further divided into 11 sub-genera. Fragments of the 18S and 28S rRNA genes were amplified for 12 *Mycale* species from various sub-genera, with a specific primer designed for the 18S fragment. The 18S and 28S fragments followed the same evolutive model and were combined to increase the total number of variable sites. Phylogenetic analyses were carried out using neighbor-joining, maximum parsimony, maximum likelihood and Bayesian methods with 4 other poecilosclerid sequences and one haplosclerid sequence as the outgroup. Results showed that *Mycale* is a monophyletic group within Poecilosclerida, but the existence of some sub-genera was not supported. Additionally, the white morph of *M. laevis* did not group with the other morphotypes of this species and could correspond to a cryptic species. The presence of palmate anisochelae in the *Mycale* genus is a valid taxonomic character to identify this taxon, but the skeletal architectural characters show homoplasy across different clades within the genus, and should not be used to determine *Mycale* sub-genera.

Taxonomy and distribution of some sponges (Porifera) of Cebu, Philippines with notes on their biomedical properties

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Twenty-nine species of sponges from Cebu, Philippines were evaluated for their biomedical potential based on published literature. Fourteen species were cited as containing a variety of new and known bioactive compounds. These belong to 14 genera, 11 families and 6 orders. An assessment of the fourteen sponge species based on the number of compounds present, range of their bioactivity and their presence in the different stations around Cebu revealed that *Hyrtios erecta* (Keller, 1889) has the most number of compounds with bioactivity ranging from anti-microbial and anti-fungal to receptor-interaction-based to cytotoxic. This is followed by *Biemna fortis* (Topsent, 1897), *Stylissa massa* (Carter, 1889) and *Cribrochalina olemda* de Laubenfels, 1954 having at least two categories of the bioactive compounds. On the sponges' distribution in Cebu, *Hyrtios erecta* (Keller, 1889) is widely distributed, present in four of the six stations. This is followed by *Plakortis lita* de Laubenfels, 1954, present in three station and *Biemna fortis* (Topsent, 1897), *Stylissa massa* (Carter, 1889), *Callyspongia (Callyspongia) aerizusa* Desqueyroux-Faundez, 1984 and *Luffariella cf. variabilis* Polejaeff, 1884, present in two stations. Of the six stations, the one in San Francisco in Camotes Islands has nine of the fourteen sponge species with bioactive properties.

Taxonomy of *Farrea* (Hexactinellida, Hexactinosida, Farreidae) from the southwestern Atlantic

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Sponges and scleractinian corals represent two of the most remarkable components of the fauna colonizing deep-water reefs. These are found in fjords, shelf breaks, continental slopes, banks and seamounts around the world, from coastal Antarctica to the Arctic Circle. In Brazil, deep-water reefs are known from Campos and Santos Basins (20.5° - 28° S, South-western Atlantic), between 550 and 1200 m depth, but they may occur from off the north-eastern to southern coasts (3° - 30° S), where important reef building species were found - *Dendrophyllia alternata*, *Enallopsammia rostrata*, *Lophelia pertusa*, *Madrepora oculata* and *Solenosmilia variabilis*. Among the commonest sponge genera found in those ecosystems is *Farrea* Bowerbank, 1862, with 19 recognized species and 11 subspecies and an overall cosmopolitan distribution, tropics to high boreal. Only seven of those species are known from the Atlantic Ocean and this is the first record of *Farrea* from the southwestern Atlantic. The material studied was collected by trawling and ROV in Campos Basin (off Rio de Janeiro State, Brazil - Southwestern Atlantic), between 835-1135 m depth. These specimens are part of a collection obtained by three deep-water assessment projects coordinated by the Research Center of PETROBRAS (CENPES): OCEANPROF - Campos Basin Deep-sea Environmental Project (2004-2008), CAP-BC - Campos Basin Deep-sea Coral Assessment Project (2006-2009) and HABITATS - Campos Basin Environmental Heterogeneity (2008-ongoing). All material was deposited in the invertebrate collection of Museu Nacional/UFRJ. We identified three species belonging to *Farrea*, including a new species: *F. herdendorfi*, *F. occa* and *F.* sp. nov. The three species are easily distinguished by the forms of their clavules: *F. herdendorfi* possesses anchorate clavules only, with heads varying between two extreme forms – an umbellate form and a thimblate form with straight claws nearly parallel to shaft; *F. occa* possesses dermal pileate clavules and atrial anchorate clavules, the typical ones; and *F.* sp. nov. possesses anchorate clavules only also, with heads varying between two forms – a common hooked anchorate form with short regular claws and an anchorate form with slightly sinuous irregularly hanging claws. The new species approaches very closely *F. foliascens* Topsent, 1906 from the Northeast Atlantic (Azores and Madeira Island). It has similar anchorate clavules (with both sinuous and hooked claws), strongly spined pentactins, uncinates with very short barbs, and oxyhexasters and discohexasters as microscleres. Nevertheless, the new species's hooked anchorate clavules are much larger (480-660 µm vs. 300 µm in *F. foliascens*), and it has no discohexasters. Additionally, the dictyonal framework meshes are regular and rectangular in *F. foliascens* while *F.* sp.nov. has irregular meshes. *Farrea herdendorfi* was collected in association with dead corals, *Enallopsammia rostrata*, *Lophelia* sp. and *Caryophyllina* sp. One of the specimens of *F. herdendorfi* was collected together with an unidentified Microcionidae and *Mycale* sp.nov. *Farrea occa* was observed to form mass occurrences in Campos Basin's sponge/coral banks, as seen in extensive video recording undertaken between 900 and 1100 m depth by CENPES/PETROBRAS. Some individuals appear to be more than 1 m long and nearly so in height.

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Confocal laser-scanning microscope and fluorescent *in situ* hybridization applied to marine sponges: Quantification and spatial arrangement of associated microbes

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Based on the microbial density hosted by sponges, the species have been categorized as "high-microbial-abundance" (HMA; former bacteriosponges), with microbial population exceeding in 2-4 orders of magnitude the microbial water density. In contrast "low-microbial-abundance" sponges (LMA) host microbial densities in the range of natural seawater both in concentration (105-106 microbes per gram of sponge wet weight) and in phylogenetic composition. HMA and LMA are considered two basic life strategies product of evolutionary processes with contrasting characteristics. LMA species use to have well irrigated tissues, high pumping rates and low concentration and diversity of microbial associates. Contrarily to HMA species with dense tissue, low pumping rates and with high concentration and diversity of microbial types different from the ones present in the water column. Despite the categorization of marine sponges in LMA and HMA, few studies devoted their time to quantify microbial cells per sponge biomass unit. In most cases, visual assessments have been used either with electron microscopy or by fluorescent *in situ* hybridization (FISH) and polarizated/epifluorescence light microscope. About one decade ago, the use of widefield deconvolution epifluorescence microscopy (WDEM) combined with FISH was proposed as low-cost alternative to confocal laser-scanning microscope (CLSM) to study the occurrence and spatial arrangement of microbes in marine sponges but no data has appeared related to microbial abundances in sponges. Quantification of microbes associated to marine sponges is complex because of the space distribution in the sponge tissue. This is especially evident in HMA species where the compact structure of microbial components similar to microbial biofilms makes the quantification in number of cells very complicated. The use of sponge tissue slurries and centrifugation on the observed microbial distribution resulted in infraestimation of cell abundance because many cells remain attached in the pellet. The use of CLSM, even it is time consuming and requires of complex instrumentation, represents a precise method to evaluate the volume of the sponge occupied by microbial partners. In order to explain the fluxes of dissolved compounds measured in the water processed by Mediterranean sponges, the microbial community of *Dysidea avara*, *Agelas oroides* and *Chondrosia reniformis* have been studied by CLSM and FISH for general bacteria and archaea. These three species have an increasing gradient of microbial abundance, being *D. avara* a LMA and *A. oroides*, *C. reniformis* HMA. In both HMA species, associated microbes appeared in aggregates which make impossible their quantification with most traditional methodologies. We proved several methods, and the most appropriated resulted to hybridize (by Card-Fish) slices of the sponges with the probes Eub I+II+III for general bacteria and Cren 537 and Eury 806 for archaea. Afterwards, several stacks of images have been taken by CLSM and the intensity of the signal was quantified by the software Metamorphc, resulting with reliable data.

Ammonia oxidizing *Archaea* in bleaching barrel sponges: Diversity, distribution and ammonia monooxygenase expression

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The giant barrel sponge *Xestospongia muta* is a prominent species in Caribbean coral reef communities. Tissues of *X. muta* contain cyanobacterial symbionts of the *Synechococcus* group, the loss of which results in two types of bleaching: cyclic bleaching, from which sponges recover; and fatal bleaching, a condition associated with the disease-like sponge orange band syndrome (SOB) and sponge death. In addition to cyanobacteria, most sponges also harbor an array of heterotrophic microorganisms, including bacteria and *Archaea*, that contribute to nutrient cycling, including nitrogen cycle processes (e.g. nitrification). However, nothing is known about how nitrification and symbiotic *Archaea* maybe affected by sponge disease and bleaching events. In this study, terminal restriction fragment length polymorphism (TRFLP) analyses, clone libraries, and relative mRNA quantification of ammonia monooxygenase genes (*amoA*) were performed using a RNA transcript-based approach to characterize the active ammonia oxidizing Archaea (AOA) community present in bleached, non-bleached and SOB tissues of cyclically and fatally bleached sponges. We found that non-bleached and cyclically bleached tissues of *X. muta* harbored a unique Crenarchaeota community similar but not identical to the ones described from other demosponges (i.e. *Cliona*; *Luffariella*; *Axinella*; *Plakortis*; and *Coscinoderma*). In contrast, fatally bleached tissue from the most degraded sponge contained a Crenarchaeota community that was more similar to free-living Crenarchaeota (e.g. sand- and sediment-derived sequences). In addition, we found that in the later stage of fatal bleaching, the bleached tissue became necrotic and *amoA* gene expression increased, probably due to greater release of ammonia from tissue degradation. Thus, results suggest that a shift in the Crenarchaeota community precedes an increase in *amoA* gene expression in fatally bleached sponges. In contrast, cyclic bleaching did not alter the Crenarchaeota community of the sponge and no significant changes in relative *amoA* gene expression were detected either. This study shows that neither the community structure of Crenarchaeota symbionts nor their *amoA* gene expression appear to be affected by sponge bleaching, unless this process is accompanied by tissue death and sponge morbidity.

Synthesis of 2′,5′-linked nucleotides by OAS from marine sponges

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2′,5′-oligoadenylate synthetases (OAS) as a component of mammalian interferon-induced antiviral enzymatic system catalyze the oligomerization of cellular ATP into 2′,5′-linked oligoadenylates (2-5A). OAS activity has also been detected in a variety of marine sponges, which exhibit different 2-5A synthesizing capacities and product profiles. Our data show that two species of the class Demospongiae, North Sea sponge *Thenea muricata* and the Mediterranean sponge *Chondrilla nucula*, can accumulate 2-5A in high amounts. Concomitantly with accumulation of 2′,5′-diadenylates, several 2′,5′-linked heterodimers were detected in the extracts of the sponges. After dephosphorylation, these 2′,5′-heterodimers were identified as A2′p5′N, where N is G, U or C (*C. nucula*, *T. muricata*); as NAD2′p5′A (*T. muricata*) and as G2′p5′N, where N is A or U (*C. nucula*). Though mammalian OASs have been characterized as 2′-nucleotidyl transferases under *in vitro* conditions, the natural occurrence of 2′,5′-oligonucleotides other than 2-5A has not been demonstrated earlier. *In vitro* assays demonstrated that in *T. muricata* ATP was a preferred acceptor for the OAS, while in *C. nucula* GTP was comparable with ATP as an acceptor for the enzyme. That can explain the natural occurrence of 2′,5′-linked heteronucleotides in *C. nucula*, which were identified in the form of G2′p5′A and G2′p5′U. The case of G2′p5′U gives evidence for the presence of non-adenylate 2′,5′-linked compound in sponge cells as a result of OAS activity. The usage of pyrimidine nucleotides as substrates for OAS was quite unexpected, since (2′-5′)oligomerization of these nucleotides by OASs has never been demonstrated. By means of OAS from marine sponges, 2′,5′-linked homo-oligomers up to trimers were obtained from the substrates UTP and CTP. To study the formation of hetero-oligomers, ATP was combined with GTP, UTP or CTP in roughly equimolar concentrations. In such conditions, a variety of 2′,5′-hetero-oligomers (up to tetramers) was obtained wherein all four ribonucleotides could serve as either acceptor or donor substrates for oligomerization. These data refer to the substrate specificity of sponge OASs that is remarkably different from that of mammalian OASs. Notably, the substrate specificity of an OAS depends on the particular sponge species, which might be explained by differences in the enzyme structures of the respective sponges. Further studies may help to elucidate the contribution of the studied catalytic properties of the enzyme to the proposed signaling or gene regulatory functions of OASs in sponges.

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Extraordinary tissue regeneration in the marine sponge *Ianthella basta*

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Given the totipotent nature of their cells, sponges have the ability to recover from damage due to physical disturbances, often developing a protective barrier within 12 hrs of injury. Here we demonstrate the remarkable ability of sponges to recover tissue biomass over a 72 hr period. Six specimens of the marine sponge *Ianthella basta* that appeared health-compromised (tissue necrosis and lesions) were collected at Orpheus Is., northeastern Australia and transported to the Australian Institute of Marine Science where they were kept in a 1,000 l flow-through outdoor aquarium. At this time, the sponges displayed substantial tissue loss, with visible gaps evident between the sponge fibers. At this point, sponges were assumed to have lost the functional capabilities of their choanocyte chambers and were considered likely to die. However, within 72 hrs the sponges displayed rapid recovery and tissue regeneration. In addition to visual comparisons of the sponges over the 72 hr period, the images were compared using the integrated density measurement of Image Tool for Windows (UTHSCA). The integrated density of the sponge tissue effectively doubled within 72 hrs (increasing by 92%), confirming extraordinary tissue regeneration in *I. basta*. The mechanisms behind the tissue loss and subsequent regeneration warrant further investigation as they have significant implications for our understanding of health and recovery in sponge communities.

Spatial patterns in the diversity of sponges off the west and south coasts of South Africa

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Most work on South African marine benthic fauna has focused on shallow sub–tidal and inter-tidal hard substrata, and our knowledge of the deep water fauna (especially from soft sediments) is relatively poor. The shelf areas along the south and west coasts of South Africa are subject to intensive trawling and mining activities and there is a need to more fully understand their faunal diversity and composition for management purposes. Here we investigate sponge communities from this region using data collected from a series of trawl surveys. The preliminary results reveal 50 species in 15 families, and they show that diversity initially declined with increasing depth to ~100-150 m on both coast, then increased to about 150 m after which it declined again. Diversity remained relatively constant with increase in longitude on the south coast to ~24 degrees E, after which it increased. Cluster analysis revealed three distinct groups of sponges (at the 35% level of similarity) on the south coast as appose to five on the west coast of which two are overlapping in the deeper shelf area. The results are discussed in the context of offshore marine protected areas.

Evaluation of the biomonitoring potential of Brazilian sponges

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The use of sponges for biomonitoring has been studied for decades, but a "Sponge Watch" program hasn´t come to reality. The great diversity of species, the specific characteristics of response to pollution, the lack of a species agreed to be used as a global model and the necessity for a deeper physiological understanding of the species-specific stress response seems to be obstacles for the widespread use of sponges as biomonitors. The present work is an effort to screen different species of the Brazilian coastline, evaluate their elemental composition and morphology in different environments subject to different stresses. *Amphimedon viridis*, *Aplysina fulva* and *Desmapsamma anchorata* were collected in two sites in Angra dos Reis, Rio de Janeiro. One site on the coast of Ilha Grande, facing the ocean with constant wave stress and the other inside the Angra Bay, in a small island frequently visited by tourists and next to a commercial shipyard. Mass spectrometry (ICP-MS) was performed on the sponges and water samples. Cr, Ni, Co, Cu, Zn, Cd, Hg and Pb were quantified. Sponge tissue and mineral skeleton (when present) were chemically characterized via X-ray Diffraction (XRD), Infrared spectroscopy (FTIR) and Energy Dispersive x-ray Spectroscopy (EDS) analysis. Morphology was studied using histological sections to account for cell and collagen density. Despite the different sites, water analysis showed no significant difference between sites, and, the elements levels were within Brazilian national environmental regulation. In ICP-MS analysis, *D. anchorata* showed different accumulations of Cr, Co and Pb depending on the site and *A. viridis* had decreased Hg concentration according to its decrease in the water. XRD analysis didn´t show crystalline accumulation of elements. EDS showed that *D. anchorata* chemical composition presents Fe and Ni only when collected in the Angra bay. FTIR didn´t show differences between sponges or collection sites. All spicules analyzed, by all techniques, were constituted solely of Si and O, with no other element. Histology revealed that sponges in the Angra bay had bigger cell density and smaller collagen density when compared to their counterparts from Ilha Grande. This study shows that different analysis may give different results, and complementary approaches must be used. *D. anchorata* revealed an extreme sensitivity to different non-polluted sites and seem less suitable as a biomonitor. *A. viridis* showed some correlation, regarding Hg, to the water and together with *A. fulva* presented a less variable chemical composition, making them more suitable for the follow up *in vitro* studies and comparison with specimens from known polluted sites.

Some features of Baikal sponge mitochondrial genome organization

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Sponge fauna in the Lake Baikal includes the families Lubomirskiidae and Spongillidae. The family Lubomirskiidae is endemic, and includes 4 genera and 13 species and 1 subspecies (Efremova, 2004). The taxonomy of freshwater sponges, in particular the Lubomirskiidae, is problematic and their phylogeny and evolution are not well understood. During the last years many scientists start to analyze the mitochondrial genome to solve of these issues. Moreover, recently the sequence of the mitochondrial genome of the Baikal sponge *Lubomirskia baicalensis* were determined (Lavrov, 2009). However, not only coding regions, but also the intergenic regions in mitochondrial genomes may provide significant data for understanding the taxonomic low level. The nucleotide sequences of the intergenic regions (IGR) of the mitochondrial genome between the genes tRNA-Tyr- tRNA-Ile and tRNA-Ile-tRNA-Met of 20 sponges (10 species) from the family Lubomirskiidae and 6 sponges (3 species) from the family Spongillidae were determined. The length of these sequences ranging from 386 bp in *E. fluviatilis* (Spongillidae) to 980 bp in *L. fusifera* (Lubomirskiidae), which is associated with the presence of multiple indels. Fore sponges were chosen for the more detailed comparative analysis, representatives of all 4 genera the family Lubomirskiidae. An analysis of these sequences has shown many inverted repeats that form secondary structures. Some of these secondary structures were previously identified in *Lubomirskia baicalensis* by D.V. Lavrov (Lavrov, 2009). The IGRs of all analyzed Baikal sponge has A-T-rich regions localized before tRNA genes, which form a stable hairpin, similar in all sponges. We have also found unknown open reading frame (ORF) between the tRNA-Tyr and tRNA-Ile genes of mitochondrial genome of sponges from the family Lubomirskiidae. This ORF encodes the putative protein which has the length to 67 aa. All amino acid sequences possess the start ATG-codon (at position 28) and the stop-codons TAG or TAA at the different positions. Despite the different length of putative protein, it has 21 amino acids conservative region at the N-end, which is the part of putative transmembrane domain. Our study showed that increasing the size of intergenic regions of the mitochondrial genome of the Baikal sponge occurs not only due to the insertion of direct and inverted repeats, but also due to presents the open reading frames. Thus the Baikal endemic sponges differ from cosmopolitan sponges in there features of the mitochondrial genome organization.

Selective filtration in the tropical marine sponge *Rhopaloeides odorabile*: impacts of thermal stress on feeding behavior

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Climate change currently represents the most significant and increasing threat to coral reef ecosystems worldwide. Elevations in seawater temperature may detrimentally affect sponges, especially those involved in tightly integrated microbial symbioses. In this study, the impact of elevated seawater temperature on feeding behavior in the tropical marine sponge *Rhopaloeides odorabile* was assessed. Sponges were reared under temperatures ranging from 27°C-32°C and filtration efficiencies (heterotrophic and phototrophic bacteria) and volume flow rates were measured for replicates from all treatments. Sponge filtration efficiency and flow rates were not significantly affected by temperatures between 27°C-30°C. This is consistent with earlier research that detected no differences in the symbiotic population or sponge health in temperature treatments between 27 and 31°C. In contrast, sponges exposed to 31°C exhibited significantly reduced flow rates and filtration efficiencies on heterotrophic bacteria at 24 hours but maintained normal filtration efficiency on phototrophic bacteria through 14 days. Sponges exposed to 32°C exhibited cellular necrosis and dramatically reduced flow rates and feeding efficiencies on both bacterial types at 24 hours. The threshold for normal sponge feeding behavior was 31°C and there was clear evidence of selective filtration (to more nutritional phototrophic bacteria) by *R. odorabile* in response to thermal stress. This thermal threshold is identical to the symbiosis threshold for corals and their zooxanthellae, indicating that sponges may be similarly threatened by climate change. *R. odorabile* exposed to 31°C for 14 days was able to recover both flow rate and filtration efficiency after a 48 hour recovery period at 27°C, indicating that this sponge is able to rapidly respond to changing environmental conditions.

Elemental composition of sponges along the Mediterranean coast of Israel

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A comparative study on metal accumulation by 8 common demosponge species was conducted to determine the most suitable species as bioindicators of anthropogenic metal contamination. Elemental composition of *Axinella polypoides*, *Axinella verrucosa*, *Sarcotragus* sp., *Psammocinia* sp., *Cinachyrella* sp., *Xestospongia* sp. *Chondrosia reniformis* and *Chondrilla nucula* was determined using ICP-AES spectrometry. Elements analysed were Ag, Al, As, B, Ba, Cd, Co, Cr, Cu, Fe, Li, Mn, Mo, Ni, P, S, Se, Si, Sr, Ti, V and Zn. A total of 73 sponge samples and 10 sediment samples were collected. The 5 different sampling sites along the Mediterranean coast of Israel were from Ashkelon in the south through Tel Aviv, Herezlia, Sdot Yam, up to Achziv in the north, at depths ranging from 3 m to 30 m. The results for each element were analysed by 2-Way ANOVA and Tukey HSD tests were preformed. When the various species were compared *A. polypoides* showed significantly higher levels of B and of S than all other species sampled. *A. verrucosa* showed significantly higher levels of Fe than all other species sampled. *Sarcotragus* sp. showed significantly higher levels of Cu and of P than all other species sampled. *Cinachyrella* sp. showed significantly higher levels of Zn than all other species sampled. *Xestospongia* sp. showed relatively high levels of Ni. When the various sites were compared, all sponge species sampled in the southern site of Ashkelon showed significantly higher concentrations of Al, Cr, Fe and Ti compared to all other sites. A higher concentration of V was also noted in Ashkelon (although statistically non-significant). Sponges sampled in Sdot Yam had the next highest levels of all these elements. Sponges from Tel Aviv showed lower concentrations of Cd and Ni and higher concentration of Cu than all other sites. In two of the collection sites, Ashkelon and Sdot Yam, there is an anthropogenic contribution to environmental metal levels due to coal unloaded from cargo ships to a mile-long pier and conveyer belt. This seems to be reflected in the elevated levels of almost all metals in the samples from these two sites. The surprisingly higher levels of Al and Fe in Ashkelon may be a result of an additional anthropogenic source such as the desalination plant active in the nearby Ashkelon power-station. In the three other sites there is no known anthropogenic contribution to environmental metal levels. Tel Aviv and Herzelia both have similar sediment and sandstone reefs while Achziv is mainly limestone. Sponges sampled from these sites showed mostly lower concentrations of almost all metals analysed, therefore they can be considered as clean or control sites for future work. Of great interest is the possible use of sponges to monitor the effect of the Al and Fe compounds used in the desalination process on the marine environment.

Molecular markers, databases and evolution; a recipe for confusion in taxonomy and systematics of *Haliclona*

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Haplosclerid sponges are some of the most difficult to identify and classify due to their simple morphology and their plasticity under different environmental conditions. Recent molecular evidence suggests an alternative taxonomic scheme is necessary. However, the genetic loci employed (the nuclear ribosomal and mitochondrial CO1 genes) have come under fire due to apparent higher substitution rates in this Order. In this presentation I will discuss these issues, focusing on the taxonomy and systematics of a greater than expected diversity of Irish *Haliclona* species. I will also present data from new genetic regions both for barcoding and phylogeny reconstruction and discuss problems with using the GenBank database for confirming species origins of sequence data.

Sponge overgrowth: a paradigm for lianas

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Sponges commonly out-compete other sessile organism in the race for suitable substrate, a resource often more limiting than food. This is the case of some sponges with superior competitive ability that allows them to grow and overgrow other sessile organism, among these some octocorals. We hypothesized that these sponge-octocorals associations can be likened to lianas growing on trees. In this study associations and overgrowth of two branching demosponges, *Desmapsamma anchorata* and *Iotrochota birotulata*, over different octocorals were studied in Carrie Bow Cay, Belize, during 2009- 2010. Results suggest that sponge attachment on octocorals is conditioned by octocoral substrates, having an advantage on bare axis or affected octocoral tissue versus live tissue, but willing and able to attach on both. There was a difference in the trend of growth and overgrowth for the two sponge species considered, as well as the effect that the sponge growth has on a given octocoral. These sponge-octocoral associations and the ability of some rope-like sponges to attach and grow on these substrates can be likened to lianas whereby the sponge benefits by having a hard-flexible substrate over which to grow, thereby increasing their exposure, while octocorals are toppled, often times towards their detriment.

Taxonomy of *Acarnus* Gray, 1867 collected from the northeastern and southeastern Brazilian coast (Poecilosclerida: Acarnidae), with description of a new species

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Acarnus Gray, 1867 is well-established phylogenetically, defined as non-fistular sponges with cladotylote echinating spicules. There are 23 known species, five of which described from the Tropical Western Atlantic: *A. innominatus* Gray, 1867 (Florida, Cuba, Curaçao, Brazil), *A. radovani* Boury-Esnault, 1973 (Pernambuco State, Brazil), *A. toxeata* Boury-Esnault, 1973 (Bahia State, Brazil), *A. deweerdtae* van Soest, Hooper and Hiemstra, 1991(Bonaire, Netherlands Antilles) and *A. nicoleae* van Soest et al., 1991 (Colombia, Jamaica, Netherlands Antilles, Aruba, Brazil). This paper presents five species collected in the Brazilian states of Alagoas, Bahia, Espirito Santo and Rio de Janeiro. Specimens from shallow-waters (0-12 m) were collected manually by scuba diving or snorckeling in Alagoas and Rio de Janeiro; and by trawling at soft bottoms on the Maraú river estuary (Camamu Bay, Bahia) Deeper water materials (60-65m) were dredged during Project REVIZEE (campaigns CENTRAL V and VI). Our results include the first record of *Acarnus innominatus* from the Brazilian continent (previous records in Brazil are restrict to oceanic areas, like Fernando de Noronha Archipelago and Rocas Atoll), new records of *A. nicoleae* and *A. toxeata*, respectively for the coast of Rio de Janeiro and that of Espirito Santo state. The spicules of *A. innominatus* are styles (380-435/11-20 µm), tylotes (195-300 µm), toxa I (240-560 µm), accolada-toxa (80-200 µm), oxhorn-toxa (55-170 µm), isochelae (11-22 µm), cladotylote I (220-315/4.5-6.5 µm), cladotylote II (95-160/1.0-3.5 µm). *A. toxeata* contain styles (180-229/6.6-12.0 µm), tylotes (319-420/3.1-5.3 µm), cladotylotes I (186-345/3.9-5.5 µm), cladotylote II (106-151/2.1-3.9 µm), accolada-toxa (138-688 µm), curved-toxa I and II (55-348 µm) and isochelae (11.9-13.8 µm). We provide the description of a new species from Camamu Bay, characterized by its thinly encrusting habit with irregular and slightly hispid surface, orange/red color and no apparent oscules. *A. nicoleae* present styles (319-505/10.6-14.0 µm), tylotes (266-425/5.3-7.8 µm), cladotylotes I (172-260 µm), cladotylote II (80-122 µm), oxhorn-toxa (34-218 µm), isochelae (11.8-15.1 µm). The spicular set of the new species is composed by two categories of cladotylotes (cladotylotes I 186-305/4.7-8.5 µm and cladotylotes II 82-172/1.3-5.8 µm), styles (292-707/5-14 µm), tylotes (212-353/2.1-6.3 µm), three shapes of toxas, unique in the genus: accolada-toxas (114-627 µm), oxhorn-toxas composed by two shapes of indistinguishable size (37-175 µm), and palmate isochelae (11.8-14.3 µm). *Acarnus* sp nov. is close to *A. toxeata*, but differs by the possession of regular four cladi on the tyle of large cladotylotes and of styles usually smooth or with sparse spines. *Acarnus* exhibit a worldwide tropical-subtropical distribution, common in shallow water habitats, absent or not recorded yet from Mediterranean (east of Greece), Hawaii Islands and presents a single species in cold waters, *A. erithacus* de Laubenfels, 1927, registered from the North coast of Alaska to United States (California).

Population dynamics of a shallow-water Caribbean sponge: survival vs. recruitment

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Variations in population size and structure of marine sessile organisms are influenced by processes such as survivorship, growth and recruitment. In this study we evaluate the relative contribution of these life history traits on the population growth (λ) of the sponge *Amphimedon compressa*, using size-based matrix models. Models were parameterized with field data on sponge growth, survivorship and recruitment obtained by monitoring twenty $1\ m^2$ quadrants for one year at Las Pelotas (LP) and at Media Luna West (MLW), two reefs in the southwest of Puerto Rico. Estimated population growth (λ) of 0.928 at MLW and 0.789 at LP suggest a declining population at both sites. Differences in λ are attributable to differences in size-specific survival between sites. Elasticity analysis indicates that λ at MLW is greatly influenced by survivorship of small individuals whereas at LP survivorship of small and large size-classes contribute notably to population growth. In general, elasticity analysis also indicates that size-specific survivorship patterns have a greater effect on population growth than reproduction or growth; survivorship contributes approximately 84% and 74% to potential changes in λ at MLW and LP respectively. Comparable life history patterns characterized by higher importance of survivorship relative to recruitment and growth have been described for long-lived species such as woody plants species and gorgonians. This suggests that the life history of *A. compressa* may be characterized by a long life span and low reproductive effort. Nevertheless, in the absence of recruitment λ was reduced by about 12% at MLW and 10% at LP indicating that this process may also play a significant role in the dynamics of this sponge.

South Florida Sponges: an online guide to identification

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The Guide to Identification of South Florida sponges is an interactive web-based platform for the shallow-water (<30 m) sponge fauna from Indian River Lagoon through the Florida Keys, Florida Bay and the Dry Tortugas suitable for use by environmental managers, advanced students, teachers and professional scientists. It includes illustrated keys and descriptions of live sponges, their diagnostic components and available habitat and biological data. The guide is permanently housed at the Nova Southeastern University Oceanographic Center http://www.nova.edu/ncri/sofla_sponge_guide/index.html. The search engine was written in JavaScript. The primary interactive component is divided into three main sets of drop-down menus: External Form, Skeletal Components, and Skeletal Architecture, with 23 characters and 161 character states. Information buttons open files that give diagnostic information about character states, including representative images. Selecting a choice from multiple drop-down menus narrows the search. The guide currently lists 97 species and includes active identification pages for 59 species. All names are given in accordance with the World Porifera Database (http://www.marinespecies.org/porifera/) and the Sponge Guide: a picture guide to Caribbean Sponges (http://www.spongeguide.org/). A workshop held in June 2009 agreed on taxonomic name changes for several taxa (e.g., *Placospongia cristata* for *Placospongia melobesioides* and *Neopetrosia proxima* for *Xestospongia proxima*).

Clearance and respiration rates of shallow *Chondrilla nucula* at varying temperature

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Chondrilla nucula Schmidt, 1862 (Chondrosida, Chondrillidae) is a common Mediterranean sponge that can be found from the surface to approximately 40 m depth. Where canopy-forming algae provide for a suitable understory, it can even colonise low-shore intertidal habitats. As such, *C. nucula* is potentially exposed to sea-surface temperature increase, as it is predicted under the current climate-change scenario. To explore the sponge´s response to temperature increase, we measured clearance and respiration rates in *C. nucula* specimens collected in September 2009 nearby Palermo (Sicily, Italy) when sea-surface temperature was ca. 23°C. Sampled *C. nucula* specimens were kept in an aquarium at the same temperature (T) for 24 h, prior to further 24 h acclimation at the experimental T and testing. Clearance rates (CR; $l\ h^{-1}\ g^{-1}$) were measured using the microalga *Isochrysis galbana* at 15°C, 19°C, 22°C, 26°C, 28°C and 32°C (n=8 per T). Respiration rates (RR; $\mu mol\ h^{-1}\ g^{-1}$) were measured in closed chambers (volume 500 ml) separately on 3 specimens per T. Volume and weight (wet, dry, ash-free) of each sample were then measured. All data were analysed by ANOVA. The observed temperature-effect resulted significant for both physiological responses. CR showed significant differences between values at 15°C and 19°C, and between values at 26°C and 28°C ($15°C < 19°C = 22°C = 26°C > 28°C = 32°C = 15°C$). Significant differences were also detected between RR at 15°C and 19°C, and between values at 22°C and 26°C ($15°C < 19°C = 22°C < 26°C = 28°C = 32°C$). Plotting CR vs. T values showed a normal-shaped curve, with maximum CR between 20°C and 26°C. Relationship between RR and T reflected the typical metabolic response increasing with temperature, with highest values between 28°C and 32°C. Maximum surface temperature values in the area are recorded in August – September and range between 24°C and 28°C (2009 values from EMIS – Environmental Marine Information System, European Commission). According to our results, these are still within the physiological limits of *C. nucula*. However, climate-change driven sea-surface temperature increase has the potential to impair the physiology of this species, with possible consequences on its vertical and/or geographical distributional range.

Sponges from Brazilian oceanic islands: an illustrated taxonomic guide to 70 species

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Well illustrated books about taxonomy of sponges, with *in situ* color photographs of species, are useful tools for experts on systematic, ecology, and biogeography, and are also important for students of biological sciences and environment managers. Yet, these sorts of publications are still scarce in the sponge literature, specially if considering the diversity and importance of Porifera. The Brazilian oceanic islands studied here are remote and isolated sites ranging 260 to 1140 km from mainland and distributed from 0°55'N to 20°30'S, playing an strategic role on scientific, political, economic and environmental scenario in the Atlantic ocean. The Brazilian government and UNESCO consider them areas of high priority for conservation. About 1300 specimens were collected by SCUBA and free dive, from 0 - 50 m depth, between the years of 1998 and 2007. This material is deposited at the Porifera Collection in Museu Nacional – UFRJ, Rio de Janeiro, Brazil. The detailed description of 70 species of sponges classified in two classes, 15 orders, 41 families, and 61 genera from four out of five Brazilian oceanic islands (Atol das Rocas, Fernando de Noronha Archipelago, São Pedro e São Paulo Archipelago and Trindade Island) was presented. Sixty-seven species belong to Demospongiae and three to Calcarea. The descriptions contain data on morphology, ecology, systematic and geographic distribution of the species, and are illustrated by color photographs of live and preserved specimens, light micrographs of skeletal arrangement, and scanning electron micrographs of spicules. Maps of local distribution of these species were produced for each island. An identification key to these species was also provided. This book contributes to the knowledge of sponges in Brazil, to a better understanding of the benthic composition of the Brazilian oceanic islands, and to the management of these areas.

How Dendy and Topsent got it right: the family 'Astraxinellidae' validated by LSU ribosomal DNA sequences.

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The monophyly of aster-bearing demosponges currently belonging to the family Hemiasterellidae has recently been shown to be unsupported by molecular data (Nichols, 2005). Partial large ribosomal subunit (28S) sequences were obtained for a number of species from Britain and Ireland. Species belonging to the genera *Stelligera*, *Paratimea* and *Halicnemia* were found to group closely in a tree based on 28S rDNA sequence data. They were distant from species of *Hemiasterella*, *Axos* and *Adreus*, which constitute the family Hemiasterellidae. The clustering of *Halicnemia* and *Paratimea* was predicted by Topsent in 1893 when he described *Paratimea constellata* as *Halicnemia constellata* on the basis of the distinctive, large, centrotylote oxea shared between these species. A new family name is needed for the group containing *Stelligera* and *Halicnemia* but the name Astraxinellidae Dendy, 1909 is not available as there is no genus *Astraxinella*. It is shown that molecular data can often support particular elements of traditional classifications and refute alternative interpretations.

Is dissolved organic matter a food source for coral-excavating sponges? A hypothesis

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Coral reefs in the Caribbean are under environmental stress. Eutrophication enhances the growth of benthic macroalgae at the expense of life coral cover. Interestingly, eutrophication also enhances the abundance of excavating sponges (Holmes, 2000). Previous studies have shown that dissolved organic matter (DOM) is an important food source for open reef and encrusting cavity sponges (Yahel et al., 2003 and De Goeij et al., 2008). We study the release of DOM by benthic macroalgae as well as stony corals and its uptake by excavating sponges such as *Cliona* sp. and *Siphoniodictyon* sp. We hypothesize that the growth and bioerosion rate of excavating sponges is enhanced by DOM. Since benthic algae may release more DOM than stony corals do per unit biomass, an increase in algal cover may also lead to an increase in DOM production per m^2 reef. This may possibly coincide with a higher abundance and bioeroding activity of excavating sponges. Enhanced bioeroding activitiy of sponges may therefore play a key role in the current demise of 3D complexity in reefs throughout the Caribbean. Preliminary results will be presented.

Primmorph cryopreservation

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Sponges are positioned among the lowest metazoa and the study of their biology should provide relevant information on the evolution of eucariotic cells. In addition, these simple animals are a precious source of natural compounds useful for cosmetics and pharmaceutics. Despite their worldwide diffusion and body simplicity, it is nevertheless difficult to obtain controlled laboratory models for the study of their biology. Nowadays, 3D sponge cell culture (primmorphs) seems to be one of the most promising approach for this target. In this study, for the first time, a protocol for freezing marine sponge cells in their 3D aggregation status has been developed. In order to achieve this, we have used primmorphs obtained from the marine sponge *Petrosia ficiformis*, a common Mediterranean sponge already studied in our laboratory previously. Although protocols for freezing cells are well established in humans, animal and plants the research on sponge cells cryopreservation system is relatively scarce. Our experimental conditions demonstrate that frozen dissociated sponge cells after thawing never gave primmorphs, they just stick to the plate. A complete different result was obtained by directly freezing primmorphs. In this study, we tested different freezing conditions and the one based on the use of natural sea water supplemented with 10% FBS and 10% DMSO gave the best reults. After thawing, we assayed different biochemical method like calcein and MTT-assay in order to verify primmorphs viability and the results were compared with unfrozen samples. Both assays confirmed the living status of primmorph after thawing. To assess the occurence of some interference of bacteria with MTT-assay (wich could give artifactual over estimation), the same test was performed also after primmorphs pre-incubation with anthibiotics. The obtained results gave similar values to the untreated samples. In the same experimental conditions, telomerase activity was assayed and the results confirmed an active cell proliferation in thawed primmorphs. Finally, an active spicologenesis was detected in thawed primmorphs by qPCR for silicatein gene expression evaluation and by quantification of silica derived from new spicules production.

Detection and Isolation of Mannose-Binding Proteins from Philippine Marine Sponges Collected in Bani, Pangasinan, Philippines

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Lectins are carbohydrate-binding proteins that play important roles in cellular recognition, adhesion, regulation, symbiosis, and innate immunity.This study aims to detect and characterize mannose-binding lectins or MBLs from marine sponges collected in Bani, Pangasinan, Philippines. Thirty-three sponges were collected, homogenized, and centrifuged. The supernatants were collected and analyzed using the Bradford protein assay. Mannose-binding proteins were detected using a lectin probe composed of a mannose-bovine serum albumin-horseradish peroxidase bioconjugate. Five sponges designated as PNG-789 (*Myrmekioderma sp.*), PNG-766 (*Plakinastrella sp.*), PNG-785 (*Agelas sp.*), PNG-771 (*Petrosia sp.*), and PNG-757 (*Agelas sp.)* with highest mannose-binding protein concentrations were tested further for corresponding specific activities using the hemagglutination assay against human blood types A,B, and O. Antifungal activity of the crude extract from PNG-766 was also detected using Kirby-Bauer Disc diffusion assay against Saccharomyces cerevisiae and *Candida albicans*. Isolation of the mannose-binding lectin from PNG-771 was done by affinity chromatography using a mannan-agarose column. The isolated lectin was further subjected to non-reducing SDS-PAGE and revealed a 42.31 kDa protein band. The biochemical properties of the affinity-purified mannose-binding lectins isolated from the marine sponges are currently being investigated. This study provides knowledge on marine sponges as a rich source of lectins that have a wide range of potential applications in glycobiology, as isolation and characterization tools for glycoconjugates and diagnostic tools for cancer.

Florida reef sponges harbor coral disease-associated microbes

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Sponges can filter large volumes of seawater and accumulate highly diverse and abundant microbial communities within their tissue. Culture-independent techniques such as fluorescent *in situ* hybridization (FISH),16S small subunit (SSU) rRNA gene analyses, and transmission electron microscopy were applied to a study of south Florida reef sponges. FISH detected several coral disease-associated bacteria (CDAB) harbored within *Agelas tubulata and Amphimedon compressa* host sponges that are potential bacterial pathogens including *Aurantimonas coralicida*, *Cytophaga* sp., *Desulfovibrio* spp., *Firmicutes*, *Serratia marcescans*, and *Vibrio mediterranei*. *A. compressa* and *A. tubulata* mesohyl 16S rRNA sequences derived from each sponge host indicated a predominance of Actinobacteria and Gammaproteobacteria respectively. Positive FISH detection of Actinobacteria and Gammaproteobacteria provided visual confirmation of the presence of these bacteria, and supported the 16S rRNA sequence data. Spatial differences in the distribution of targeted bacteria were seen within sponge hosts. Transmission electron microscopy of *Aplysina fistularis* and *A. compressa* indicated there was a higher concentration of bacteria in the choanosome compared to the ectosome in both species. These results have interesting implications for ongoing coral reef disease research and marine sponges as sentinels of ecosystem health or reservoirs of potential microbial pathogens.

A preliminary analysis of the transcriptome of *Tethya wilhelma* – putative gene targets for functional studies on contraction and integration

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The small globular *Tethya wilhelma* (Demospongiae, Hadromerida, Tethyidae) is one of the few sponge species that can be bred and maintained in laboratory aquaria fully independent from natural sources. Due to its reliable asexual reproduction by budding, which can be induced by changing environmental conditions, *T. wilhelma* shows high potential as a model system for asexual reproduction and other aspects of demosponge biology. At present, however, we lack knowledge on sexual reproduction in laboratory cultures. Nevertheless, adult and juvenile specimens as well as buds are available in higher numbers for studies on living and fixed animals. In particular, the macroscopic and microscopic anatomy, contraction behavior and kinetics, physiology of signal integration and endogenous rhythms as well as the dynamics and development during budding have been studied in detail or are under investigation in *T. wilhelma*. In addition, the mitochondrial genome of *T. wilhelma* is completely sequenced. However, the nuclear genetic data were not available until recently. Here we report on large scale 454-sequencing efforts of the *T. wilhelma* bud/juvenile transcriptome and an attempt to establish an RNAi protocol for functional gene knockdown studies. In a first analysis of the transcriptome data, we aimed at analyzing genes putatively involved in contraction and integration. Based on a preliminary EST sequencing project, we additionally aimed at developing a working RNAi protocol for a demosponge. For RNA extraction, a large number of buds and juvenile sponges were collected from actively reproducing aquarium cultures. A first batch was used for random sequencing of a cDNA library. Data from this approach was used to clone actin as a first target into an RNAi vector which was used to produce dsRNA with which juvenile sponges were treated for up to 9 days. Actin mRNA levels were determined by qPCR and referenced to the transcription levels of TALDO (transaldolase of the pentose phosphate pathway) and RPS5 (40S ribosomal protein S5). In a another approach a second, larger batch of around 100 buds and juveniles were used for more complete transcriptome sequencing using the 454-Titanium platform. Data from this approach were analyzed for contraction- and integration-related genes. A first analysis of the transcriptome revealed information on proteins and pathways putatively involved in contraction and integration of *Tethya wilhelma*. Here, we focus on cytoskeleton proteins as well as receptors with agonists which were tested in previous physiological studies. In our RNAi pilot study we were able to partially down-regulate the expression of actin over several days. Our results demonstrate that functional studies by gene knock-down are possible in sponges.

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A close look at a diversity of diversities in the Mediterranean sponge *Spongia lamella*

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Understanding diversity is a major ecological goal that has expanded to other social fields. The term biodiversity commonly refers to ecosystem, species and genetic diversity and the complex set of interactions within and between those levels. Diversity is linked to variability in and of life and it is a dynamic concept. However, little efforts have focused on the connections between multiple levels of diversity. Understanding the relationships between multiple levels of diversity may shed light on the mechanisms that generate and maintain biodiversity. In this study, we investigated the genetic, bacterial, and chemical diversity of the endangered Mediterranean sponge Spongia lamella (previously Spongia agaricina). These levels of diversity are intrinsically related to sponge evolution and could have strong conservation implications. We developed microsatellite markers and used multi-locus-heterozygosity and gene diversity to assess genetic diversity. We used denaturing gel gradient electrophoresis (DGGE) and quantitative PCR to compare, identify, and quantify main bacterial clades in the sponge. We used HPLC chromatograms to obtain chemical profiles. Then, we applied the Shannon index of diversity to calculate bacterial and chemical diversity. We calculated these three diversity indexes in multiple populations across the western Mediterranean and tested whether the three diversity levels investigated in this study were associated at a population level. Genetic analyses revealed that sponge populations were highly and significantly differentiated. The population from Ceuta presented the highest genetic diversity. We also observed bottlenecks for many populations, Ceuta being the least affected. DGGE revealed semi-quantitative intra- and interpopulation variation and significant differences in bacterial fingerprints between sponge populations. Quantitative PCR performed on Chloroflexi, Acidobacteria, and Actinobacteria (which are found in high abundance in this species) also differed significantly between populations. The Atlantic population (Berlangas, Portugal) presented the greatest differences for all the bacterial clades amplified. Comparison of chemical profiles showed that only nine compounds out of 22 differed between sponge populations. Chemical diversity was quite low, because of the presence of 2 major compounds (nitenin and an unidentified compound), but differed significantly between sponge populations. We found higher dissimilarities in genetic, bacterial, and chemical diversity with increasing geographic distance between populations. However, we failed to detect significant correlation between those three levels of diversity at a population level. This suggests that multiple geographically variable factors could regulate the genetic, bacterial, and chemical diversities investigated in this study, highlighting the complex nature of the mechanisms behind biodiversity.

Antiophidian properties of marine sponges against *Bothrops jararaca* snake venom

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Snakebites are a health problem in many countries because of the global incidence of accidents that exceed 2.5 million per year leading victims to death. Envenomation by snakes is characterized by hemorrhage, necrosis, clotting, neurotoxicity and hemolysis. To counteract such effects, antivenoms have been regularly used for more than a century. Apart from side effects such as anaphylactic effects, antivenoms did not neutralize local tissue damage contributing to an increase of the severity and morbidity. Thus, it is relevant to search for new strategies to improve antiserum therapy. A great number of researchers are trying to identify molecules from natural sources with antiophidian properties and marine organisms are good candidates. A variety of pharmacologically-active chemical metabolites in marine organisms has been reported such as antifungal, anticancer, antiviral, antimicrobial molecules. Marine sponges are among the richest sources of these metabolites. In this work, we report the antiophidian properties of crude organic extracts in 10 Brazilian marine sponge species (*Amphimedon viridis, Aplysina fulva, Chondrosia collectrix, Desmapsamma anchorata, Dysidea etheria, Hymeniacidon heliophila, Mycale angulosa, Petromica citrina, Polymastia janeirensis, Tedania ignis*) against *Bothrops jararaca*'s biological activities: coagulation, hemorrhage, hemolytic (indirect hemolysis test) and azocaseinolytic. Marine sponges were collected in Rio de Janeiro state, extracted with acetone (after liophilization) and dissolved in dimethylsulfoxide. All extracts were able to inhibit the proteolytic activity of *B. jararaca*, except that of *H. heliophila*. Only *P. citrina* prevented efficiently plasma clotting and protected mice from hemorrhage induced by venom. Our data demonstrate that sponges are capable of inhibiting such activities with different potencies. We conclude that Brazilian sponges may be useful for treatment of *B. jararaca*'s accidents and have potential for the discovery of antiophidian molecules.

Studies on the mode of cell division during differentiation of archeocytes (pluripotent stem cells) in *Ephydatia fluviatilis*

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We are trying to clarify the stem cell system using the asexual reproduction system (gemmule hatching) of a freshwater sponge, *Ephydatia fluviatilis*. The principals of the stem cell systems are regulation of differentiation and self-renewal, which might involve cell division or the microenvironment. How archeocytes differentiate to certain types of cells during gemmule hatching is one of the important issues to be clarified. Since archeocytes are actively migrating stem cells, we hypothesized that their differentiation to certain cell types may be regulated by particular environments such as contact with, or secretory signals from, specific types of cells. For the first step, we focused on the mode of cell division of archeocytes to get clues about the microenvironment for archeocyte differentiation. We succeeded in identifying an archeocyte-specific gene, EfMusasahiA, by *in situ* hybridization screening. Musashi is a RNA binding protein originally found as an important player in maintaining neural stem cells during asymmetric cell division in *Drosophila* neural cell differentiation. Accumulating studies about Musashi family genes in various cell systems in various organisms suggest that Musashi family genes have a fundamental role in maintaining the stem cell state. Identification of EfMusashiA as an archeocyte molecular marker enabled us to investigate the mode of cell division during differentiation of archeocytes. First we elucidated the precise localization of EfMusashiA mRNA and protein during cytokinesis. i) There was no difference of the future daughter cells in volume in all of the detected mitotic-phase (M-phase) archeocytes. ii) Both EfMusashiA mRNA and protein were uniformly distributed during mitosis of archeocytes. Furthermore, iii) M-phase archeocytes could be divided into two types. In one type (Type I), high expression of EfMusashiA mRNA and protein were maintained during M-phase, whilst in the other type (Type II), the expression levels of both EfMusashiA mRNA and protein were decreased to 1/2 and 2/3, respectively, of those in Type I. We speculated that Type I archeocytes were undergoing selfrenewal and Type II archeocytes were undergoing differentiation. Based on these results, we propose that archeocytes undergo symmetric cell division both to self-renewal and to produce differentiated cells. These results are the first step forward for clarifying regulatory mechanisms of archeocyte selfrenewal and differentiation.

Infection by Aplysina Red Band Syndrome alters sponge-associated bacterial communities

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The decline of shallow water coral reefs has been attributed to a number of factors, including contagious diseases of reef organisms. Most studies of coral reef diseases have focused on scleractinian corals, whereas sponge diseases have been less frequently documented. Investigations of Aplysina Red Band Syndrome (ARBS), which affects Caribbean rope sponges, indicated that the rust-colored leading edges of the lesion were likely due to the presence of filamentous cyanobacteria. Although the presence of this distinctive coloration is used to characterize the diseased state, it is not yet known whether these cyanobacteria are the causative agents of this disease. In an attempt to better characterize the bacterial and cyanobacterial associates of both healthy and visibly ARBS-affected tissue, terminal restriction fragment length polymorphism (T-RFLP) analyses were conducted. Samples collected in Belize and the Bahamas in 2008 and 2009 showed significant differences in the associated communities by date (year of collection), health status (healthy or ARBS-affected), and location. These data suggest that the bacterial communities associated with *Aplysina* sponges vary over both temporal and spatial scales, even though the ARBS lesions appear visually similar. Additional studies to evaluate the impact of nutrients on ARBS showed no significant effect of exogenous nitrogen on the composition of the associated bacterial communities. Clone libraries generated from healthy and ARBS-affected tissue indicated that *Synechococcus* and *Prochlorococcus* species were more abundant in healthy specimens while *Leptolyngbya* spp., filamentous cyanobacteria, were more abundant in ARBS-affected sponges. Members of the Actinobacteria and Chloroflexi were common constituents in both healthy and ARBS-affected samples. Characterizing the etiologic agent(s) and the host responses to disease may help to explain the emergence, distribution, and virulence of marine pathogens.

Elevated metals in tropical sponges, *Halichondria phakelloides* and *Spheciospongiae vagabunda*, in Darwin Harbour, northern Australia.

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Darwin Harbour (Northern Territory, Australia) is a tropical, macrotidal estuary. Water quality in Darwin Harbour is good (Darwin Harbour Region Report Cards 2009, NRETA) due to the relatively low population (117,000), low industrial development and the rapid dilution of solutes through large tidal range. Increased industrialization and urbanization is expected, and potential increases in seawater temperatures will add pressure on the marine environment. There are already localized impacts such as sewage discharge and stormwater drainage. Because solutes are readily diluted, biota such as sponges that integrate contaminants over time can be more robust bioindicators for assessing water quality. We assessed impact at different sites in Darwin Harbour by measuring sponge, sediment and water chemistry. We collected four sponge species: *Halichondria phakelloides, Spheciospongia vagabunda, Iotrochota* sp. and *Paratetilla* sp., seawater and sediment at each of four sites in Darwin Harbour: (1) Larrakeyah diffuser (at the sewage effluent discharge pipe), (2) Larrakeyah control (~400 m from discharge), (3) Stephen's Rocks (control site) and (3) East Point (unknown impact). Filtered (0.45 μm) seawater was acidified and stored at 4°C. Sponge tissue was freeze-dried and ground. Sediment was sieved to < 2 mm, oven dried and ground. Sponge samples were digested using HNO_3/H_2O_2; sediment samples were digested using $HNO_3/HClO_4$. All samples were analysed by ICP-MS for Al, P, V, Cr, Fe, Mn, Mo, Co, Ni, Cu, Zn, Ga, As, Cd, Pb, and U. Compared to other sites, seawater and sediment collected at the diffuser had higher levels of almost all elements measured, particularly P, Mn, Fe, Cu and Zn, indicating impact from discharged effluent. Although solutes are readily diluted from the diffuser, sponges on or near the diffuser will be filtering contaminated water and in contact with contaminated sediment. None of the sponges tested, however, showed significant differences in metal concentrations between sites. Of significant note though, was the level of molybdenum measured in *H. phakelloides*, which was on average 100 times greater than levels measured in other sponge species, particularly near the diffuser. Molybdenum was barely detectable in most of the sediment samples and was present at background levels in seawater and sediment at all sites. Cyanobacteria require molybdenum for nitrogen fixation and cyanobacteria DNA signatures have been detected in *H. phakelloides*. This association is the focus of ongoing studies. We also measured very high levels of Co, Cd, Zn and Ni in *S. vagabunda:* up to 400 times greater than in other sponge species. This phenomenon occurred at all sites and using multiple approaches including X-ray microanalysis we seek to determine if it is a function of the sponge or its microbes.

Macrofauna inhabiting the sponge *Paraleucilla magna* (Porifera, Calcarea) in Brazil

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Sponges have long been considered to be "living hotels" due to the great diversity and abundance of organisms that can be found living inside them. These associated organisms may find habitat, food, substrate, and protection in the sponge's body. Despite the ecological and taxonomic importance in studying the associated fauna of sponges, there are no publications on this subject to calcareous sponges. In fact, the only work that tried to find associated fauna in a Calcarea, found no associated organisms. Therefore, this is the first work to present the macrofauna associated to a calcareous sponge. *Paraleucilla magna* (Porifera, Calcarea) is a very abundant and widely distributed species in Brazil, occurring from Rio de Janeiro to Santa Catarina states (around 1,400 km apart) and it is considered to be an exotic species in Brazil. The aim of this work is to describe the diversity and abundance of the associated fauna of *P. magna* in Rio de Janeiro, Southeast Brazil. Up to five specimens were monthly collected during one year in Rio de Janeiro, totalizing 54 specimens. The individuals were carefully fragmented under a stereomicroscope in order to remove the associated macrofauna. This macrofauna was separated in morphotypes and identified with the help of specialists. Forty eight taxa were identified, representing ten phyla: Arthopoda, Annelida, Mollusca, Ectoprocta, Chordata, Cnidaria, Echinodermata, Platyhelminthes, Porifera and Nemertea. The most frequent groups were: Crustacea (70.4%), Polychaeta (57.4%), Mollusca (48.2%) and Ectoprocta (40.7%). Ascidians, hydrozoans and echinoderms were found only in a few individuals, and platyhelminthes, pycnogonids, sponges and nemerteans were found only in one individual. The most diverse group was Crustacea with 11 species, followed by Polychaeta with five. These groups and also echinoderms have been reported as the most common organisms associated to demosponges all around the world, including Brazil. In *P. magna*, many juvenile individuals (including crustaceans, molluscs, polychaetes, and echinoderms) and pregnant females (amphipods) were found, suggesting that this sponge may serve as a shelter for these organisms at least in some period of their life cycles. Many of the species found in association with *P. magna* are widely distributed around the globe and are commonly found associated with sponges (Demospongiae) but also with other benthic organisms. This indicates that these organisms are not specialist sponge dwellers. Our results reveal that calcareous sponges also harbour a great sort of invertebrates and reinforce the idea that poriferans are true "living hotels".

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Sponge feeding by the reef fish *Holacanthus ciliaris* (Pomacanthidae) in São Pedro e São Paulo Archipelago, Brazil

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The diet of the reef fish *Holacanthus ciliaris* is composed mainly of sponges, which comprise 90-95% of its stomach contents in the Caribbean and in Bahia State, Brazil. São Pedro e São Paulo Archipelago (formerly St. Paul's Rocks) is a small (1.5 km^2) and remote oceanic site (0°55'N - 29°21'W, 1010 km from mainland) where lives an isolated population of *H. ciliaris*, with individuals presenting rare and diverse chromatic patterns. The sponges of the archipelago are usually cryptic and the community has low diversity, with only 26 species known. It was hypothesized that in such conditions *Holacanthus ciliaris* may shift its diet towards more abundant benthic prey, especially algae. The goal of this study was to determine the qualitative and quantitative composition of the diet of *H. ciliaris* in the archipelago and to relate it to the abundance of sponges in the area. Five adult specimens of *H. ciliaris* were collected in Spetember 2007 through spear fishing between 5-15 m depth and had their stomachs removed and fixed in 70% alcohol. The stomach contents were examined, weighed and analyzed individually. Sponges, algae and bryozoans were separated in morphotypes according to external morphology and subsequently identified and weighted to determine their proportion in the fish diet. Sponge abundance in the field was estimated in 40 quadrats 0.25 m^2, randomly distributed from 5-20 m depth, and compared to abundance in the diet through Manly's resource selection function. The diet of *H. ciliaris* in the archipelago had a higher proportion of sponges (56-83%) compared to algae (25%) and bryozoans (6%). Thirteen species of sponges were identified in the stomach contents. The most frequent were *Clathria calla* (33%), *Spirastrella hartmani* (24%), and *Tedania ignis* (14%). The most abundant sponges in the field were *Scopalina rutzleri* (43.2%), *Chondrosia collectrix* (24.7%), and *Spirastrella hartmani* (7.6%). There was no correspondence between sponge abundance in the field and in the diet. Our results suggest that *Holacanthus ciliaris* feed on rare species either because the most abundant sponges are chemically defended or because fish predation is reducing the abundance of its preferred benthic preys in the archipelago.

Sponge distribution in the ABBA cluster, Pangasinan, Philippines

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Sponges are part of the least understood fauna in the Philippines, receiving little attention amidst extensive researches on other marine organisms. They play important roles in the marine ecosystem and these assemblages are highly influenced by the natural biophysical and anthropogenic factors. This study reports the distribution of sponges and different sponge growth forms in the Anda-Bolinao-Bani-Agno (ABBA) cluster in Pangasinan, Northwestern Philippines in relation to present reef conditions. The sponge collection yielded more than 300 sponges from eight sites spanning four municipalities in Pangasinan, Northwestern Philippines. The sponges were representative of 155 morphospecies and 16 different growth patterns. Diversity is apparent as almost 60% of the sponges were encountered only once while not one sponge was seen at all 8 sites. The sites were dominated by haplosclerid and dictyoceratid sponges. The encrusting, massive and repent growth forms were most ubiquitous and abundant. Principal component analyses showed that low energy areas with high sedimentation tend to have upright forms (arborescent, tubular) sponges while higher energy areas tend to have low profile forms (encrusting, repent). Also, high species richness and also morphological richness was found in vertical habitats, probably due to presence of more microhabitats (overhangs, crevices) and high exposure. Fish diversity and abundance may also play a role in disturbance and spongivory. The presence of the different morphological types may be indicative of their ecophysiological adaptations, physiognomic functions and interactions that sponges have in the reef. The knowledge of the distribution of the sponges will contribute not only to biodiversity studies in the Philippines but also to other applied researchers.

A multi-gene assessment of heat stress response in the Great Barrier Reef sponge *Rhopaloeides odorabile*

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Increasing sea surface temperatures are causing detrimental effects on tropical ecosystems worldwide. Research attention has focussed on coral stress responses and the partnership between corals and their symbiotic zooxanthellae. Porifera are also a critical component of coral reefs due to their abundance, diversity and their role in benthic-pelagic coupling. However, few studies have assessed the response of sponges to thermal stress. We developed a quantitative PCR assay for the Great Barrier Reef sponge *Rhopaloeides odorabile* to assess variation in gene expression following thermal stress. We quantified expression levels of six target genes: 1) actin related protein (arp 2/3) involved in controlling actin polymerization, 2) calmodulin (CaM) involved in signal transduction, 3) ferritin involved in iron storage, 4) ubiquitin conjugating enzyme (ub. conj. enz.) involved in protein ubiquination, 5) heat shock protein 90 (HSP90) and 6) heat shock protein 40 (HSP40). In an attempt to detect a molecular stress response prior to visual signs of necrosis, we applied qPCR to samples experimentally exposed to heat stress. Temperature treatments included 27°C (ambient), 31°C and 32°C and sponges were sampled after 1 d, 3 d and 14 d. The assay incorporated biological triplicates and technical duplicates for each sampling time and temperature. Results are expressed as fold changes relative to the 27ºC treatment at each time point. Reference genes (a-tubulin, 28S and ubiquitin) were validated by NormFinder. In sponges treated at 31ºC and 32ºC there was a significant reduction in expression of the arp2/3 and ferritin genes after 1 d and a further reduction in sponges kept at 31ºC for 3 d. CaM expression was significantly reduced in sponges at 32ºC after 1 d and both the CaM and HSP40 genes were down-regulated in sponges kept at 31ºC for 3 d. Whilst the HSP 90 gene was down-regulated after 1 d in sponges at 31ºC and 32ºC, after 3 d the 31ºC sponges maintained the decreased expression whereas sponges at 32ºC increased expression of HSP90. Whilst expression of the ub. conj. enz. was significantly reduced in sponges at 31ºC after 3 d, expression was increased in sponges at 32ºC. The rapid decrease in expression of most genes (arp 2/3, ferritin, CaM, and HSP90) at 31ºC and 32ºC within 24 h indicates that the sponge goes through an initial shut-down of its molecular systems in response to thermal stress. These results are consistent with research showing a suppression in transcription after an initial stress treatment (Kassahn, 2006). The increased expression of HSP90 after 3 d at 32°C indicates a slow activation of the heat shock response system but is consistent with its role as a chaperone for directing degraded proteins to proteolysis. Whilst sponges at 32ºC only survived for the first 3 days, none of the genes in sponges at 31ºC were significantly different to the 27ºC controls after 14 d. This indicates a very strict thermal threshold for *R. odorabile* between 31 and 32ºC and is consistent with previous findings based on sponge necrosis and symbiotic disruptions in this species.

Variations in composition, abundance, diversity and cover of reef tract sponge assemblages in Broward County, Florida, 2002-2006

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Sponges are major macrofaunal components of coral reef ecosystems, often outnumbering corals in richness, abundance and coverage. Although reefs along southeastern Florida have exhibited substantial deterioration over the last few decades and are currently under intensive scrutiny, most investigations have omitted sponges. This study focuses on reef sponge assemblages along Broward County, Florida, from 2002 through 2006, and examines changes in assemblage composition and areal coverage over time. The photographic database derives from an ongoing study of stony and soft coral coverage carried out by the National Coral Reef Institute (Nova Southeastern University) and includes twenty-six 20 x 2-m transects. Forty images covering 1 m^2 each were taken per transect (two rows of 20) via scuba. Sponges were identified and their areal coverage measured in a total of 4,600 images using Coral Point Count with Excel extensions (CPCe) software. Data includes species counts, minimum and maximum coverage by species, transect and year, as well as total and average areal coverage by transect and year. Dominant species include *Niphates erecta*, *Amphimedon compressa* and encrusting species. Comparisons among species, transects and years are being carried out using Primer software. Initial analyses indicate a decrease in overall areal coverage by sponges over the duration of the study, but also that some species (e.g., *Niphates erecta*) have exhibited an increase in abundance. During 2005, multiple transects were affected by hurricanes Katrina and Wilma, which damaged or removed some sponges and buried others. The more common species appear to have recovered more rapidly, but it is not yet clear whether the same dominance hierarchy was retained.

Taxonomic diversity and distribution of sponges at La Restinga Lagoon, Venezuela

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The composition of sponge communities in mangroves are usually taxonomically different from the composition found in neighboring communities, such as seagrass beds and coral reefs. Because of this divergence and considering the ecological importance of this group in Caribbean mangroves, this study was aimed at assessing the taxonomic diversity of Porifera in the Laguna La Restinga. In order to conduct this assessment, two goals were established: (1) the quantification of taxa, considering taxonomic afiliation; and (2) test the null hypothesis that the distribution of species of sponges in the Laguna La Restinga is spatially homogeneous. Seven localities were chosen within the lagoon. At each locality two line transects (15 m long each) were laid along the mangrove fringe separated by 15 m. We conducted five surveys within July of 2004 and August of 2005, with three months between each sampling event. We quantified the number of species, genus, families, order and classes for each locality; and using this information we created a similarity matrix based on the taxonomic dissimilarity coefficient described by Clarke and Warwick. The matrix was analyzed using a cluster analysis by the method of group averages and a SIMPROF test was run to verify that the separation of groups was significant. As a result, 45 species were identified, being Chalinidae (10 species), Mycalidae (6 species), and Halichondriidae (4 species) the richest families in the lagoon. Moreover, class Calcarea identified two orders with four species in total. The greatest number of species was found in the entrance of the lagoon, followed by middle and edge locations having the lowest richness. Localities from the entrance of the lagoon displayed greater complexity and taxonomic equitability. The locality with less structure or taxonomic evenness observed was Gieles (an internal lagoon). This locality had a structure significantly different than expected because of its species richness when compared to the taxonomic structure in the whole Laguna La Restinga. Gieles presents virtually all orders, but with one to three species per family, far less than other locations. Furthermore, species in the lagoon showed a clear spatial pattern of distribution. This pattern showed three dissimilar groups (SIMPROF test, $p < 0.05$). This grouping coincides with the spatial closeness among localities but also with the strong environmental gradient existing in Laguna La Restinga. The richness of sponge species observed in the Laguna La Restinga is among the highest values reported for other Caribbean mangrove ecosystems and the country.

Richness and taxonomic composition of sponges in coastal habitats of high ecological importance in Venezuela

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Based on the Natural Geography In Shore Areas project (NaGISA), part of the international Census of Marine Life (CoML, www.coml.org), the set objective was to conduct a monitoring plan to study the distribution patterns of coastal marine biodiversity along the Venezuelan coast. This study focused on rocky shores as they are coastal habitats of high ecological importance and wide global distribution. Porifera are of great ecological importance within ecosystems such as reefs and mangroves, and very few studies on this group have been done on rocky shores in Venezuela. For this two reasons we thought it was necessary to determine the richness and taxonomic composition of sponges in this habitat along the Venezuelan east-central coast. To estimate the coverage and richness of benthic species, we used a spatial hierarchical design of three nested scales: regional (central and eastern: hundreds of kilometers apart), local (three localities in each region separated by tens of kilometers), and punctual (three sites in each locality: separated by 500-1000 meters). Sampling was conducted within 2008 and 2009. Each site was sampled at two depths: shallow (2-3 m) and deep (5-7 m), using 1 m^2 quadrats. Between both depths, the rocky subtidal assemblages of sponges represented 5.37% of the total coverage in 2008 and 3.72% in 2009. In both years of sampling, the eastern region had the highest percent cover of sponges (6.12% and 6.83%, respectively) while the central region had the lowest cover of sponges (4.75% and 2%). In terms of richness, a total of 51 species of Porifera, all belonging to the class Demospongiae, were distributed in 11 orders, 27 families and 32 genera. Four of these Porifera are new records for Venezuela: *Tedania* aff. *klausi*, *Mycale* (*Aegogropila*) *arndti*, *Aaptos duchassaingi* and *Monanchora arbuscula*. The species composition varied substantially among sites in each locality (12% dissimilarity), but the rate of species similarity between localities and regions was very high (95-99%). This work is part of the first efforts to understand the distribution and taxonomic characterization of Porifera within the benthic assemblages associated with rocky bottoms in Venezuela. It is important to continue with this type of work and to expand to other ecosystems to better understand the taxonomic composition of Porifera within various habitat.

The current state of sponge fishing in the Republic of Croatia

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Sponge fishing is a century-old tradition in Croatia and represents cultural and historical heritage. Primarily it was a trademark of the island of Krapanj in Sibenik's surrounding. Today fishing grounds that are exploited by Croatian sponge divers include locations in the North and Middle Adriatic and partly in the South Adriatic. Refined natural bath sponges originating from the Mediterranean are still considered to be valuable products in world markets. However, since they are a sensitive natural resource their exploitation should be properly managed. For that purpose we need to know more about the technology of the fishing, fishing grounds, quality and quantity of the catch. Although not many people in Croatia are involved in the business, recent official statistics are not properly kept so it is almost impossible to assess the real amount of sponge catch per year. Also, it is not known which species of bath sponges are targeted, proportions of their catch and habitats affected. In order to answer these questions we made a research consisting of: 1. survey among sponge fishermen through a structured questionnaire and 2. sampling of fresh and dry catch for determination of species and biometry of specimens. We also took into consideration the official statistics despite their incompleteness. In this contribution the main results of survey among fishermen as well as qualitative and quantitative estimation of sponge catch in Croatia are discussed. *Spongia officinalis* makes the biggest proportion of the catch. Almost all the catch is exported to other countries in a dry, non-refined state so the traceability of this Croatian product from the Adriatic Sea is practically lost.

Boring and insinuating sponges in stylasterids (Cnidaria: Hydrozoa) from Indian Ocean

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Numerous sponges live in association with the skeletal structures of calcareous corals as precious corals (*Corallium, Paracorallium*), scleractinians, helioporids or milleporids. Several of these sponges are able to bore the calcareous substrata, while other species insinuate into the excavations produced and after left vacant by the boring sponges. The calcareous coenosteum of stylasterid corals is very compact, composed by calcite or aragonite crystals and it is perforated by a densely anastomosed three-dimensional canal network hosting the living tissue. To date only two boring species have been described in association with stylasterid hydrozoans. *Alectona microspiculata* Bavestrello et al., 1998 was found in the inner part of the stem of *Distichopora* sp. and an unidentified hadromerid sponge was recorded in the basal portion of a colony of *Errina dabneyi* (Pourtalès, 1871). This work provides for the first time a list of boring and insinuating sponges associated with some species of *Distichopora* from Indian Ocean. The observation of 77 colonies of *Distichopora* spp., revealed the presence 22 sponges species into the basal portion of 15 colonies. The identified species belong to 11 genera and include 14 boring and 8 insinuating species. The recorded boring species belong to *Cliona*, *Pione*, *Aka*, *Cliothosa* and *Spiroxya* genera, while the insinuating ones belong to *Acarnus*, *Jaspis*, *Geodia*, *Plakortis*, *Crella*, *Amorphinopsis*. In a single colony of *Distichopora* it was possible to find up to 6 different sponges. It is generally recognised that boring sponges are able to colonize exclusively portions of corals lacking of living tissue but *Aka* spp. produce a secondary metabolite that kills the surrounding host tissue. The basal portions of the studied *Distichopora* colonies were often died, covered by algae and bryozoan and usually the sponge colonization is limited to this area. Nevertheless the colonies associated with *Aka* spp. are the only ones bored also in the living portions where the sponge directly interacts with the stolon network of the stylasterid.

High levels of undescribed species diversity in encrusting sponges confirmed by LSU ribosomal DNA sequences - the genus *Eurypon*.

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Intensive collecting of encrusting sponges by SCUBA diving around Ireland and Britain has resulted in the discovery of a large number of undescribed species which should belong to the genus *Eurypon* (Raspailiidae). These species often co-occur on slightly silted rock faces and have different colours and surface appearances, but similar spiculation and simple skeletons. Comparison of LSU ribosomal sequence data from these specimens has confirmed the species level status of closely allied taxa and suggests that the genus *Eurypon* is polyphyletic with respect to other Raspailiidae. It is suggested that in the Raspailiidae the transition from encrusting to erect growth forms and vice-versa has occurred on multiple occasions, and that similar skeletal architecture is frequently a convergent character and of limited phylogenetic value.

Characterization of a new pyoverdin from a sponge of the Portuguese coast with unique characteristics

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Sponge-associated bacteria have been an important topic of research in the past few years. These studies have contributed to the fundamental knowledge of Marine Biology and Chemistry, and allowed the identification of inumerous new natural products with potential technology and health related applications. In the present work, we report on the identification of a bacteria of the genus *Pseudomonas* associated with the sponge complex *Cliona viridis* collected on the Portuguese coast. The bacterial strain, currently designated NB3, was isolated and cultured in laboratory, and the 16S RNA was extracted and sequenced. NB3 produces the typical fluorescence of several *Pseudomonas* species. The molecule responsible for the fluorescence was purified, and structural determination is undergoing. Taking into account results from amino acid sequence analysis, FT-IR and NMR spectroscopies, and mass spectrometry methods, the molecule was identified as a pyoverdin, also known as bactopecton. This is a family of siderophores that may also be involved in intercellular communication. This molecule has apparently chemical and photophysical properties that are unique among pyoverdins. Some of these features include a very high hydrophilicity, and stability in solution, the absence of a pronounced photobleaching under continuous illumination, and the precipitation at basic pH. The molar absorption coefficient, position of the fluorescence spectra, the quantum yield and fluorescence lifetime are pH and metal ion dependent. Further studies of the strain NB3 and of its pyoverdin, including fluorescence properties and function will, hopefully, shed light into novel roles of sponge-bacteria association in ecosystems.

New species of *Corvoheteromeyenia* Ezcurra de Drago, 1979 (Spongillidae/Porifera) in the São Francisco Basin (Brazil)

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Corvoheteromeyenia Ezcurra de Drago, 1979 is exclusively known to Neotropical Region with two species recorded: *C. australis* (Bonetto and Ezcurra de Drago, 1966) and *C. heterosclera* (Ezcurra de Drago, 1974), the former reported to Argentina and Southern Brazil, the latter to Venezuela, Curaçao, Argentina (Parana Basin) and Northern Brazil (States of Maranhão and Rio Grande do Norte). The genus is characterized by gemmoscleres birotulates insert radially on gemmule, megascleres exclusively oxeas, can present sparsely microspined. Microscleres pseudorotule, exceptionally present in two distinct series. Based on the material collected in São Francisco basin, we characterize a new species to the genus. The material was collected in cages fish farming from Paulo Afonso municipality, State of Bahia, in the shores of São Francisco River. The specimens was removed with a spatula and fixed in ethanol. In laboratory slides of spicules and gemmules were done to identifying proceeding. *Corvoheteromeyenia* sp.nov. is characterized by presents gemmoscleres acanthostrongyles with tuberculate spines differing from the other species of genus that present birotules with spined axis. The shape this sponge ranges from incrustant to slightly massive, do not exceeding 1 cm of thickness. The color was dark green *in vivo* and light green in ethanol. Its megascleres are fusiform, slightly curved oxeas, with surface can be smooth or with sparse microspinations. The microscleres have very varied shape with two types of pseudobirotules. The Type 1 is the most common shape, which has three large spines forming rotules, with a short axis can be smooth or spined, and the Type 2, the most rare shape, is longer than the first which has more discreet spines forming rotules; these present spines distributed equitably along of axis. Gemmoscleres inserted radially on the wall of gemmules, acanthostrongyles, with tuberculate spines with only one category. Gemmules normally located in the basal portion of sponge. Acanthostrongyles gemmoscleres with tuberculate spines were not founded neither species of *Corvoheteromeyenia* nor another freshwater species, what motived us to propose a new species. Notwithstanding a similarity of the other spicular types founded only in species of *Corvoheteromeyenia* allowed allocate this species in the genus. This species can suggest that birotulate gemmoscleres have arisen at various times in the freshwater sponges or still the same was an intermediate stage between those with acanthostrongyle and birotulate gemmoscleres.

A new species of *Radiospongilla* Penney and Racek, 1968 from brazilian inland waters

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The genus *Radiospongilla* Penney and Racek, 1968 has a cosmopolitan distribution, being that to Neotropical Region only *R. crateriformis* (Potts, 1882) and *R. amazonensis* Volkmer-Ribeiro and Maciel, 1983 are known, the latter with restricted distribution to Brazil. *R. amazonensis* is widely distributed in Brazil being reported in 12 of 26 states and seven of 12 brazilian hydrographic basins. This wide distribution can be associated to a cryptic species complex. Despite of Brazil have a large diversity of freshwater sponges, to the State of Pernambuco did not exist records yet. From material collected in the Rio do Prata basin we identified specimens of genus *Radiospongilla*. The material was collected in channels and cages of Aquaculture Station in the Universidade Federal Rural de Pernambuco, Recife, State of Pernambuco, Brazil. The specimens were collected with knife and fixed in ethanol. All material was housed in the Porifera collections of Universidade Federal de Pernambuco. *Radiospongilla* sp. nov. is characterized by the presence of megascleres oxeas (200-266.9-300/9-11.9-20 µm) normally straight and sparsely microspined; microscleres absent. The gemmoscleres are acanthostrongyles straight (57-67.7-78/3-3.1-5 µm) with large abundance of spines distributed homogeneously in the axis with a spine at the tips. The gemmules often scattered throughout the sponge, occasionally observed at the base. Sponge ranging from incrustant to massive with sparse, minute, cylindrincal projections; the color is beige *in vivo* and brown in the ethanol; the consistence is soft. Only two species were recorded to Neotropical Region, which will compare with *Radiospongilla* sp. nov. When compared the collected material from Recife (PE) with holotype from *R. amazonensis* (MNRJ 0088), we constated this specimens present significative morphological differences that justify the proposition for a new species. The external morphology of these species is completely different. In *Radiospongilla* sp. nov. the specimens possess a large abundance of spongin, giving them a soft texture. In other hand, *R. amazonensis* possess scarse spongin. The megascleres of *R. amazonensis* are predominantly acanthostrongyles and strongyloxeas, however in *Radiospongilla* sp. nov. the megascleres are exclusively oxeas. The gemmoscleres of *R. amazonensis* are acanthostrongyles halter-like, due to forming distinct spine aggregations at the tips, which differs from the gemmoscleres of *Radiospongilla* sp. nov. by its homogeneous distribution of spines along axis. *R. crateriformis* is the other species with biogeography affinity which has a cosmopolitan distribution, and also was recorded to Neotropical Region (West Indies). In spite of the megascleres morphology resemble the *R. crateriformis*, these are slightly bigger than new species (390 vs. 300 µm). In opposite, when we compared the gemmoscleres in *R. crateriformis*, these differ mainly in morphology, where as the former presents gemmoscleres pseudorotules in opposition to gemmoscleres acanthoestrongyles of *Radiospongilla* sp. nov. There is a difference in the dimensions these spicules once *R. crateriformis* has 69-148 µm vs 57-78 µm from the new species.

Eocene representatives of *Stoeba*-like demosponges (Astrophorida, Pachastrellidae) from SW Australia and their evolutionary significance

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Completely preserved *Stoeba*-like sponges have been collected from the Late Eocene (about 35 millions years old) spongiolithic rocks of SW Australia (Fitzgerald River National Park area) where they are associated with a rich fauna of lithistid sponges. Other soft bodied demosponges are also common but represented mostly by disassociated spicules. As is typical with fossil material, no microscleres belonging to these *Stoeba*-like specimens are associated with body fossils. A choanosomal skeleton that consists exclusively of short shafted dichotriaenes suggests that the investigated sponges are closely related/may belong to the Recent genus *Stoeba* Sollas, 1888. At least 3-5 species of this sponge could be differentiated, based on considerable differences in morphology, from branching to encrusting, and all free-living in habit. In all specimens the choanosomal spicules are very similar in shape but differ in size and robustness. These differ from Recent representatives of the genus *Stoeba*, that have dichotriaenes or triaenes free in the choanosome, by having the dichotriaenes loosely articulated with their cladomes. This feature makes them intermediate between typical desmas of lithistid sponges and triaenes of nonlithistd demosponges. The fact that choanosomal spicules are loosely articulated, has allowed the intact preservation of the sponge body. The Recent genus *Stoeba* Sollas, 1888 (Demospongiae; Astrophorida: Pachastrellidae) is represented by at least 12 living species known from tropical to warm temperate waters around the world[1]. No fossil representatives of this genus have been known so far. The only known recent Australian species is *Stoeba occulta* (Hentschel) but it differs from the fossil sponges described here in having loose regular dichotriaenes as choanosomal spicules and exhibiting, typical of the genus, a cavity-filling habit. The presence of these fossil sponges in the late Eocene deposits of SW Australia indicates a longer evolutionary history pattern of the pachastrellid sponges than generally accepted.

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The lithistid fauna of Western Australia: general characteristics and distribution.

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Very little is known about the lithistid fauna of Western Australia. The first and only prior report on the group (by Lendenfeld in 1907) described two new species of *Theonella* (*T. levior* and *T. discifera*) collected on the Gazelle expedition to the Dampier Archipelago in 1888–89. Commencing in 1997 a series of surveys encompassing a wide range of latitudes (35.5°N;12.4°S) and depths (intertidal to 1600 m) have been undertaken off the coast of Western Australia. We investigated the diversity of lithistids collected during these surveys and assessed their occurrence against physical and oceanographic features of the Western Australian coast. We estimate this fauna includes 46 species from 18 genera and 10 families. Although southern latitudes were sampled, living lithistids were only found from the Perth Canyon (~32°S) northwards. Two dead lithistids (lacking living tissue) were collected from stations at ~35°S, which suggests that lithistids have occurred in more southern localities. Only 7 species were found from Perth Canyon to the Ningaloo region (23°S); 2 in the Perth Canyon, 1 at Two Rocks (~32°S), 1 off Jurien and 2 off the Houtman Abrolhos (~30°S), and 1 off Kalbarri (~28°S). The other 39 species (and 11 dead specimens) were obtained from the tropical region of Western Australia (north of 23oS). In some tropical regions and depths lithistids were a significant component of the sponge faunal biomass. Most of the species (85%) occurred at depths greater than 50 m. Several species (7%) were found at depths less than 50 m, including 5 species of the family Theonellidae and 2 species of the genus Microscleroderma (Scleritodermidae). The shallowest lithistid was a theonellid, which was collected at 8 m depth in the Dampier Archipelago (~21°S). The deepest lithistids were a *Callipelta* species (436 m), a *Corallistes* species (400 m) and a *Herengeria* species (480 m). Species of the family Theonellidae dominated the lithistid fauna, followed by the Scleritodermidae (8 species), the Azoriciidae (6 species) and the Siphoniidae (4 species). These families all occurred at relatively shallow depths (Theonellidae < 100 m; Scleritodermidae < 200 m; Azoriciidae < 300 m depth; and Siphoniidae < 140 m). A monographic description of the lithistid fauna of Western Australia is ongoing.

Abundance, size distribution patterns and regeneration rates of the Indonesian giant barrel sponge *Xestospongia testudinaria*

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In addition to being some of the most conspicuous and charismatic invertebrates on coral reefs, barrel sponges fulfil a number of important functional roles, filtering large volumes of water, contributing to habitat complexity and as spatial competitors. For example, in the Caribbean they can occupy over 9% of available substrate. Recent studies have shed light on a number of aspects of barrel sponge ecology, but much of this work has focused on the related, but chemically different, Caribbean giant barrel sponge, *Xestospongia muta*. Here we present data on the abundance, size distributions and regeneration rates of *Xestospongia testudinaria* on Indonesian reefs. This study was carried out at two sites in the Wakatobi Marine National Park, SE Sulawesi. The first site, Sampela, is a degraded shallow fringing reef that experiences high levels of sedimentation and the second, Ridge 1, is located 700 m offshore and has higher coral cover and experiences much lower sedimentation rates. Three 50 m x 5 m transects were surveyed at each study site and the volume of each barrel sponge was calculated from in situ measurements and photographs. In total 70 barrel sponges were surveyed across the two sites. Sponge densities were similar to those previously reported for *Xestospongia testudinaria* on the Great Barrier Reef with 0.132 sponges/m^2 at Sampela (33 sponges in total) and 0.16 sponges/m^2 at Ridge 1 (40 sponges in total). The mean volume of barrel sponges at Sampela was 0.07 $m^3 \pm 0.01$ m^3 (SE) compared with 0.17 $m^3 \pm 0.03$ m^3 (SE) at Ridge 1. During surveys we noticed sponges that barrel sponges were predated upon by a number of fish species, so we also investigated regeneration rates for 19 individuals of *Xestospongia testudinaria* at each study site by monitoring the area and depth of 4 cm^3 holes over 20 days. We found no significant differences in regeneration rates between the study sites, with a mean regeneration rate of 1.01 $mm^3 \pm 0.09$ mm^3 (SE) per day. These results add to a substantial body of evidence that sponges are highly efficient at recovering from damage and to the best of our knowledge is the first time that abundance, size distribution patterns and regeneration rates have been described for this species in Indonesia.

What are the key drivers of spatial variability in Indo-Pacific sponge assemblages?

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Coral reefs across the globe are currently declining due to a plethora of threats. Currently, much research focuses on investigating and mitigating the impacts of these threats on coral reefs, however, the vast majority does not examine its effects on sponges. In order to understand how anthropogenic changes may influence sponge assemblages, it is important to understand the factors driving sponge spatial and temporal variation. Here we focus on the factors driving the spatial distribution patterns of coral reef sponges. The abundance and spatial distribution patterns of sponges are influenced by both physical factors, including water flow and sedimentation, and biological processes such as spatial competition and predation. The relative importance of biological and physical factors in driving sponge abundance patterns is still unclear in many regions, but could have far reaching implications when attempting to predict the impacts of environmental degradation on sponge assemblages. The aim of this study was to quantify a number of key environmental and biological variables across a gradient in habitat quality and use multivariate techniques to determine the most proximal factors structuring sponge abundance and diversity in the Wakatobi Marine National Park (WMNP) in SE Sulawesi, Indonesia. Surveys were conducted at ten sites at a depth of 10-12 m. The abundance and diversity of sponges was quantified at each site using a divided 0.5 x 0.5 m^2 quadrat. The environmental and physical variables measured were sedimentation, temperature, salinity, turbidity, chlorophyll-a concentration, substrate angle, nutrient levels, flow rate and habitat complexity. We also surveyed spongivorous fish densities and the percentage cover of other major groups of benthic competitors. This presentation will describe the results of this survey, and we discuss which biological and physical factors are most important in structuring sponge assemblages in the WMNP, a key step in predicting the impacts of habitat degradation on sponge assemblages and developing sound reef management practices in this region.

Parazoanthus axinellae epibiosis. *Axinella damicornis* or *Axinella verrucosa*? This is the dilemma

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Epibiosis is a common strategy among benthic organism, increasing the possibility to occupy both hard and soft substrates. Sponges have a huge spectrum of epibionts everywhere in the world and in the Mediterranean Sea the association between *Axinella* species and *Parazoanthus axinellae* can be considered the commonest. *P. axinellae* can live both on rocky and biological substratum such as several sponges or naked axes of sea-fans. Anyway, as the specific name suggests, *Axinella* species represent its typical living substratum and among *Axinella* species *A. damicornis* and *A. verrucosa* are the commonest choice. Respect to other sponge-zoanthids associations, here *P. axinellae* settles on the sponge ectosome developing encrusting colonies that can be easily detached from its living substratum. We evaluate the differences in the frequency of *P. axinellae* colonies on *A. damicornis* and *A. verrucosa*, comparing the percentage of covering of the epibiont on the basibionts at three different depths (35, 25, and 15 m) along the Portofino Promontory (Ligurian Sea, Italy). Moreover, the population structure of these two sponge species was recorded by 50 x 50 cm^2 frames. *A. damicornis* has generally a bushy shape and never exceed 6 cm in height while *A. verrucosa* develops branching colonies that oaccasionally can be 15 cm tall. The density of *A. damicornis* and *A. verrucosa* is similar at all considered depths (F= 2.79, P=0.11). Regarding the frequency of sponges covered by *P. axinellae* we evaluated that for *A. verrucosa* the highest covering frequency is in the medium class height (4-5.9 cm). For *A. damicornis* the covering is more frequent in the 2-3.9 cm class height. Comparing dimensions of covered and uncovered sponges, at 15 m depth *A. damicornis* is bigger when covered. In depth this response to epibiosis disappears. *A. verrucosa* is affected by the occurrence of *P. axinellae* colonies in the same way but only at 35 and 25 m depth, this response to epibiosis disappear in shallow habitats. An hypothesis is that sponges, to keep an efficient ratio between volume and inhalant surface, are stimulated to increase their final size when covered by *P. axinellae*. *P. axinellae* grows preferentially on *A. verrucosa* than *A. damicornis* at all depths ($P < 0.05$). Patterns of epibiosis are here interpreted under the light of morphological features, suggesting that this association may be regulated by physical more than chemical cues.

Bacterial antagonisms within the microbial compartment of the temperate calcareous sponge *Leuconia johnstoni*

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In our ongoing program devoted to elucidate the role of microorganisms associated to marine sponges, we are involved in studying chemical mediators of bacterial antagonisms. *Leuconia johnstoni* Carter 1871, collected off Concarneau (Brittany, France) is a temperate calcareous sponge (Calcaronea, Baeriida) which dichloromethane crude extract exhibits a persistent annual antimicrobial activity against the Gram positive strain *Staphylococcus aureus*. We showed by fractionation of dissociated sponge suspension that the microbial compartment of the sponge was responsible of this activity. Ultrastructural analysis and fluorescent *in situ* hybridization revealed diverse bacterial morphotypes and phylotypes in the mesohyl of this sponge. In order to determine whether the antimicrobial activity in *L. johnstoni* crude extract could result from the diversity of antagonistic interactions within the sponge ecosystem, we used a culture-dependant method to study the associated cultivable bacterial community. From a series of sponge samples collected from 2005 to 2009 we demonstrated that the cultivable microbiota was composed mostly of gamma- and alpha-Proteobacteria as well as a few Firmicutes, Actinobacteria and Bacteroidetes. About 15% percent of these cultivable associated bacteria had antimicrobial activity against the reference strain *S. aureus*. Among numerous bacterial antagonisms within the sponge *L. johnstoni*, bacterial interactions between the *Microbulbifer* sp. L4-n2 strain (γ-Proteobacteria) and the *Bacillus* sp. L6-3e strain (Firmicutes) were particularly studied. Nine esters of para-hydroxybenzoic acid isolated from *Microbulbifer* sp. L4-n2 were identified as responsible of the anti-*S. aureus* activity. In addition, these compounds also proved to be inhibitors of the bacterial strain *Bacillus* sp. L6-3e growth. We confirmed their *in situ* production within the sponge host by LC/MS experiments through the detection of their chemical fingerprint in sponge extracts. Furthermore, we demonstrated that exposure of the bacterial stain *Microbulbifer* sp. L4-n2 to extracts of the competing bacteria *Bacillus* sp. L6-3e, increased the production of para-hydroxybenzoic acid esters. Interestingly, *Bacillus* sp. L6-3e, which is inhibited by various native sponge strains, was also shown to biosynthesize antioxidant compounds and diketopiperazines which might be implicated in the quorum sensing, in order to maintain its population within the sponge host. In conclusion, bacterial metabolites which are implicated in the competition between bacterial populations in the sponge were shown to contribute to the sponge antimicrobial activity.

New data about demosponge diversity in the Adriatic Sea

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The Adriatic Sea can be considered as a large, mostly shallow embayment of the Mediterranean Sea. The longitudinal SE-NW position of the Adriatic results in considerable climate differences along it. When combined with other factors three ecologically distinct areas can be distinguished: the North, Middle and South Adriatic. The North Adriatic is the northernmost part of the Mediterranean, it is the shallowest part of the Adriatic and it is under the strong influence of the river Po inflow. The South Adriatic is the deepest part of the Adriatic and it is under the strong influence of the incoming warm current from the south. The Middle Adriatic can be considered as a transition zone between these two extremes. Most of the Adriatic shelf was submerged recently, after the last glaciation, during the last 18,000 years when the Adriatic Sea level rose for approximately hundred meters. In this work we compiled the list of demosponges recorded in the Adriatic Sea up to now: we performed a thorough literature check and we added results of our decade long research. In order to check if there is any preference among sponge species towards certain parts of the Adriatic all the records were assigned to the North, Middle or South Adriatic according to their geographic location. In the contribution we discuss the results of the statistical analysis. Altogether 266 species of demosponges have been recorded up to now in the whole Adriatic (our findings and literature survey). Among the 124 species that we found so far in our research 11 are new records for the Adriatic Sea (5 of which were noted only in caves). These species are: *Penares candidata* (Schmidt, 1868), *Timea geministellata* Pulitzer-Finali, 1978, *Discodermia polydiscus* (Bowerbank, 1869), *Rhabderemia spinosa* Topsent, 1896, *Hymedesmia* (*Stylopus*) *coriacea* (Fristedt, 1885), *Phorbas dives* (Topsent, 1891), *Myxilla* (*Myxilla*) *macrosigma* Boury-Esnault, 1971, *Axinella vaceleti* Pansini, 1984, *Dendroxea lenis* (Topsent, 1892), *Pleraplysilla spinifera* (Schulze, 1878) and *Hexadella racovitzai* Topsent, 1896.

Partner-derived chemical defense in a two-sponge symbiosis: the challenge of identifying costs and benefits when multiple partners interact

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The ecological significance of positive interactions (e.g. mutualisms) in marine communities has drawn increased attention over the last decade. In the Florida Keys, a symbiosis between two sponges, *Geodia gibberosa* and *Amphimedon erina*, provides a model system to examine important ecological aspects of beneficial interactions between species. This system also permits exploration factors that may influence partner specificity as several species of sponge may be involved in this association to greater of lesser degrees. We used a series of field and laboratory experiments to test the hypothesis that the symbiotic relationship between *A. erina* and *G. gibberosa* represents a defensive mutualism, with chemically defended *A. erina* providing palatable *G. gibberosa* protection from predators. In a series of tests using artificial foods containing crude chemical extracts, we found that spongivorous starfish and crabs, and a suite of potential predators in the field, avoided feeding on foods containing *A. erina* extracts. We also conducted several field-based experiments with intact and manipulated two-sponge symbioses, and found that removal of *A. erina* resulted in tissue loss from predation for the internal *G. gibberosa*. *Echinaster* seastars appear to be the major consumer of *G. gibberosa* in the habitats we worked in, and these predators have a negative response to several of the sponges that can be found on the surface of the host *Geodia* sponge. The combined data support the hypothesis that *A. erina* (and other symbionts) effectively deters predators of *G. gibberosa*, and thus represents an acquired chemical defense. Chemically defended sponges are commonly involved in associational defenses with a variety of organisms, such as mangroves and hermit crabs, but this sponge-sponge interaction meets criteria to be considered a defensive mutualism. The association provides unique opportunities to discover forces that drive species into more specialized, and thus narrower, niches.

First record of calcareous sponges from bathyal and abyssal depths in the Antarctic Ocean

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Calcareous sponges have traditionally been regarded as shallow water organisms, a persistent myth created by Hentschel[1], partly supported by the problematic question of calcareous skeletal secretion under high partial CO_2-pressure below the CCD in the abyss. Up to now, only few species of the sponge class Calcarea have been described from depths below 2000 m world-wide. From the Antarctic Ocean, by far the largest number of records of Calcarea is known from shelf areas between 50 and 400 m depth. They have only been sporadically recorded on the lower shelf and the upper slope at depths between 570 and 850 m. From depths below 1000 m in the Antarctic there are no previous records of calcareous sponges. It was therefore a big surprise when the first true deep-sea Calcarea from the Antarctic Ocean were collected at depths between 1120 m and 4400 m during the ANDEEP I, II and III expeditions[2,3]. All together, 5 calcarean species and some indeterminable fragments were collected from 5 different stations, and all except 1 species were single individuals. 3 of these species were new to science and 4 have never been recorded in the Antarctic before. The three new species represent the genera *Ascaltis*, *Clathrina* and *Leucetta*[4]. The 5 Calcarea species belong to 5 different genera and 5 families; they make up c. 7% of the total 76 sponge species, but only 1.6% of the about 500 sponge specimens collected in total during ANDEEP I-III. Taken the very patchy and isolated collections in the deep Weddell Sea, any generalization concerning the distribution of Antarctic deep-sea Calcarea should be done with caution. However, although calcareous sponges are rare in the Antarctic deep sea they seem to constitute a constant component of the fauna, as they turned up at every ANDEEP expedition. Therefore we assume that high proportion of calcarean sponge species new to science is still to be expected from the Antarctic deep-sea.

[1]Hentschel E, 1923/1925. Porifera. In Kükenthal W (ed): Handbuch der Zoologie, Band I. pp 307- 418; [2]Janussen D, 2003. Berichte zur Polar-und Meeresforschung 470, 104-108; [3]Janussen D, 2006. Berichte zur Polar-und Meeresforschung 533, 174–178; [4]Rapp HT, Janussen D, Tendal OS. Deep-Sea Research, ser. II, CAML Special Issue (in press)

New reports of freshwater sponges from British Columbia, western Canada

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The province of British Columbia (BC), Canada, is a large, mostly temperate land mass, ca 945,000 sq km, exceeding all but two European countries (Russia and Greenland), but its freshwater sponge fauna is very poorly known. In the scanty and scattered published literature, documentation of only 3 species, *Ephydatia muelleri* (Lieberkühn), *Eunapius fragilis* (Leidy), and *Spongilla lacustris* (Linnaeus), all very common North American (NAm) forms, from only 10 locations can be confidently accepted from 8 original reports, beginning with Bowerbank, 1863. Here I summarize new species location reports from BC, adding occurrence of four additional species (133% increase) and 23 new locations (233% increase) to the known freshwater sponge data for the province. New occurrences, 1 new location each, include *Anheteromeyenia argyrosperma* (Potts), *Heteromeyenia baileyi* (Bowerbank), *Heteromeyenia tentasperma* (Potts) and *Trochospongilla pennsylvanica* (Potts). These are all common or rare (*H. tentasperma*) species distributed from Ontario-Minnesota-Wisconsin-Illinois to the east. The first species, *A. argyrosperma*, has been listed only once as occurring in western NAm (California) in an unconfirmed report (no details given) by Neidhofer (1940). The others, *H. baileyi*, *H. tentasperma* and *T. pennsylvanica*, have never been reported from western NAm, although *H. baileyi* does extend towards the west in its southern range (101.5°W in Mexico). It has been postulated previously, on the basis of overall reports, that freshwater sponges occur less frequently and have lower diversity in western NAm. My collections, based upon ad hoc comparison of results per unit effort in eastern and western regions support this generalization. Since, however, there has been a very low search effort carried out in western NAm, it is likely that wider surveys will uncover a much greater freshwater sponge species diversity in this region than previously expected.

Taxonomic revision of Brazilian *Tethya* (Porifera: Hadromerida) with description of four new species

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The taxonomy of the genus *Tethya* from the Brazilian coast is revised. Five species are described, four of which are new to science: *Tethya maza* Selenka, *T. beatrizae* sp. nov., *T. nicoleae* sp. nov., *T. parvula* sp. nov., and *T. solangeae* sp. nov. Five earlier records of *Tethya* from Brazil (*T. aurantium, T. diploderma, T. maza, T. japonica, and T. seychellensis*) are re-evaluated and only *T. maza* is confirmed. The other previous records of *Tethya* from Brazil, viz., *Tethya brasiliana, T. cyanae, T. ignis and T. rubra*, have been recently described in detail and are considered valid without need of redescription. An ammendement to the nomenclature of micrasters is made, recognizing at least two different morphologies of each type of micraster (strongylasters types 1 and 2, tylasters types 1 and 2, and oxyasters types 1, 2 and 3). *Tethya diploderma* from the Gulf of Mexico is reexamined and presents four types of micrasters: strongylasters type 2 (spined only at the tips), tylasters type 2 (rays cylindrical with coarse spines at the tip), oxyasters type 2 (smooth), and oxyasters type 3 (with large center and conical rays with spined tips). *Tethya maza* also has four types of micrasters, subtly different from those of *T. diploderma*: strongylasters type 1 (spined all along the rays), tylasters type 1 (short rays, fully spined), oxyasters type 1 (spined), and oxyasters type 2. *Tethya beatrizae* sp. nov. can be distinguished by its three types of micrasters: strongylasters type 2, oxyasters type 1, and oxyasters type 2, all in low size range (11–15 µm in diameter). *Tethya nicoleae* sp. nov. also has three types of micrasters: strongylasters type 2, tylasters type 2, and oxyasters type 2. *Tethya parvula* sp. nov. has a thin cortex, and its only micrasters are strongylasters type 2. *Tethya solangeae* sp. nov. is distinguished by a firm consistency and choanosomal lacunae; its micrasters are strongylasters type 2 and oxyasters type 2. Neotypes were designated for *Tethya maza* and *T. diploderma*, which lacked type specimens. Four new species were described here and four previous records were considered invalid; therefore, the known diversity of *Tethya* in Brazil still remains in nine species. However, its rate of endemism increased from 44% to 89%.

A comparison of chemical profiles of buds and adults of *Tethya maza*

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Antipredation activity of secondary metabolites is well known in several marine organisms such as sponges, corals, algae, mollusks, etc., and this defensive activity represents ecological advantage in the establishment of such organisms. Currently, it is known that several structures of propagation are also chemically defended (e.g. zygotes of macroalgae, larvae of many invertebrates including sponges and eggs of nudibranchs). Sponges reproduction may involve sexual and asexual processes. In the sponge *Tethya maza*, asexual reproduction occurs through buds that grow up beyond the surface of the sponge. This species, abundant in the intertidal zone, has active secondary metabolites against predation by hermit crabs. However, the chemical composition of buds are not known. The aims of this study were to analyze the chemical profile of buds and adults of *T. maza* and to compare them. Specimens of *T. maza* were collected in Paraty (Rio de Janeiro State) through free diving. In laboratory, buds were carefully separated from adult, stored, lyophilized and extracted with acetone. Crude extracts were analyzed by gas chromatography-mass spectrometry (GC-MS). The most abundant compounds found both in buds and adults were cholesterol, ergosta-5,22-dienol-3 and stigmasta-5,24-dienol-3. There is a high probability that chemical defenses are present in buds due to their resemblance to the profile of adults examined.

Antifouling potential of tropical marine sponges

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Benthic marine organisms are constantly exposed to settlement of larvae, propagules and microorganisms on their surface. Secondary metabolites may act preventing or reducing this kind of interaction, what represents a major ecological advantage for sessile organisms. Several studies showed the high antifouling potential of metabolites from sponges around the world (e.g. Caribbean, Mediterranean, Indian and Pacific). In Brazil, until the present only three sponge species had their antifouling properties studied: *Aplysina fulva*, *Mycale microsigmatosa* and *Geodia corticostylifera*. Among these, *Geodia corticostylifera* showed strong antifouling activity preventing the establishment of the bivalve *Perna perna* in laboratory tests and *Mycale microsigmatosa* selectively inhibited the establishment of barnacles in field experiments. Considering the existence of more than 300 species of marine sponges along the Brazilian coast, there is still much to be investigated. The objective of this study was to test crude extracts of ten abundant species of sponges from Brazil against the settlement of the mussel *Perna perna* through laboratorial assays. The sponges *Petromica citrina, Desmapsamma anchorata, Polymastia janeirensis, Tedania ignis, Aplysina fulva, Mycale angulosa, Hymeniacidon heliophila, Tethya rubra, Tethya maza* and *Dysidea etheria* were collected in Arraial do Cabo, Angra dos Reis and Paraty (Rio de Janeiro State) and Salvador (Bahia State). In laboratory, organisms were frozen and freeze-dryed before extraction with acetone. The crude extract of three species significantly inhibited the attachment of byssus: *Tethya rubra* ($p = 0.0009$), *Tethya maza* ($p = 0.0039$) and *Hymeniacidon heliophila* ($p = 0.00003$). The existence of antifouling activity in these sponges seems to reflect the evolutionary history of each species, once it is not related to a pattern of taxonomic affinity or geographical distribution. Also for the other species it remains the question of how they avoid predation and competition since the secondary metabolites do not seems to act by inhibiting the settlement of other organisms over them.

A new species of *Isodictya* (Porifera, Demospongiae, Poecilosclerida) from the Antarctic Ocean

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A new Antarctic species of the genus *Isodictya* Bowerbank, 1864 is described. All the specimens were collected in Antarctic waters: three in the Antarctic Peninsula, Marguerite Bay, Low Island, and Deception Island and one in Peter I Island. Its presence in the last one is quite significant as this is 390 km away from the nearest coast in Bellingshausen Sea and very few scientific results have been published about it, as it has been seldom visited by scientific expeditions. However the Bentart 03 Expedition seems to indicate that it has a wide variety of benthonic organisms, in contrast to the deep adjacent areas of Bellingshausen Sea. The material was collected by the Spanish Antarctic expeditions Bentart 03 in February 2003, Ecoquim 06 and Bentart 06 in February 2006. Samples were collected at depths from 97 to 210 m by a Box Corer and a modified Agassiz Trawl between February 2003 and February 2006. *Isodictya* sp. nov. is characterized by its fragile and thin morphology and by the fact that it has microxeas as additional microscleres. It is a filiform sponge, which is fixed to the substratum by means of small roots. Dimensions: 24 cm long and 3 mm diameter in its middle part and 1 cm at the top of the apex. It has a smooth and hard texture, flexible but consistent shaft. At first sight neither oscula nor perforations can be seen. In its upper part, an apical multispicular axis can be made out, with secondary fibres in perpendicular arrangement to this axis. Her colour is beige *in vivo* and white in alcohol, except for its basal area and in its apical area, where it is brown. Choanosomic skeleton made up of bundles of oxeas along the stalk of the sponge. The ectosomic skeleton is made up of oxeas with few spicules, with scattered isochelae that form no defined layer, among which we may also find microxeas. The main spicules are erect or slightly curved oxeas, which end with a very short tip and the microscleres are palmate isochelae and microxeas. This peculiar morphology of the collected specimens is similar to the design of *Isodictya delicata* megachela (Koltun, 1964). In the specimens that is object of study, an additional spicular category has been also observed: the microxeas that are absent in the other species of the genus *Isodictya* of the Antarctic.

This is a contribution to the projects Bentart and Ecoquim.

Preliminary results of sponges collected by Mozambique 0309 expedition in South East Africa (Indian Ocean)

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Mozambique 0309 expedition is included within the East-Centre Atlantic Fisheries Program of Spanish Institute of Oceanography, in cooperation with Spanish Sea General Secretariat and Fisheries Ministry of Mozambique. As part of the results of this expedition, we collected 159 specimens of sponges in 60 tows between 240 and 690 m depth, by means of "Baka trawl" named Conakry in the Mozambique region on board R/V Vizconde de Eza. This communication presents the preliminary results about this phylum. Most of the samples belong to Hexactinellida Schmidt, 1870 (53%) and Demospongiae Sollas, 1886 (47%) classes. The individuals of Hexactinellida class mostly belong to genera *Rossella* Carter, 1872, *Euplectella* Owen, 1841, *Aphrocallistes* Gray, 1858, *Hyalonema* Gray, 1832 and *Myliusia* Gray, 1859. As regards Demospongiae class, the most abundant specimens belong to *Astrophorida* Sollas, 1887 (30%) and *Hadromerida* Topsent, 1894 (21%) orders but we also noted the existence of specimens included in the orders Poecilosclerida Topsent, 1928 (18%); Spirophorida Bergquist and Hogg, 1969 (7%); Halichondrida Gray, 1867 (5%); Haplosclerida Topsent, 1928 (4%) and Lithistida Schmidt, 1870 (1%).

Ecological aspects of sponges in mesophotic coral ecosystems

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Mesophotic Coral Ecosystems (MCE) represent a new realm of unexplored habitats that range from 30-100 m deep. Given the worldwide coral reef decline, MCE research has created great expectations because of their potential as refugia and as a viable source of larvae and nursery for commercial and endangered reef species. Within these deep reef systems sponges play an important ecological role in terms of abundance and richness of species, coupling water column productivity to the benthos and providing rugosity, complexity and refuge for other species. However, information regarding the taxonomic composition and ecology of sponges in MCE is scarce. The main goal of this study was to characterize MCE sponges in five areas of Puerto Rico (La Parguera, Guánica, Vieques, Desecheo and Bajo de Sico). Thirteen (km-long) photo-transects obtained with the Seabed Autonomous Underwater Vehicle (AUV) provided the 1,116 images that were analyzed, covering over 3.7 km², to derive percent cover and species richness, color and morphology. The possible influence of several factors (location, transect, depth, geomorphology, water turbidity, distance from land, level of analysis) in the distribution of MCE communities was statistically evaluated with ANOSIM, SIMPER, NMDS, cluster and regression analyses. Slope is a determinant factor for community composition. The species richness found (77) tend to decrease with increasing depth. Morphology of sponges seems to be related to depth and location, whereas color seems to be related to geomorphology, turbidity and location. Sponges with carotenoid pigmentation are the most abundant and widely distributed. Tube, cup-like and massive forms not only provide rugosity and microhabitats for other organisms, but they may conform the most adaptive morphologies to an optimal water-circulation in MCEs. Branching and erect forms may represent an adaptation to steep slopes, while encrusting morphologies are probably opportunistic.

Exploring the homology and evolution of microRNA biogenesis in Porifera and Eumetazoa

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MicroRNAs(miRNAs) are short, 22 nucleotide noncoding RNAs that play an important role in the post-transcriptional regulation of eumetazoan messenger RNA, which regulate numerous developmental and cellular processes. miRNAs are transcribed from the genome as single stranded pri-RNA precursors with a stem-loop secondary structure which is cleaved by the Drosha/Pasha enzyme complex 'microprocessor', exported from the nucleus, and further processed by Dicer into the 22nt mature strand which recognizes complementary sequences on the target messenger RNA. In eumetazoans, miRNA genes are acquired over evolutionary time and are rarely secondarily lost, exhibiting a lineage-specific pattern of occurrence which mirrors the hierarchical organization of the eumetaozan phylogeny. miRNA gene acquisition also correlates well with increasing organismal complexity. Demosponges have been found to possess miRNAs, however there are no miRNA genes in common with the eumetazoa and miRNAs are absent in the calcisponges and the placozoans. Demosponge miRNAs also do not share secondary structure of the Eumetazoan pre-miRNA regarding the position of the mature sequence on the stem-loop pre-miRNA. Indeed demosponge pre-miRNAs appear to have a more plant-like secondary structure. This leads to the hypothesis that demosponge miRNAs have an independent evolutionary origin. The key enzyme complex for processing of eumetzoan miRNAs is the microprocessor, consisting of the RNAseIII enzyme Drosha, and the double-stranded RNA binding enzyme Pasha. Several sequence-specific RNA recognition enzymes and helicases also associate with the microprocessor and the pre/pri-miRNA to modify processing in a subset of miRNAs. Previous studies have determined that the demosponge *Amphimedon* possesses the key miRNA enzymes Drosha and Pasha; and that Pasha is missing in placozoans. However it was not shown that the demosponge enzymes were phylogenetically homologous with respect to the Eumetazoan enzymes. In the current study, phylogenetic analysis of the identified *Amphimedon* Drosha and Pasha show that they do appear to be homologous to the eumetazoan enzymes. The homology of demosponge and eumetazoan Drosha and Pasha strongly suggests a common evolutionary origin for metazoan miRNAs. However the constrained secondary structure of the eumetazoan pre-miRNA relative to the more plant-like demosponge miRNA secondary structures, as well as the apparent lack of acquisition of miRNA genes over evolutionary time requires further investigation. The constraint of the eumetazoan miRNA secondary structure is potentially the result of the microprocessor associated sequence-specific RNA recognition enzymes and helicase activities. Further analysis is presented in this poster to show whether *Amphimedon* possesses homologous copies of these enzymes.

Life cycle and larval ultrastructure of two Mediterranean Poecilosclerida sharing habitat

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Hemimycalle columella and *Crella elegans* are poecilosclerid sponges, which share semi-dark habitats in the western Mediterranean sublittoral. Both species show a thick encrusting habit, have a pale orange to rose coloring, and are covered by round densely distributed pore-sieves on the surface. Furthermore, the two species release larvae at the beginning of autumn in the study area. The phenotypic convergence of the genera *Hemimycale* and *Crella* has been responsible for some wrong identification of Pacific and Red Sea species. *H. columella* and *C. elegans* are hermaphroditic and release a typical parenquimella larva in later September-early October. To gain knowledge on the possible interactions of these two species, we investigated their timing of gamete and larva production, their reproductive investment, and the ultrastructural aspects of gametes and larvae for both species. In *C. elegans*, both oocytes and spermatic cysts were produced from Maig to July depending on the specimen, and embryos were incubated from July to september. The life cycle of *H. colummella* is slightly delayed with respect that of *C. elegans*: oocytes were present from July to September, spermatic cysts were abundant in August September, embryos appeared in September and coexisted with mature larvae in October. No reproductive elements were found in any of the two species from November on. Some individuals of both species disapeared after the reproductive period has finished while others experience a resting period likely for the rearrangement of the aquiferous system. Whether individuals in a resistance stage remained in place to develop new adults or dissapear after some months is not resolved yet. The larval ultrastructure shows distinct features in both species, with notably higher collagen density in *C. elegans*.

The elephant ear sponge *Ianthella basta* - Growth and potential of farming for biomedical applications

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The elephant ear sponge *Iantella basta* is a common member of the benthic communities on micronesian reefs, but was apparently introduced on Guam, where its presence is restricted to Apra Harbor. This sponge species is very conspicuous with its bright blue color and fan- to funnel-shaped growth form. What is even more impressive is the large size of older individuals with sizes of 1 m height and diameter not being uncommon and individuals reaching over 2 m in height being recorded in recent surveys (Rohde, unpublished). The tissue of *Ianthella basta* has recently received attention by biotechnological studies, which identified a for sponges unusual chitin skeleton and suggested that this chitin scaffolds may have great potential in tissue engineering and other biomedical applications. However, the use of marine products is often restricted by the supply limitations. To explore whether *I. basta* constitutes a sustainable source for tissue harvest, we determined specific growth rates and growth patterns of the natural population in Guam and tested multiple techniques to grow this species in an aquaculture setting. Sponges are considered to have relatively slow growth rates, but data on sponge growth are very rare and non-existent for the genus *Ianthella*. Experiments determining growth rates over the course of one year revealed a mean specific growth rate of 1.30 ± 1.02 $year^{-1}$ and growth rates decreasing with increasing sponge size. These impressive high growth rates support the establishment of aquaculture for the supply of *I. basta* tissue for biomedical applications. Different options for farming, like initial explant sizes, attachment methods and depth were tested and results will be discussed.

Comparison of tissue preservation methods for total RNA extraction from diverse marine sponges for the Porifera Tree of Life project (PorToL)

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The primary goal of the Porifera Tree of Life Project (PorToL) is to provide a phylogenetic context that will improve scientific understanding of various aspects of sponge biology. For example, resolving the branching order of Poriferan families (Tier 1), reconstructing relationships and character evolution within each major clade, exploring relationships within clades by providing a community sequencing service, and developing an Internet-accessible database (PorToL: The Porifera Tree of Life Project - available at www.PorToL.org) will facilitate the integration of molecular and morphogoical data to resolve longstanding questions on sponge phylogeny. Part of the Tier 1 strategy of PorToL is based on sequencing 7 nuclear genes from mRNA transcripts (that lack intron sequences) (Sperling et al., 2007; See A. Hill et al. poster; Also D. Lavrov on mitochondrial genomes). This approach requires the isolation of high quality RNA that must be converted to cDNA before use in reverse transcription-PCR (RT-PCR) of the nuclear genes with universal or sponge-specific primers. However, the samples available to PorToL from the 120+ potential sponge families can vary in age and tissue quality. Thus this study represents a systematic comparison on the yield of total RNA based on the type of preservation method (frozen, 75% ethanol, Trizol® reagent, RNA Later®, DMSO), and secondly on the taxonomy of the sample. RNA extraction of sponge samples followed the routine Trizol extraction protocol. Results showed that a recently developed preservation protocol of sample immersion into a series of 75% ethanol baths was highly effective and convenient (for field collections) for the preservation of RNA integrity. Not surprisingly, freshly collected samples yielded higher RNA quantities in contrast to samples archived at -20°C for several years. However, the commercial reagent, RNA Later, did exhibit sufficient RNA yields (>100 ng/ul) for cDNA synthesis for some older samples. Another important factor likely affecting RNA yields was the taxonomy of each sponge sample. For example, some deepwater demosponge taxa (Theonellidae) appeared to yield good quality and quantities of RNA across several methods, while by contrast, freshly collected deep water hexactinellid species yielded lower amounts. From these results, it appears that sponge taxonomy remains a difficult predictor for high quality RNA yield due to complex factors that may be involved such as variable environmental conditions, microbial symbiont compositions, and biomass:spicule ratios.

Chemical and microbial investigations of the calcareous Mediterranean sponge *Clathrina clathrus*

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Marine sponges are a rich source of structurally unique natural compounds, several of them displaying a wide variety of biological activities. They are also known to host a wide diversity of micro-organisms and, since several years, these associations raise the question about the origin of the metabolites that sponges produce. However, despite this growing interest in marine symbiotic association research, the complex interactions between sponges and associated microsymbionts remain difficult to investigate. Most symbiotic microorganisms are uncultivable, and thus the putative microbial origin of natural compounds isolated from sponges is often inferred from cellular localization data. Calcispongia (calcareous sponges) are good models for this kind of investigations. Unlike Demospongiae, they have been little studied both in terms of chemistry and microbiology. Their relatively low microbial density and diversity might represent an advantage to better understand the origin of the secondary metabolites they produce and the putative role of their endosymbionts. From crude extracts of the Mediterranean sponge *Clathrina clathrus* (Calcinea, Clathrinida), several compounds were isolated, including two major aminoimidazoles derivatives: the known antifungal clathridine and the new clathridimine which is inhibitor of *Candida albicans* and *Escherichia coli*. Different populations of bacteria have been localized in the mesohyl of *C. clathrus* and their morphology and distribution have been examined in details at the ultrastructural level through electron microscopy. In order to determine which cells are responsible for the production of secondary metabolites, we investigated the cellular localization of these compounds by separating bacterial populations and sponge cells of *C. clathrus* using differential centrifugation. Our first results are discussed with regards to the origin of *C. clathrus* metabolites and the possible association between some sponge cells and some bacterial morphotypes.

Isolation and cultivation of fungal strains from *in vitro* cell cultures of marine sponges

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In the last 20 years, marine sponges have been considered one of the most prolific sources of natural products, with notable chemical diversity. However, many of these substances are produced by associated microorganisms, and not by the sponge itself. Most sponges harbor large numbers of symbionts, sometimes comprising 40-60% of the total biomass and including bacteria, cyanobacteria, unicellular algae, archaea and fungi. In many cases, these communities are species-specific and independent of the environmentally available strains. Despite the more than 800 fungi strains identified from sponge samples, little evidence has been published to support the idea of true sponge-associated symbioses. In addition, sponges are also exposed to large numbers of different microorganisms from the environment, making it difficult to distinguish between true sponge-associated microorganisms and environment-derived contaminants. In the present work, fungal strains were isolated and cultivated directly from *in vitro* sponge cell cultures and from single cells. Twenty-seven fungal strains were isolated from the sponges *Hymeniacidon heliophila* Parker, 1910 (Halichondriidae) and *Haliclona melana* Muricy and Ribeiro, 1999 (Chalinidae). Twelve strains were obtained from *in vitro* cell cultures and considered truly associated with the sponges. The other fifteen strains were isolated from whole tissue and considered transient. Five different strains were obtained from *H. heliophila* isolated cells, while five were identified from cytospins and two from primmorphs of *H. melana*. The fungal strains obtained from *in vitro* cell cultures were different in both sponges and none of them were detected in the whole tissue. Four fungal strains isolated from the cell cultures have been previously found in association with marine sponges. Nevertheless, this is the first report for the other eight strains. Eight *H. heliophila* and six *H. melana* fungal strains belong to potentially new species. The use of isolated cells and *in vitro* cultures eliminates transient microorganisms and allows the initial development of the strains to be monitored, enabling the selection of those truly associated with sponges.

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A molecular and morphometric re-evaluation of Placospongia (Porifera, Demospongiae)

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The genus *Placospongia* is characterised by the presence of selenasters as cortical spicules. According to Systema Porifera, it has 3 valid species (*P. melobesioides*, *P. carinata* and *P. decorticans*) and 6 synonymous species (*P. mixta*, *P. intermedia*, *P. labyrinthica*, *P. cristata*, *P. graeffei*, and *P. anthosigma*). A recent molecular study, based on ribosomal spacer (ITS) sequences, suggested that there might me a large number of cryptic species in the genus, but no concomitant morphological analyses were performed, neither were proposed boundaries between the putative species. To try to address those issues, we have studied worldwide samples of *Placospongia* to verify their specific status, through morphological, morphometric and genetic analyses. Spicules were analysed through optical and scanning electron microscopy, and six molecular markers were analysed: cytochrome oxidase subunit 1 gene (CO1), ribosomal internal transcribed spacers 1 and 2 (ITS1 and 2), and the new mitochondrial markers ATP synthase subunit 6 (ATP6), cytochrome oxidase subunit 2 (CO2), and two spacers delimitated by the ATP6/CO2 genes (SP1) and ND5/12S genes (SP2). Specimens of *P.* aff. *carinata* from 6 localities and specimens of *P.* aff. *melobesioides* from 4 localities, as well as a specimen of *P. decorticans*, and the holotypes of *P. carinata*, *P. intermedia* and *P. mixta* were analysed. *P. carinata* and *P. melobesioides* proved to be species complexes, with three new species in the former and one in the latter. The holotypes of *P. intermedia* and *P. mixta* were genetically very distinct from the other species of the genus, that, and the clear differences in morphology and geographic distribution confirm their specific status. More surprisingly, sequences from the holotype of *P. carinata* (a species diagnosed, among other things by the presence of spirasters) clustered with those from species of the *P. melobesioides* group, composed of sponges without spirasters. A careful analysis of the holotype of *P. carinata* failed to find any spirasters, even though they were depicted in the original description of the species. The common occurrence of both species in sympatry may have been the cause for such confusion, and indicate that a neotype must be designated for the species.

New polymorphic mitochondrial markers for sponge population genetics

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Population genetic studies in the Porifera have been hampered by the lack of variable DNA markers. The traditionally used invertebrate markers, such as the mitochondrial cytochrome oxidase subunit 1 gene (CO1), present low levels of polymorphism in sponges. Therefore, most population genetics and phylogeography studies of sponges have been limited to allozymes, which are problematic by the need of fresh or frozen samples. Some sponge population genetics studies have been based on microsatellites, but their general utility is limited by their species specificity. In the present study four new mitochondrial polymorphic markers were tested in six sponge species that encompass five different orders belonging to the class Demospongiae. Whenever possible, two populations of each species were used in the analyses. Nucleotide and haplotype diversities of these four markers were compared to those of CO1. Two species (*Aplysina fulva* and *Hymeniacidon heliophila*) showed no variation across all tested markers. Additionally, some primer combinations did not work in some species. This last result could be due to differences in gene order and may reflect the immense genetic diversity within demosponge orders. In spite of that, in the other four species studied (*Chondrosia* aff. *reniformis, Placospongia carinata, Cliona delitrix* and *Cinachyrella* sp.) the new mitochondrial markers presented higher levels of variation, sometimes up to 2.3 times higher than those observed for CO1, making them suitable for alpha-level systematics and population genetics studies.

Growth of freshwater sponges, Corvospongilla spp. in the Pong River, northeast Thailand

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Growth of freshwater sponges, *Corvospongilla* sp. 1 and *Corvospongilla* sp. 2 were investigated in the Pong River, northeast Thailand during July 2009 - April 2010. The study site is located at 16°46'20.40"N, 102°42'48.22"E where *C.* sp. 1 is dominant. The large colonies of *C.* sp. 1 and introduced *C.* sp. 2 were cut into small explants with approximately 21 cm^2. Explants of both species were attached to the plastic mesh and settled depth of 1, 2, and 3 meters at different locations as near the river bank and far off the river bank. Seventeen physicochemical parameters of water quality were measured monthly throughout the study period. After six months exposure, survival and growth rates of explants were monitored by SCUBA diving. Photographs of explants were taken and the surface areas of all explants were calculated from digitized images. Growth of explants was determined by comparing the surface area of explants at the beginning and after six months. The result showed that percentage of survival rates of both species was 100. Two-way ANOVA indicated that growth differed significantly between species, location, and their interactions ($p < 0.05$). Increase surface area of *C.* sp. 1 (319.7 cm^2) was higher than *C.* sp. 2 (51.0 cm^2) and growth of both species at the far off river bank (220.9 cm^2) was greater than those of the near river bank location (146.9 cm^2). However, growth of both species did not significantly different between depths. In addition, explants of *C.* sp. 1 began to die in February, 2010 whereas *C.* sp. 2 is still alive. The result of physicochemical parameters to support this situation is ongoing.

Variability of natural products in the sponge *Aplysina aerophoba* over a two-year survey

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Given the huge number of interactions between marine invertebrates, specially between sessile organisms, they are provided with an efficient chemically-mediated system based on bioactive compounds. For that reason, marine invertebrates are a potential source for natural products with pharmaceutical and biotechnological interest. In order to obtain the required amount of those compounds to set up the bioactivity tests and not to threaten species with extinction, it is essential to know the production cycle of natural products and when it is higher. Sponges are one of the most known producers of compounds with bioactive properties. However, changes in its production over time are far to be fully reported. The demosponge *Aplysina aerophoba* is well known to harbour large amounts of brominated alkaloids (BAs) used as a defence system which abundances vary between tissues. This species is used as a model to assess the variability of its chemical production over a 2-year survey in the ectosome and the choanosome of the sponge. Samples were taken monthly from five healthy specimens randomly chosen in Portbou coast (Northwestern Mediterranean). Samples were freeze-dried and 30 mg of each one were extracted with methanol. We used high performance liquid chromatography (HPLC) to obtain the chemical profile of each sample. Four major BAs were identified due to its retention time and UV profile and were quantified according to standard curves. Our results show evidence of a striking variation in compound abundance between tissues. The choanosome was more enriched in BAs than the ectosome. Our results also show a different pattern of BA abundance over time. There seems to be a seasonal regulation of secondary metabolite production, although factors behind this variation are still unknown. Temperature fluctuations all over the year might be one of the factors to be taken into account to understand what is causing those changes. Our findings are a further step in the search for drugs from marine organisms with the purpose of obtaining the required amount of natural products without threatening the marine biodiversity.

Richness and distribution of sponge megabenthos on hard benthic reefs of the Agulhas continental shelf

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The Agulhas Continental Shelf is a broad extension of the South African coastal plain with a roughly triangular shape and a mean depth of just over 100 m. Approximately 60% of the shelf consists of hard substratum, and low/high-profile reefs exist in a number of places, both close inshore and extending offshore south of Cape Agulhas (20°E). Inshore and offshore reefs were sampled from 2008 to 2009 and were found to contain a high diversity of marine sponges, representing a highly unique fauna in the Agulhas biogeographic province. Gradients in species richness and taxonomic composition were not correlated with the distance between reefs or their latitude, but with the spatial heterogeneity of bottom profile and a combined effect of both distance from shore and latitude. Spatial pattern of the sponge assemblages was characterized by (1) high species turnover per depth range within and between reefs, and (2) high levels of reef occupancy in the component species, with most species recorded in low profile/high rugosity reefs. Reef variation in diversity and species composition within or between reefs suggests that biological patterns may be finer-scaled than the spatial resolution of conventional bioregional units. Consequently, from a perspective of conservation planning, a single reef structure on the Agulhas continental shelf is unlikely to accurately represent the regional faunal diversity, considering the high beta diversity and low biotic similarities of communities between the reefs and the limited distributional ranges of the component species.

The study of benthic habitats and biological communities of the deep sea with a new photogrammetric and quantitative method

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Some deep areas of the Cantabrian Sea and off Galicia (North of Spain) were studied in frame of the ECOMARG and INDEMARES projects. The study focused on benthic-demersal ecosystems, with a multidisciplinary approach (www.ecomarg.net, www.indemares.es). In areas where the presence of rocky outcrops hampered the use of classical sampling methods we used a non-intrusive method based on a towed vehicle that collected continuous high definition video and periodic digital still imagery of the seabed along transects. The system was designed by the Spanish Institute of Oceanography (IEO), Santander laboratory, with the aim of sampling deep macro-epibenthic communities quantitatively in a cost-effective manner, in order to eliminate the need for high vessel performance or support from technicians. In each transect, between 1 and 2 km length along the sea floor, the number of pictures ranged between 800 and 1000 and 1-2 hours of video footage were taken. Using an image processor, the scale and surface of each picture was calculated from the four laser point's geometry analysis. The coordinates estimated were used to resampling, surface calculations and for sizing species. These features defined the photogrammetric sledge as a real quantitative sampling system. The coverage of each element (facies, habitats, species, etc.) was also obtained. The classification scheme was built up of different main three information layers: 1) facies; 2) habitat and 3) human impacts. Facies was used to describe the different geological and sedimentological appearance (sand, mud, rocky outcrops, etc.). To define the different habitat we used the EUNIS hierarchical classification. Two sub-classes of anthropogenic impacts were defined: (1) fisheries (lost gillnets, long lines, trawl damages, i.e.) and (2) rubbish. In addition, other environmental characteristics (water temperature and salinity, reflectivity, etc.) were included in the analysis to detect their effects on habitat and communities spatial distribution. The analysis of information provided a detailed habitat mapping and benthic community structure. This new methodology is a valuable tool to locate, characterize and provide high resolution maps of the deep-sea vulnerable habitats. The use of this non-extractive methodology, which does not cause damage or alterations to benthic communities, is particularly necessary when establishing a programme of MPA monitoring using a visual transect, in order to ground-truth the image records by either incorporating information into *in situ* survey data, and studying the effectiveness of closed areas on the recovery of structure-forming invertebrates from disturbance, particularly bottom trawling and gillnets.

Unravelling the moons: taxonomic insights of Tetillidae in Indonesian waters

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Moon sponges include the genera *Paratetilla* and *Cinachyrella*, which are globular sponges that are similar in external appearance with numerous porocalices, resembling the lunar surface. It has long been considered that the two species *Paratetilla bacca* and *Cinachyrella australiensis* are widespread and abundant with a geographic range from the Red Sea to the Central-Pacific in a variety of habitats. These species are almost always present in species lists of surveys from the Indo-West Pacific. However, an examination of up to 200 specimens deemed to be '*Paratetilla bacca*' (Selenka, 1867) and '*Cinachyrella australiensis*' (Carter, 1886) collected in East Kalimantan, Java, Bali, and West Papua in Indonesia as well as in various localities in the West Indo-Pacific such as Taiwan, Singapore and Palau, showed that there was great variation in morphological characters. The variation in morphology was so high that we are certain that this is taxonomically relevant, but we were limited in being able to assign a valid taxonomic name to our specimens. A review of the taxonomic history of this group revealed some ambiguous statements, including incomplete descriptions, intermingled identifications, and tens of different species synonymised. The objective of our study was to provide a detailed taxonomic description of the different morphotypes we identified. In summary, three different *Paratetilla* morphotypes and five *Cinachyrella* morphotypes were recognized. The presence of calthrops-type spicules in *Paratetilla* species is the character distincting them from *Cinachyrella* species. Within the genus *Paratetilla*, ranges in calthrops size, oxeas and microxeas were used to diagnose species; the validity of *P. bacca* is discussed. For *Cinachyrella* species, the presence or absence of acanthose microxeas and siliceous spheres, triaene geometry and size of sigmaspires were used as diagnostic characters for species identification. Based on our observations, we propose to resurrect the species *C. porosa* (Lendenfeld, 1888), *C. anomala* (Dendy, 1905), *C. vaccinata* (Dendy, 1922) and *C. paterifera* Wilson, 1925. Illustrations and a taxonomic key for the eight species will be presented.

A new species of Jaspis Gray, 1867 (Porifera, Demospongiae, Ancorinidae) from the western Atlantic

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The genus *Jaspis* Gray, 1867, of the Demospongiae family Ancorinidae Schmidt, 1870, comprises encrusting or massive sponges without triaenes and characterized by oxeas and euasters without a marked centrum (Hooper & Van Soest, 2002). The genus is currently considered to comprise 33 valid species (Van Soest et al., 2008). Six species assigned to *Jaspis* were previously recorded from the Atlantic Ocean: *Jaspis eudermis* Lèvi & Vacelet, 1958, *Jaspis griseus* Lèvi, 1959, *Jaspis incrustans* Topsent, 1890, *Jaspis johnstoni* Schmidt, 1862, *Jaspis salvadori* Boury-Esnault, 1973, and *Jaspis velezi* Wintermann-Kilian & Kilian, 1984. The present paper describes a new species of *Jaspis* Gray, 1867 collected by trawling on board of the RV 'Astro Garoupa' in three campaigns of the Project of Environmental Characterization and Monitoring of Potiguar Basin, on the northern coast of Rio Grande do Norte State, north-eastern Brazil. Samples were fixed and preserved in 70% ethanol. The specimens were housed in the Porifera collections of Universidade Federal de Pernambuco (UFPEPOR) and Departamento de Invertebrados do Museu Nacional, Universidade Federal do Rio de Janeiro (MNRJ). Spicule slides were prepared by dissociation of a small fragment of sponge in boiling nitric acid. Transverse sections of the skeleton were mounted on microscope slides for identification. Photomicrographs were taken with a digital camera Sony DSC-W50 coupled with a Nikon Eclipse E-200 light microscope. Illustrations of the spicules of all species were carried out in Scanning Electron Microscope Jeol model JSM-6390. *Jaspis* sp. nov was massive, coloured externally dark brown and internally brown. Surface was covered with algae, eunicid polychaetes, corals and bryozoans. Oscules scattered over the surface. Consistency was firm, slightly compressible. The skeleton was confusing, with oxeas interlocking, oxyasters and strongylasters dispersed in choanosome. Ectosome possessed a well differentiated cortex of choanosome, formed by oxyasters and strongylasters. *Jaspis* sp. nov. had oxeas and oxyasters smaller than *J. eudermis*, however oxeas and oxyasters were larger than in *J. griseus* and *J. salvadori*. *Jaspis* sp. nov had only one category of oxeas while *J. johnstoni*, *J. salvadori* and *J. velezi* have two categories. *Jaspis* sp. nov. was different from *Jaspis velezi*, with larger oxeas, one category of spinal oxyasters and one of strongylasters, whereas *J. velezi* does not have oxyasters and has two categories of strongylasters with larger diameters.

Measuring sponge feeding habit: an analysis across the literature

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The ability to understand and predict the eco-physiological response of marine organisms to environmental drivers is of great importance within the current climate-change scenario. Given its central role for feeding, respiration, excretion and reproduction, measuring "sponge filtration/pumping activity" may prove a biologically relevant and relatively simple proxy for sponges' eco-physiology. To assess the state-of-the-art on the subject, and identify a standard methodology, we performed an extensive literature screening for the key-words "sponge" AND "Porifera" AND: "filtration", "pumping", "water flow", "retention", "feeding", "clearance" (including alternative spelling), identifying 148 relevant sources for the period 1892-2010. Of these,16 are review papers and do not provide primary data. No information beyond the title level on additional potential 12 sources was found. The 132 retained sources providing primary data involve 122 species from 11 basins/environments (including polar, tropical, temperate and freshwater habitats), investigated for any facets of the "filtration/pumping activity" of sponges. We intentionally use such a general definition because of the different activities addressed and definitions reported in the literature. These include: filtration (mostly intended as a volume of water cleared of particles in a unit of time, measured by a direct method), clearance rates (i.e., the volume of water cleared of particles in a unit of time, measured by an indirect method), retention (often used for filtration but mostly focused on the particles retained, rather than on the volume of water filtered), removal (often used for retention, but mostly focusing on changes in the surrounding water composition), flow rates (direct measurements of water volumes flowing from oscula), pump rates (often used for flow rates, but also pertaining to experiments on pressure/speed). Almost any of these activities has been measured both in the field and in the lab, but by different authors and on different species. Variable experimental set-ups have been adopted to measure the same activity and results can only rarely be compared due to the use of different units (e.g., some sources refer to sponge volume, others to wet, dry or ash-free dry weight). Unfortunately, out of the 132 sources providing primary data, just 12 measure sponge filtration/pumping at varying temperatures, and only 20 also report respirometry data (yet not always associated to T information), which could give useful indications on responses to climate drivers. This variability is due to the different purposes of research addressing sponges, which includes not only basic but also applied studies (e.g., cultivation, bioremediation). The extreme diversity of the phylum and the complex ecology of sponges harboring associated microorganisms further complicate the situation. While offering highly valuable insight on sponge eco-physiology, the literature does not provide evidence-based information to predict responses to climate-change driven temperature increase, nor advice on the ideal experimental methodology to address it. We suggest that temperature effects and tolerance limits of sponges be analyzed by simple and easily reproducible methods (e.g., clearance and respiration rates) so that results from different authors can be compared for several species/habitats and provide a comprehensive view of the subject.

Choosing settlement substrate: sponges in soft-bottom assemblages at the shelf-break frontal area, Argentine Sea

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The shelf-break frontal area of the Argentine Sea supports a high biological production as a result of high input of nutrients, consequently influencing higher trophic levels. Patagonian scallop fishing grounds (between 37° and 45°S, at approximately 100m depth) are located in the mentioned area and are characterized by the presence of soft bottoms, mainly composed by sand, as the majority of the Argentinean shelf. Given the absence of hard substrates like rocks, sessile species of this ecosystem colonize secondary hard bottoms composed by parts of living and non-living organisms. Sponges at Patagonian scallop commercial fishing grounds represent 5-10% of the total epibenthic community biomass, although at specific areas with low scallop biomass (i.e. near 40°S) they can reach higher values. Annual monitoring of the benthic assemblage associated to the scallop fishery is developed as part of the management strategy. The results here presented are a compilation of data collected in the past decade during the monitoring cruises carried out by INIDEP from selected and preserved sponge samples in shelf areas subjected to scallop trawling. More than 50% of the benthic species richness of the Patagonian scallop fishing grounds corresponds to epibiotic sessile taxa, and from those species, 40% (47 species until present) is represented by sponges. Settlement substrate (at least one possible) was successfully recorded for more than 80% of the identified species. In all the registered cases mollusks or their empty shells were found to be the substrate for the following species: *Iophon proximum*, *Clathria microxa*, *Clathria discreta*, *Clathria* (*Microciona*) *antarctica*, *Clathria* spp., *Dasychalina validissima*, *Isodyctia setifera*, *Hymedesmia* (*Stylopus*) *antarctica*, *Calyx kerguelensis*, *Tedania mucosa*, *Tedania massa*, *Tedania spinifera*, *Tedanis* sp., *Mycale doellojuradoi*, *Mycale magellanica*, *Amphilectus fucorum*, *Dictyonella* sp., *Stelodoryx argentinae*, *Stelodoryx cribrigera*, *Callyspongia ramosa*, *Pachychalina* sp., *Phorbas* sp., *Ulosa* sp., *Halichondria* sp., *Hymeniacidon* sp., *Lyssodendoryx* (*Ectodoryx*) spp., *Haliclona* spp., *Sycon* sp., *Oceanapia* sp., *Eurypon* sp., *Chalinula* sp., Suberitidae and Clionaidae (holes). Among these species, 14 were registered on *Zygochlamys patagonica* (mainly empty shells) and 33 on *Fusitriton magellanicus* (see Schejter et al., poster presentation). Some of these species (*Halichondria* sp., *Phorbas* sp., *Haliclona* cf. *bilamellata*, *Haliclona* (*Reniera*) sp. and *Haliclona* (*Haliclona*) sp.) were registered in this benthic community using other substrates like crab carapaces, dead coral, Rajoidea egg capsules and polychaete tubes. Due to sampling procedure (dredges and trawls), sponges (and other benthic organisms) were frequently damaged or fragmented and the recognition of the original settlement substrata was not always possible. The commercial species *Zygochlamys patagonica* and the hairy snail *Fusitriton magellanicus* allow the development of highly diversified sponge gardens, a peculiar habitat that can host a wide variety of endobionts, mainly microcrustaceans and polychaetes (see Schejter et al. (2), poster presentation). The risk of overexploitation of scallop beds could lead to a general loss of biodiversity, or at least to a transformation, because living molluscs are able to host the majority of the species, but their dead valves were not preferred by the same species.

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The gastropod *Fusitriton magellanicus*: a very suitable substrate for the settlement of sponges in soft bottoms of the Argentine Sea

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Fusitriton magellanicus is frequently collected in the by-catch at *Zygochlamys patagonica* commercial fishing grounds in the Argentine Sea, located in the shelf-break frontal area, one of the most productive ecosystems in the SW Atlantic Ocean. Preliminary results showed that *F. magellanicus* would be the second most important living substrate colonized by encrusting organisms in this community, fact that highlighted its importance in the maintenance of the species richness in the exploited bottoms influenced by the shelf-break front in Argentine waters. However, sponges using this gastropod shell as settlement substrate deserve more detailed studies. In this sense, we studied living *F. magellanicus*, empty shells and pagurized shells that were collected between 37°00'27" and 45°01'70" S and 54°40'46" and 60°25'62" W, along the 100m isobath and between 81-150m, during 3 research cruises in 2007 and 2008. In total, 556 shells were studied (443 living *F. magellanicus*, 86 empty shells and 27 pagurized shells). More than 70% of all the studied shells presented encrusting organisms. More than 20% of the living studied shells was encrusted by sponges (only 8.14 and 7.40% were encrusted with sponges, for empty and pagurized shells, respectively). In total, 25 species were identified by means of the classic methodology described by Rützler (1972). The most frequent sponge species encrusting living individuals of *F. magellanicus* were *Hymedesmia* (*Stylopus*) *antarctica* (20%), *Tedania* spp. (9%) and *Dictyonella* sp. (7%). These species were often found heavily fouling the shells. Moreover, *Tedania mucosa* can reach twice or three times the volume of the living gastropod, resembling a "motile sponge". Other sponges less frequently registered on living gastropods and empty shells (i.e., 5%) were *Clathria* (*Microciona*) *antarctica*, *Callyspongia ramosa*, *Stelodoryx argentinae*, *Stelodoryx cribrigera*, *Amphilectus fucorum*, *Iophon proximum*, *Clathria* (*Microciona*) spp., *Lissodendoryx* (*Ectodoryx*) spp., *Ulosa* sp., *Hymeniacidon* sp., *Haliclona* spp., *Halichondria* sp., *Chalinula* sp., *Eurypon* sp., *Oceanapia* sp. and *Phorbas* sp.. We also found other species belonging to Suberitidae. More than 70% of the empty shells (but only 12% of the living organisms) presented holes, possibly bored by Clionaidae sponges. Only 6 (6%) of the living gastropods were encrusted simultaneously by 2 sponge species while monospecific sponge colonies were registered in the majority of the individuals. Even so, sponges could share the shells with other epibionts, sometimes competing for the free space; the sponges overgrowth and cover other epibionts that could eventually die (i.e. bryozoan colonies). Considering that the study area is a soft bottom and the only available substrates for settlement of sessile species are hard parts of other living organism (or empty shells or carapaces), we confirm that shells of the gastropod *F. magellanicus* play a very important role for settlement of sessile species, and that they represent the substrate hosting the highest species richness of sponges in the area.

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Endobiont fauna of Porifera collected in the shelf-break frontal area of the Argentine Sea. Preliminary results on *Mycale magellanica* (Ridley, 1881)

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Literature about sponge endobionts and other associate species is very extensive. Many studies and reviews have shown the ecological, biochemical and physiological impact of these associations that could be sometimes species-specific. According to other studies in different regions of the world, polychaetes and amphipods are the most common endosymbionts. The type of endobionts and the species hosted are a function of both, morphology of the sponge and biotic interactions, but apparently no relationship was found considering abundances of endobionts and volume of the sponge. Endobionts obtain structural or chemical refuge, direct or indirect sources of food and sites for reproduction, among others. Endofauna of many sponges from North Atlantic, Mediterranean, East Pacific and Antarctic waters was studied, but few species were studied in the SW Atlantic Ocean (i.e. *Hymeniacidon sanguinea*, *Mycale (Aegropila) angulosa* and *Mycale microsigmatosa*). In this study we present preliminary results on the endofauna of *Mycale magellanica,* one of the most widely distributed sponges in the Argentine Sea.Samples were obtained with trawling devices in four localities along the shelf-break front, at nearly 105m depth and between 39° 24' S - 41° 51' S and 55° 56`W – 58° 09' W. Sponges were collected as part of the Patagonian scallop assemblage during stock assessment cruises. Consequently, due to this sampling procedure, specimens often were damaged and only the biggest pieces of *M. magellanica* were preserved for the present analysis. A total of 2.7 kg (six colonies between 340 g and 660 g) of wet material was dissected and examined under binocular microscope at laboratory and 846 individuals were sorted (average 350 individuals per kg). The number of taxa identified was 22. Crustaceans, mainly Amphipoda and Isopoda, reached between 66 and 90 % of the total number of specimens. The most abundant taxa were the amphipod Lysianassidae and the isopod *Caecognathia* sp. Other peracarids registered were *Leucothoe* sp., *Seba cf. saundersii*, *Colomastix bastidai*, *Liljeborgia* sp., *Iathrippa* sp., *Fissarcturus* sp. and *Acanthoserolis schythei*. We found juvenile stages and incubating females of some peracarids, what suggests their close relationship with *M. magellanica.* The rest of the endobiotic taxa were molluscs (mainly *Hiatella meridionalis*), echinoderms (*Ophiactis asperula*, *Ophiura lymani*, *Ophiacanta vivipara* and *Pseudocnus dubiosus*), polychaetes (Spionidae, Polynoidae, Phyllodocidae) and the decapod *Nauticaris magellanica*. These results represent the first attempt to elucidate the composition of endobionts in sponges from the deep waters of the Argentine Sea. *Mycale magellanica* seems to be important in providing habitat for at least 22 species of small invertebrates, mainly crustaceans, in the study area. This is of particular interest considering the fishing effort exerted in the different Patagonian scallop banks there distributed, subjected to removal of benthic species. In this sense, sponges in general could contribute to enhance biodiversity in areas scarcely disturbed.

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Defensive strategies of tropical Pacific sponges: Examples from Micronesia

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Sponges, like any other sessile benthic organisms, have to deal with several ecological threats like predation, fouling, or competition. To overcome these threats, they have evolved a wide array of defensive mechanisms, such as morphological or chemical defenses. The aim of this study was to determine how common sponges from Guam and surrounding Micronesian islands employed secondary metabolites as defense against predators and microbial pathogens. To test for antipredatory defenses choice feeding assays were conducted in the field with omnivorous fishes and in the laboratory with the pufferfish *Canthigaster solandri*. Therefore, sponge crude extracts were incorporated at natural concentrations into an artificial diet and offered simultaneously with food containing solvent only to potential fish predators. In addition, the same sponge extracts were tested at natural concentrations for possible antimicrobial activity in disc diffusion assays against ecologically relevant bacterial isolates. Bacterial isolates had previously been isolated from biofilms of reef rock and crustose coralline algae. A subset of the investigated sponges was further analyzed with regard to dynamic defensive strategies, such as activated or induced defenses, to evaluate whether such strategies are an exception or widely represented among various Porifera species. Furthermore, we investigated whether allocation of defenses followed strategies such as the Optimal Defense Theory.

Natural and anthropogenic induced changes in shallow water sponge communities on Guam reefs

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During the assessment of benthic invertebrate assemblages along permanent transects at five reef sites around Guam, we observed a pronounced change in the abundance of the encrusting marine sponge, *Terpios* sp. at Double Reef, Guam. Percent cover of *Terpios* increased from 13% to 23% during the course of one year (2007 to 2008). In the past *Terpios* has been reported as a nuisance species covering large areas of hard substrate and actively overgrowing live and dead coral. While Bryan[1] implied that *Terpios* is nutritionally benefitting from coral, growing four times faster on living coral surface compared to bare reef substrate, Plucer-Rosario[2] found that growth rates were fastest in this order: cleaned bare rock > live coral > bare reef rock, and further suggested that microbes on the surface of bare reef rock inhibit the growth of *Terpios*. Our aim was to evaluate the substrate preference and growth patterns of *Terpios* sp. in the field and laboratory, as well as to characterize its chemistry in terms of possible allelopathic activities, which could aid the sponge to overgrow its scleractinian hosts. Furthermore, several mechanisms, which possibly aid in its dispersal and spreading, were evaluated. Investigating the mechanisms of dispersal and how *Terpios* can overgrow and occupy various substrates might help to understand the pronounced changes in benthic cover of this nuisance sponge. We also monitored the sponge community in Apra Harbor, Guam, with regard to anthropogenic impacts from the dredging and subsequent construction of a 400 feet wharf extension during 2009 and 2010. Sponge assemblages were monitored along 81 transects (25m x 1m each) at various distances from the construction site. In addition, changes in sponge size were monitored by taking 6 permanent photo quadrates at the beginning of each transect and taking three-dimensional measurements of the sponges inside the quadrates. Results will be discussed in relation to oceanographic and modeling data from the sediment plume of the construction site.

[1] Bryan 1973. Micronesica 9 (2), 237-242; [2] Plucer-Rosario 1987. Coral Reefs. 5, 197-200.

Seasonal fluctuation in tissue structure of long-living Spongillidae from Lake Baikal

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The sponge fauna of Lake Baikal is represented by two families: endemic Lubomirskiidae and cosmopolitan Spongillidae. We consider Lubomirskiidae to be derived from Spongillidae; these latter have greatly changed their short-term life cycles in specific environment of the giant ancient lake. Sponges of *Trochospongilla* genus are suggested as possible ancestors of Lubomirskiidae. Representatives of this genus inhabit the littoral zone of open Baikal and do not produce gemmules. Our investigations show that Baikal *Trochospongilla* spp. live at least more than one year, vegetate through all-year-round and reproduce sexually. The current study is aimed to analyse the seasonal changes of their tissue structure. *Trochospongilla* spp. were collected from the depths of 1.5–17 m, on Berezovy test site (South Baikal), which is a typical zone of open littoral. Samplings were accomplished in different seasons during 2001–2003. Histological approach was used in order to investigate tissue organization. It was found out that Spongillidae tissues changed noticeably during the year. The sponges collected in summer demonstrated a normal tissue structure, characterized by aquiferous system connected with choanocyte chambers, inhalant and exhalant canals lined with endopinacocytes. Mesohyl contained different cell types: archaeocytes, eosinophilic cells etc. Late oocytes, larvae, spermatic cysts were also observed in mesohyl. Histological analysis clearly evidence on the disordering of aquiferous system during the late autumn. Canals were rare in the sponge, often they were lacking of the endopinacocytes lining. The most of choanocyte chambers were destroyed. A number of free laying choanocytes and a lot of archaeocytes were observed in mesohyl. Archaeocytes demonstrated a trend to aggregation. These cells' cytoplasm got rich in basophilic inclusions (most likely phagosomes). A few sponge specimens contained early oocytes. Winter sponge tissues consisted almost exclusively of archaeocytes with numerous phagosomes. No choanocyte chambers or canals were observed. One specimen contained embryos at the stage of cleavage. Archaeocytes were still the prevalent cell type in sponge tissues at the end of March – beginning of April. However, the clusters of choanoblasts in mitotic activity and rare choanocyte chambers were also observed. Early oocytes were found in one of the sponges. Thus our data evidence that long-living Spongillidae of the open Baikal annually experience deep reorganization of their tissue structure. This process includes reducing of the aquiferous system and changing of cell composition during the autumn – winter and their reestablishing during the late spring – early summer. Reproductive elements are usual in summer and are sporadic in other seasons. Unlike *Trochospongilla* spp., Lubomirskiidae collected in the same biotope at the same time demonstrate permanent tissue organization in any season of a year. It is well known that Siberian Spongillidae form gemmules and do not survive in winter. Consequently, the life cycle of the open Baikal Spongillidae differs from both Lubomirskiidae and Siberian Spongillidae life cycles. This enables us to speak about evolving of the three diverse patterns of sponge life cycles, which reflect differences in sponge phylogenetic positions and environmental conditions of biotopes.

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Sponge biodiversity of mesophotic coral reefs and their biotechnological potential

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Mesophotic coral reefs [MCRs] are deep (30-150 m) fore-reef communities comprised of low-light adapted macroalgae and zooxanthellate corals, sponges, gorgonians, and fish. There is some overlap in species composition with the adjacent shallow reefs, but MCRs also harbor unique species not found in the shallow community. Because MCRs are typically found farther from anthropogenic disturbances than their shallow reef counterparts, there has been much hope that they would serve as both a refuge for corals, sponges and fish as well as a potential source of larvae that could contribute to the resiliency of degraded shallow reefs. Moreover, these extreme habitats have the potential to harbor unique phenotypes and/or genotypes with rich biotechnological products. Between 2003 and 2009, we surveyed the sponge communities of the MCRs at Lee Stocking Island, Bahamas and Little Cayman Island using technical diving procedures. These surveys were compared to similar surveys conducted in nearby shallow reef communities to assess the degree of variability in sponge community structure within and between depths and geographic locations. In both locations there appears to be a "species break" at about 60m depth, with deeper transects yielding several new species. Sponge samples collected from these shallow and mesophotic coral reefs were assessed for potential pharmaceutical activity against several common human pathogenic microbes. Almost twice as many MCR samples exhibited bioactivity in these assays than did shallow reef samples. In addition, certain species that co-occurred across the entire depth gradient were significantly more bioactive at MCR depths, indicative of quantitative and/or qualitative differences in their natural product constituents. Our results indicate that MCRs represent an important source of sponge biodiversity and chemodiversity, with tremendous biotechnological promise.

Population dynamics of a coral-infesting sponge, *Terpios hoshinota* in Taiwan: A preliminary study of the role of typhoons

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A population explosion of the encrusting sponge *Terpios hoshinota* was noticed in the corals reefs of two off-shore islands east of Taiwan in 2007. Densities up to 62 individual/100 m^2 was recorded among 32 survey points around Green Island and Orchid Island, although most sponges are <100 cm in longest axis. Most sponges were found in shallow waters, although a few occurred down to 10 m depth. Sponge sizes were significantly larger in shallow 2-3 m than in 5 m depths. They covered live corals and killed the underlying host tissues without specificity. The average expansion rate is 0.18 cm/d on live corals, with a smaller rate in non-live-coral surfaces. Two nutrient-experiments in the field failed to find supporting evidence of the role of nutrients on the growth rates of the sponge. The abundance of the sponge in the sea is also not related to the geographic distribution of human population. Experimental light blockage, on the other hand, induced tissue degeneration in a couple of weeks. This light-dependency reaction explains the depth distribution of the sponge. Strong wave actions caused by typhoons can either kill the sponges or reduce the sponge sizes as indicated by a photo-survey comparison before and after a typhoon in 2009. Possibly due to the typhoons, the sponge is kept in check and the densities and the coverage of the sponge did not increase in 2009.

Do physical forces promote or inhibit the coral-killing sponge, *Terpios hoshinota*?

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High densities of a black encrusting sponge, *Terpios hoshinota,* were found growing on all species of stony corals at Green Island and Orchid Island, southeast of Taiwan in 2007. They grow on surface of live corals and eventually cover the whole colonies and kill them. The average expansion rate on the coral-sponge fronts was 1.8 mm per day for measurements done between March and July 2008. No effects of nutrient addition on expansion rates were detected in two field experiments, and the sponge densities were not related to the densities of human inhabitants on the islands. Blocking of light inhibit the growth of this sponge with symbiotic cyanobacteria. The abundance of the sponge is higher in shallow (2-3 m) than in deeper waters (5 and 10 m). The sponge can also grow on non-live-coral substrate. Moreover, they grow on artificial substrates such as glass slides, plastic sheets and meshes. This capability endows the sponge to cross and infect many unconnected coral colonies on the reefs. In addition, sponge fragments, with or without coral skeleton, are able to infect new coral hosts. These fragments may be generated by strong waves. The largest sponge patch was 8 m across. However, the expansion rates on non-live-coral surfaces are about 50% of that on live corals. Using the 2008 densities, sizes and the expansion rates of sponges, as well as the coverages of stony corals, we simulated that the sponge has the potential to cover 70-100% of the shallow reefs of Green Island in 5 years. The above simulation did not consider recruitment, predators or physical factors that may promote or inhibit the growth of the sponge population. The 2009 survey indicates that the sponge coverage and densities, based on more than 30 sites in the two islands, were below the prediction. Obviously, some important factors are inhibiting the sponge. We are currently testing two hypotheses that may undermine the sponges. The first is the possible extreme temperatures that the sponges may not be able to acclimate to. The second is the wave actions generated by typhoons and monsoons that may kill the sponges. These two hypotheses are not mutually exclusive.

Reproduction and development in *Haliclona indistincta* (Phylum Porifera: Order Haplosclerida)

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The intertidal species *Haliclona indistincta* is a common inhabitant of the marine intertidal regions around Ireland and Britain, its suggested range extends from the English Channel, to St. George's Channel, Irish Sea, western seaboard of Ireland and Scotland. It is an important member of its environment, providing a habitat for numerous species of flora and fauna. Little work has been done on this species beyond the initial descriptions of the mature individuals and larvae. The species is viviparous: brooding embryos in the mesohyl until they are released as free-swimming larvae. The reproductive cycle was investigated using histological and electron-microscopy methods. Thirty adult specimens are samples on a monthly basis from Corranroo, Co. Galway, Ireland from September 2009, and fixed in Bouin's solution. To observe the progressive development and differentiation of the cells into gametes, the Bouin's fixed specimens were sub-sectioned, embedded in paraffin, sectioned, and stained with eosin and hematoxylin. To study the ultrastructure of the developing larvae, embryonic packets inside the parent sponge are sampled from individuals from the shore once a week throughout the reproductive season of this species, May-July. The embryonic packets are fixed in Glutaraldehyde solution and prepared for Transmission electron microscopy (TEM) and Scanning electron microscopy (SEM) by standard protocol. To secure free-swimming larvae, three methodologies were employed. Firstly, *H. indistincta* specimens were placed in a closed system, in 250 liter tanks, in a constant temperature room of 15°C throughout their reproductive season. The water was aerated and specimens were fed once a week. Secondly, settlement panels were developed and placed in regions were there was a high population of this species. Thirdly, larval traps containing collection tubes were designed and placed over specimens in situ. The results of this work will be presented.

Patterns of polychaete preference for mangrove sponge hosts

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Sponges growing on mangrove roots provide habitat for smaller invertebrates like crustaceans, cnidarians, and polychaetes. Because each root consists of a unique assemblage of sponge species and the distance between roots potentially limits dispersal, the extent to which polychaetes actively choose their host is unclear. The mangrove sponge host preferences of surface dwelling syllid polychaetes were examined using laboratory choice experiments and *in situ* observations. For laboratory choice experiments, pieces (~1.5 cm^3) of 5 sponge species (*Biemna caribea*, *Halichondria magniconulosa*, *Tedania ignis*, *Spongia obscura*, and *Haliclona implexiformis*) were collected from Twin Cays, Belize. While sponges were submerged in seawater, all polychaetes were removed under a dissecting microscope. The initial abundances were recorded as well as the body and intestine color since the polychaetes appear to prey on the host. Pieces of each species were combined in 7 replicated beakers with fresh seawater drips. Approximately 10 polychaetes were added to each beaker and the proportion of total polychaetes on each sponge species was recorded after 2 hours. *In situ* abundances were also quantified. Pieces (~3cm^3) from 4 randomly selected individuals of the 5 sponge species were preserved for polychaete abundance quantification in 4% formaldehyde and seawater. *In situ*, *H. magniconulosa* and *T. ignis* hosted the highest polychaete densities at 16 and 10 polychaetes/cm^3, and *S. obscura* and *H. implexiformis* hosted the lowest polychaete densities at 1 and 0.5 polychaetes/cm^3. *Halichondria magniconulosa* hosted significantly higher polychaete densities than *S. obscura* and *H. implexiformis*. *Tedania ignis* was the only species that hosted polychaetes that were the same color as the sponge. Polychaetes on the other species were colorless with a dark intestine. In the choice experiment, the average proportion of polychaetes was significantly higher on *B. caribea* (0.5) than all other species, and significantly higher on *H. magniconulosa* (0.3) than *T. ignis*, *S. obscura*, and *H. implexiformis*. Additionally, orange polychaetes initially removed from *T. ignis* were found dwelling on all other sponge hosts except for *S. obscura*. This study suggests that the highest polychaete densities *in situ* are found on *H. magniconulosa*, and, when polychaetes are given a choice between these 5 species in the laboratory, higher proportions of polychaetes are found on *B. caribea* and *H. magniconulosa*. *Spongia obscura* and *H. implexiformis* hosted the lowest polychaete densities *in situ* and the lowest proportion of polychaetes in the lab experiment. The pattern of polychaete distribution on *T. ignis* appeared to differ between *in situ* observations where it hosted high polychaete densities similar to *H. magniconulosa* and the choice experiment where the low proportion was similar to *S. obscura* and *H. implexiformis*. This pattern may be due to exudates from *T. ignis* produced because the pieces were not healed before the choice experiment or from stress in the laboratory environment. The patterns of polychaete preference observed in this study conducted in May are similar to previous choice experiments and *in situ* observations conducted in December. Future choice experiments should use cleaned and healed sponge pieces that are presented to polychaetes *in situ*.

The impacts of anthropogenic sedimentation on sponge diversity and abundance on the North shore of Jamaica

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Jamaica's growing dependence on tourism has led to construction of new hotels and resorts on the north shore of Jamaica. These resorts must either truck in or dredge sand to create artificial beaches on the rocky shore. A higher level of sedimentation is found in areas near these hotels and has a direct impact on benthic reef organisms. Due to the high filtration requirements of sponges, suspended sediment levels may act as an environmental parameter controlling the distribution and abundance of sponge species. Surveys were completed at three sites along the north coast of Jamaica to determine if species diversity and abundance were impacted by sedimentation over spatial and temporal scales. Both species diversity and abundance were lower in the sedimentation site as compared with the control sites. Sedimentation appears to be a controlling factor for the distribution of sponges on the reefs of Jamaica. Increasing coastal development will further exacerbate the reef degradation in this area by decreasing the habitat complexity of the reef.

A long-term change in distribution and abundance of a marine sponge, *Neopetrosia* sp. in the Gulf of Thailand

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Neopetrosia sp. is a dominant sponge found in coral communities in the Gulf of Thailand. The purpose of this study was to compare distribution pattern and population density of *Neopetrosia* sp. in coral communities of Khang Khao Island, the inner part of the Gulf of Thailand between two surveys in 1998 and 2010. A quadrat (0.5×0.5 m^2) was placed randomly on coral zones of the study sites. Number of colonies, colony size and substrate type were recorded by SCUBA divers. The mean population density in 2010 (1.04 colonies/m^2) was significantly higher than that in 1998 (0.37 colonies/m^2). *Neopetrosia* sp. attached mostly to a massive coral, *Porites lutea*, a zoanthid, *Palythoa* sp., and rocks. Partial mortality of colonies of *Neopetrosia* sp. was obviously due to a sea slug, *Jorunna funebris*. We found significant differences of population densities of the sponge on *P. lutea* between the two study periods, 0.20 colonies/m^2 in 1998 and 0.95 colonies/m^2 in 2010. An increase on the population density of *Neopetrosia* sp. attached on *P. lutea* may indicate the degradation trend of coral communities in the Gulf of Thailand.

Patterns in shallow water sponge richness and biogeography within the Western Indian Ocean

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Sponges are, in many respects, ideal organisms for the study of biogeographic patterns on shallow, marine hard benthic reefs. They have relatively similar species numbers in any one large region from temperate to tropical regions, and comprise representatives of three major phyla, which, though divergent phylogenetically, have a series of similar functional forms. The tropical Indo–West Pacific is the largest coastal biogeographic region on earth and also has the most sponges recorded for any one region, with approximately 250 sponge species for the Indian Ocean alone. An analysis of the large scale patterns of sponge species richness has, however, never been performed in the WIO. Our analysis revealed the following patterns: 1) species turnover is lowest within and between the (North Red Sea, South Red Sea and Arab Basin) and the (Mascarene Plateau, Somali Basin and Mozambique Channel) branches. On the other hand, species turnover is at its largest at the southern extreme, i.e. at the boundary between Cape Point and Natal-Delagoa bioregion. The positioning of Delagoa bioregions with Mozambique, demonstrates that this bioregion acts as a bleeding zone between the Indo-Pacific and the South African east coast. The analysis revealed that the WIO can be split into at least three biogeographical realms: 1) the red sea and associated Arab Basin, 2) the asymmetrical circumtropical region stretching from the horn of Africa to southern Mozambique including the Delagoa bioregions of South Africa, and 3) Southern Africa excluding the Delagoa bioregion. Given the preliminary patterns revealed here, it is becoming clear that important taxonomic turnovers occur within the latitudinal and longitudinal gradients of the WIO, which is not all that homogeneous an area as has long been thought. The biogeography of the WIO can be explained by 1) dispersion ability, 2) current patterns, and 3) recent geological history. We also found that Rapoport's rule cannot be upheld across the latitudinal gradient studies here. This pattern is contradictory to the rule that dictates that species' range sizes decrease towards lower latitudes, i.e. Rapoport's rule.

Specific growth process in a hexactinellid sponge of the genus *Aspidoscopulia*

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A new representative of the genus *Aspidoscopulia,* characterized (besides specific spicules set) by giant sizes, dichopodial-monopodial branching and two types of symmetry (bilateral and metameric=pseudometameric), possesses a specific type of growth. Formation of the carina (line of fusion margin-to-margin of the opposite sides of the expended wall during oscular marginal growth which led to dichotomous-isotomous branching), well known for many other hexactinellids belonging to the order Hexactinosida and for other species of the same genus, is replaced in this unique species by fusion of the atrial surfaces by their inwardly directed surfaces. Extended fused surfaces of the walls form the ear-like, flat processes in which the atrial cavity is absent (being eliminated between two met and fused walls). The peculiar body shape of *Aspidoscopulia* sp.n. is explained by a scheme.

A new type of deep-water reef constructed by a glass-sponge *Sarostegia oculata* Topsent (Porifera: Hexactinelida) from the Mount Error Guyot (NW of the Indian Ocean)

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A new reef-builder among the hexactinellid sponges, *Sarostegia oculata* Topsent, 1904, is found in the archive of photos. Unlike known before, huge reef constructions of the hexactinellid sponges (*Heterochone calyx* and *Aphrocallistes vastus*) described from the shelf of the coast of SW Canada, the newly found constructions are much smaller, being rather small banks. Besides they are notably deeper, bathyal, situated at a depth of about 1200 m at slope of the Mount Error Guyot (NW of the Indian Ocean). Similar to the former, the newly found banks function as other reefs, when the dyed bodies provide substratum for the successful settlement of larvae of same species and their further growth.

Identification of sponges (Phylum Porifera) inhabiting pre-selected sites in Talim Bay, Lian, Batangas

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Sponges in nine sampling sites (Clubhouse 2A, Galvez 5A, Inner Talim 6A, Middle Duenas Reef, Layag-layag 3A, Outer Talim 7A, Torso 4A, Roces Mangrove waterway and Roces) in Talim Bay, Lian, Batangas were collected from May 2-8, 2009. Twenty-four different sponges collected belong to class Demospongiae. Only 21 were identified and classified into 10 families: Halichondriidae, Clionaidae, Axinellidae, Chondropsidae, Ircinidae, Petrosiidae, Microcionidae, Chalinidae, Niphatiidae, and Callyspongiidae; and 14 genera: *Ciocalypta*, *Spheciospongia*, *Axinella*, *Phoriospongia*, *Sarcotragus*, *Petrosia* (*Strongylophora*), *Xestospongia*, *Clathria*, *Haliclona*, *Haliclona* (*Gellius*), *Chalinula*, *Niphates*, *Callyspongia*, and *Callyspongia* (*Cladochalina*). The sponge collection was dominated by the genus *Haliclona* obtained from Clubhouse 2A, Outer Talim 7A, Torso 4A, Roces mangrove waterway, and Roces. At the species level, only *Callyspongia* (*Cladochalina*) *samarensis*, *Haliclona* (*Gellius*) *sigmadocia*, *Xestospongia testudinaria*, and *Chalinula nematifera* were identified and verified. Comparison of the current survey with Elmido et al. (1995) revealed more diversity in terms of genera and species identified. Seven (29%) of the 24 sponges collected in the present survey were of genus *Haliclona*, of which only *Haliclona* (*Gellius*) *sigmadocia* was identified and verified. None of the 41 sponge species in the collection of Elmido et al. (1995) belonged to the genus *Haliclona*. *Petrosia* and *Xestospongia* were the most dominant genera in the sponge collection of Elmido et al. (1995), and both genera were also identified in the current study. Other less represented genera in 1995 that have been likewise identified in the current survey are *Xestospongia*, *Clathria*, *Petrosia*, *Petrosia* (*Strongylophora*), *Ciocalypta*, *Callyspongia*, *Callyspongia* (*Cladochalina*), and *Niphates*.

A taxonomic inventory of sponges from abyssal depths in the Norwegian-Greenland-Iceland (GIN) Seas

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The abyssal (>2000 m depth) zones of the GIN Seas (Greenland-, Iceland- and Norwegian Seas) are parts of the Arctic deep-sea region. While depths of more than 2000 m are found in only a small part of the Iceland Sea, depths to more than 4500 m occupy considerable areas in the Greenland- and Norwegian Seas. Depths larger than 2000 m of the GIN seas have been investigated by a number of expeditions and cruises during the last 130 years and in many cases sponges were reported (Hansen 1885; Lundbeck 1902, 1905, 1910; Hentschel 1929; Koltun 1964; Borojevic & Graat-Kleeton 1965; Steenstrup and Tendal 1982; Brattegard et al 1988; Tendal 1989; Barthel & Tendal 1993; Janussen et al 2003; Cardenas et al 2010). Characteristics of the fauna are a high number of endemic species, a generally low diversity as compared to the Atlantic abyssal fauna, and extremely high abundance of some species. Our results show that the GIN Seas sponge fauna bears clear similarities to the Arctic Ocean and the north-western Pacific sponge faunas. It is a general trend in all oceans that as to number of species calcareous sponges (Calcarea) inhabit mainly the coast and shelf areas, the siliceous sponges (Demospongiae) dominate the bathyal zone, while the siliceous sponges and the glass sponges (Hexactinellida) are the equally represented in the abyssal zone. In the abyssal zone of the GIN Seas the calcareous sponges are highly abundant both on soft and hard substrates and together with a core group of species within the Caulophacidae, Theneidae, Cladorhizidae, Hymedesmiidae, and Polymastiidae they represent a quite uniform widely distributed sponge fauna. The BIODEEP and H2DEEP cruises during 2006, 2007, 2008 and 2009 have provided fresh samples of a major fraction of the sponge fauna in the area and around 40 species have so far been identified. Use of ROV provided unique ecological data and HD quality video and still photos of dominating sponge taxa in the area, including even specimens less than 1 cm in size.

Implementation and use of a genomic DNA bank to screen the genetic diversity of Brazilian marine sponges

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The most important information of a species is the one in its genome. DNA banks, or databases, are a way to preserve biological diversity. Resources of DNA banks will enable scientists to access biological samples that are stored in a proper way and gathered in different points of time and space. It will enable not only to study the present state, but also to observe the trends of genetic changes in ecosystems, species and populations. Thus, DNA banks which enable systematic collection, preservation, and distribution of samples will play a vital role in conservation genetics studies of, biodiversity, taxonomy, and comparative genomics. However, few efforts have been made in the sampling and documentation of genomic DNA as a genetic resource. In Brazil, there are several DNA banks, mainly of endemic and threatened Brazilian species, mostly vertebrates and plants. The main objective of this work was to implement and explore the first DNA bank of Brazilian marine sponges and to develop tools for detection of genetic variability. The bank consists on two types of products: extracts of cells and purified total genomic DNA. These samples are classified and preserved in deep freezers or liquid nitrogen. From 394 cell extracts, 203 have "vouchers" stored in the Museu Nacional / Universidade Federal do Rio de Janeiro (MN/UFRJ) collection. A survey of the amount of cell extracts and their bulks was done, yielding volumes varying between 0.1 and 15.6 mL, from which 375 had their pigment content catalogued. The great majority of the yielded DNA samples are of high molecular weight and reasonable concentration. Some are somewhat degraded because the sponge from which they were extracted was preserved for a long time in museum collections, whereas others are more dilute than desirable. But, some sponge species never yield much DNA, due to their molecule composition (viz. amount of complex polysaccharides). The analyzed extracts comprise 11 of the 15 orders of Demospongiae. Among the most representative genera are *Aplysina* Nardo, 1834 (6 sp., 88 specimens); *Amphimedon* Duchassaing & Michelotti, 1864 (2 sp., 68 specimens); *Cliona* Grant, 1826 (29 specimens of *C. celata* Grant, 1826); *Dragmacidon* Hallmann, 1917 (5 morphotypes, 42 specimens of *D. reticulatus* (Ridley & Dendy, 1886)); *Geodia* Lamarck, 1815 (8 sp., 40 specimens); *Hymeniacidon* Bowerbank, 1858 (44 specimens of *H. heliophila* Parker, 1910) and *Mycale* Gray, 1867 (8sp., 64 specimens). Additionally, the ongoing analysis of the COI genetic marker (standard "Folmer" fragment or "Rot" fragment) applied to the DNA bank samples, would allow investigation of populations' structure, resolution of taxonomic uncertainties, individual identification, and the determination of levels of genetic diversity.

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Porifera LifeDesk: an internet-based tool for collaborative systematic documentation of species

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Over 8,300 species of Porifera have been described; however, because the phylum is estimated to contain at least 15,000 species, many additional species remain to be fully documented and named. As part of the Porifera Tree of Life (PorToL) project, we are working with the Encyclopedia of Life (EOL; www.eol.org) to develop Porifera LifeDesk (http://porifera.lifedesks.org/), a freely accessible tool for collaborative documentation of biodiversity. In our collaboration with the LifeDesk programming team, we serve as early users who can provide feedback to improve this resource. By sharing information more efficiently among sponge systematists, we hope to accelerate the documentation of sponge species, including naming and describing species that are currently poorly understood. LifeDesk and EOL use the taxonomic classification provided by the World Porifera Database; both websites can be explored using a classification tree. Our poster illustrates the methods by which LifeDesk can be used to share information. At the LifeDesk, users can add web pages for known and unknown sponge species using the graphical, tree-based browser. Pages can be added for morphotypes or variants as well as for entire species. Users can upload images of *in situ* specimens, isolated spicules, and histological sections. A variety of text fields are available to report synoptic descriptions, species distributions, and life-history traits. Supporting documentation can be submitted as PDF files. For images, text, and files, LifeDesk provides options to limit access to registered users or to publish the information to the entire web. One of the strongest features of LifeDesk is the availability of commenting boxes for each category of text and each image. For unpublished (or "draft") material, the submitter can create commenting boxes for use by registered users only. These boxes allow for real-time discussion of any aspect of sponge biology that users can imagine. LifeDesk complements existing databases (e.g., World Porifera Database, Sponge Barcoding Database) by providing a forum for taxonomic discussion and description of new species. In addition, LifeDesk can be easily linked to other databases documenting sponge biodiversity. For example, we will link each LifeDesk page to morphological and molecular data in the Porifera Tree of Life project database (www.portol.org). Ultimately, material placed into LifeDesk will mature into EOL web pages, yielding a freely accessible and durable platform for documenting studies of taxonomy, systematics, and biodiversity. We will be available during the conference to assist individuals that wish to give LifeDesk a trial run and to provide feedback on how this resource can be made even more useful.

Species composition and zonation of sponges on the “Schulz Massive” – sponge grounds on an arctic seamount dominated by hexactinellid sponges

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Seamounts host many of the known mass occurrences of sponges and because they are often remote, they were until recently undisturbed by human activities. Their ecosystems are vulnerable and higher fishery pressure has led to negative impacts on their sessile fauna. In contrast to more southern seamounts around the Azores, off West Africa and Portugal, etc., which are being intensively investigated, there are only few studies on arctic seamounts (Heinrich et al. 1992). The Schultz Massive is situated on the mid ocean ridge in the transition zone between the Norwegian and Greenland Seas on 73.5°N, and extends from a depth of 3000 m up to 550 m. The BIODEEP and H2DEEP cruises during 2006-2009 have revealed a unique and undisturbed type of hexactinellid sponge ground and have provided fresh samples of a major fraction of the sponge fauna. Use of ROV also provided ecological data and resulted in HD quality video and still photos of the dominating sponge taxa in the area. The hexactinellid fauna of the region is only fragmentary investigated but some early reports provide a sound basis for this study (Hansen 1885; Schulze 1900; Burton 1928; Koltun 1964, 1967; Barthel and Tendal 1993). Literature survey (Klitgaard and Tendal 2004) and recent investigations have shown that going north and into negative temperatures the Hexactinellida become dominating on sponge grounds contrary to Astrophorida in boreal and temperate waters. Our preliminary data suggest that there are about ten species of Hexactinellida on this particular seamount, dominating in the deepest and the shallowest part. The Calcarea are in general poor in species but are highly abundant in the deepest part. Different groups of Demospongiae are found over the entire depth gradient, but they are particularly dominant at intermediate depths where *Geodia hentscheli* and *G. phlegraei* form dense aggregations. New species and records for the Norwegian- and Greenland Seas are reported.

Larval release and ultrastucture of the egg shell and larva of *Cliona viridis*

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Although several features on the *Cliona viridis* reproduction have been reported in a set of recent publications, whether the larva is a parenquimela or a coeloblastula is still controversial. We collected sponge egg masses attached to seaweeds at the Blanes littoral in May and monitored larval hatching though a stereomicroscope. Larvae released the eggs within two hours after collection. Several stages of the process were fixed and examined through SEM and TEM. The eggshell is made of a fibrillar material clearly different from collagen fibrils. Maternal cells are enrobed within the fibrillar material. A monospecific bacterial strain proliferated on and within the fibrillar, mucopolysaccharide-like, material and appeared to be involved in he eggshell break, which may facilitate larval release. The larva is 80-100µm in diameter, evenly and completely ciliated. It shows all the cell characteristics of a typical parenquimella: it is completely solid, with a higher cell density than that reported for other typical parenquimella. Cilia are 15 µm long and finish in an enlargement. The cells of the pseudostratified external layer are elongated with an apical zone mostly occupied by the basal cilia apparatus and a high amount of small vesicles, a central zone with the nuclei, and a basal long zone full of lipid and yolk inclusions. The inner zone is full of nucleolated, archeocyte-like cells filled with high amounts of reserve (yolk and lipid droplets) materials, which indicates their larval nature. A noticeably higher abundance of yolk compared with that of the already described viviparous parenquimella is to be highlighted. Archeocyte-like cells share the larval inner zone with collencytes and densely packed collagen bundles. Neither bacteria nor zooxanthellae were noticed through TEM within the larva tissue.

Genetic similarity between the Indo-Pacific giant barrel sponge, *Xestospongia testudinaria* (Lamarck, 1815) and the Caribbean giant barrel sponge, *Xestospongia muta* (Schmidt, 1870)

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The Caribbean barrel sponge, *Xestospongia muta*, and the Indo-Pacific barrel sponge, *Xestospongia testudinaria*, are morphologically indistinguishable yet are recognized as two distinct species. While the ecology and population genetics of the Caribbean barrel sponge are the subject of a number of studies, less is known of its Indo-Pacific counterpart. In this study, barrel sponges were collected from northwestern (Pangasinan) and central (Romblon) Philippines and identified on the basis of morphology and sequence of a mitochondrial phylogenetic marker, the I3-M11 partition of the cytochrome oxidase subunit 1 (CO1) gene. Sequences of this marker from fifteen sponges morphologically identified as *X. testudinaria* are found to be at least 99.6% similar to those of *X. muta*. Of the four haplotypes reported for *X. muta*, two are identical to two of the four *X. testudinaria* haplotypes observed so far; in fact, the most common *X. muta* haplotype in the Caribbean is also commonly observed in the *X. testudinaria* specimens. Only the presence of a haplotype not found in *X. muta* but appears common in *X. testudinaria* hints that these two spatially exclusive species are at least different populations. While these observations may not serve as definitive basis for recognizing the conspecificity of the two taxa, they at least support this hypothesis and thus highlight the need to further investigate this taxonomic issue.

Time-dependent spiculogenesis and effect of dissolved silica and iron on spicules production in primmorphs of the demosponge *Petrosia ficiformis* (Poiret, 1789)

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The building of skeletons is considered in Metazoa the most important process for the development of body plans, whose architecture is the main mechanical support. Spiculogenesis is considered the first step in the morphogenesis of a sponge. The formation process of spicules has been clearly detailed in literature from a descriptive point of view, but the rate of spicule production remains an important step for the comprehension of modalities of construction of skeletal elements for a sponge. Production of spicules in primmorphs of the sponge *Petrosia ficiformis* is here investigated after long term cultures and by evaluating the influence of different cultures conditions, enriching sea water medium with silica and iron, alone or combined. According to previous reports, both elements resulted to affect the production of spicules. The primmorphs cultures enrichment with these elements determined a different production of spicular types: silica affects production of fusiform oxeaes, while iron does on the production of a sort of strongyloxeaes, a spicular type absent in *Petrosia*. In all conditions however strongyles are always rare. The addiction of silica generated not only an increase in number of spicules, but also thicker and longer respect to seawater. Results showed no difference in the short time observations between the addiction of silica and both silica and iron, but differences arose on the second and third week of observation. Therefore low concentrations of iron seemed to have a role on spicule formation and silica deposition, while silica gave confirmation of its influence on the rate of spicule production. The variability in types, shape, and size of the spicules produced depending on variations of physical and chemical parameters, such as ions concentrations, confirmed once again important implications for taxonomy.

Host specificity in sponge-inhabiting barnacles (Crustacea, Cirripedia)

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The subfamilies Acastinae and Bryozobiinae and the genus *Membranobalanus* are the three groups of barnacles (Cirripedia: Crustacea: Archaeobalanidae) with species known to be obligate symbionts of Porifera. Few studies have attempted to identify both sponge and barnacle taxa in these relationships. Even less is known regarding the species-level specificity of the relationships. Taxonomic data from identified specimens sampled at the Porifera collection of the Queensland Museum is compiled with sponge identifications made from the sponge-inhabiting barnacles in the Cirripedia collection at the California Academy of Sciences. The combined dataset of barnacle taxa and their sponge hosts is examined for host specificity for barnacle species, genera and subfamily levels. The barnacle species in the subfamily Acastinae live in a broad range of Porifera (at least 81 species of Porifera from several different orders); barnacle species in the genera *Membranobalanus* and *Bryozobia* are obligate symbionts of clionaid and *Clathria* sp. (*Microcionidae*) sponges, respectively. *Bryozobia* was originally believed to be a symbiont of the calcareous bryozoan on which it was discovered, but the present data show it to be an obligate symbiont of *Clathria* sp. growing in or on calcareous substrata. The Acastinae is likely a polyphyletic group, perhaps having evolved symbiosis more than once in different groups of sponges. The bryozobiinae and *Membranobalanus* are more specific in their host requirements. It's possible that these two groups evolved more recently from independent archaeobalanid lineages not directly related to the Acastinae. However, it is interesting to note that although Acastinae are found in Microcionidae, they are not presently known from hosts in Clionaidae. Preliminary phylogenetic analyses of some of the sponge-inhabiting Cirripedia taxa using DNA sequence data confirm the concordance of barnacle and sponge host taxon relationships. Additional phylogenetic work and chemical ecological studies of the Porifera hosts have the potential to tell us more about the evolutionary ecology and history of these relationships. It is well known that barnacle larvae use chemical signals for settlement, therefore it seems likely that sponge chemistry plays an important role in dictating potential host taxa.

A phylogenetic analysis of the genus *Rossella* (Hexactinellida, Rossellidae)

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The taxonomy of *Rossella* is in need of revision. The species content of the genus has changed dramatically over the years, expanded or contracted depending on the criteria used by the taxonomic authority consulted. The genus was regarded as a putative monophyletic unit showing a bipolar distribution. However, recent molecular evidence has suggested that *Rossella* is in fact polyphyletic, and that *Rossella* s.s. is probably restricted to the Southern Oceans, where it shows a circum-antarctic and subantarctic distribution. Herein, we provide a molecular phylogenetic assessment of the genus *Rossella*, based on a fragment of the nuclear ribosomal large subunit (28S) DNA. We aim to further test the monophyly/polyphyly of the genus that was previously reported, as well as the proposed circum-antarctic cohesion of the Antarctic *Rossella* component.

The Sponge Genetree Server v.2: a phylogenetic pipeline for an ever current tree

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Molecular phylogenetic studies of the phylum Porifera have accumulated over the years a considerable number of sequences from different markers, both mitochondrial and nuclear. This massive body of information has been brought together in the Sponge Genetree Server, an initiative to provide an ever up-to-date overview on the phylogenetic signal of sponge sequences. Here we present an extension to the Sponge Genetree Server (www.spongegenetrees.org) that allows its complete automatization. Our phylogenetic analysis pipeline automatically downloads and filters poriferan rDNA sequences from NCBI's GenBank, aligns the sequences to established rDNA structural alignments, and proceeds to infer phylogenetic hypotheses on a routine basis. The ever up-to-date, ever growing phylogenetic tree that is inferred this way serves to summarize the increasing knowledge on sponge molecular systematics and provides insight into the phylogenetic signal of popular markers for the study of Porifera.

A high-throughput, low-cost Porifera DNA barcoding pipeline

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DNA barcoding provides a simple and standard way to screen sponge collections for diverse applications such as taxonomy, systematics, biogeography, ecology, etc. However, the massive number of sponges deposited in museum collections around the world, and the inherent difficulties involved in sponge molecular biology requires establishing laboratory methods suited for the challenge. Rapid, effective, reliable, and relatively inexpensive laboratory protocols have been published elsewhere. The suitability of these protocols remains to be tested for sponge DNA barcoding. Here we present a quantitative assessment of our barcoding pipeline established at the Molecular Geo- & Palaeobiology Lab., LMU, Munich for the Sponge Barcoding Project (www.spongebarcoding.org). We also provide a discussion on some problems and pitfalls found in sponge barcoding techniques and prospective ways to solve them.

Phylogenetic diversity of Antarctic sponges: Insights and prospects

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Sponges are important members of Antarctic benthic marine ecosystems, where large sponges provide refuge to other invertebrates, and increase the structural complexity of the benthos. Several species have been described to date, and sponge species richness in Antarctica matches that of tropical communities. Despite their importance, the phylogenetic relationships of the sponges occurring in Antarctica remain unknown. Here we present the first results on the molecular systematics of Antarctic sponges. Based on these results, we explore the phylogenetic relationships of the Antarctic sponges and provide insights into the phylogenetic diversity of Porifera in this region. Finally, we discuss the implications of the new results for the future development of Antarctic sponge systematics, biogeography and molecular ecology.

Evolution Pattern of Baikal Sponge Fauna since Late Miocene

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Baikalian endemic sponge fauna of Lubomirskiidae family is one of the unusual representatives of the ancient baikalian fauna. In spite of all the research efforts, many questions on the evolution of baikalian sponge are still to be answered. The most ancient spicules of Lubomirskiidae were found by G.G. Martinson in early Tertiary sediments of South Baikal terrigenous deposit in Tunka region (Martinson, 1938, 1948). The age of these sediments is probably Oligocene (36 -24 Ma). The modern molecular investigations estimate relative adolescence of Baikalian endemic Lubomirskiidae family, approximately 1,3–3,8 Ma. (Itskovich et. al., 1999, 2005). Thus, certain contradictory data originating from paleontological and molecular investigations are available. The baikalian sponges fossilized in sediments in the form of isolated skeleton needles, i.e. spicules, that have been well preserved for millions of years. The progress in the modern methods of drilling enables us to obtain baikalian bottom sediment samples aged a few millions years. According to our study (Weinberg, 2001; Weinberg et. al., 2003) of these sediments, almost all recent species of baikalian sponge from Lubomirskiidae family had existed in Late Pliocene, 3.2 Ma ago. However, the sponge fauna in Late Pliocene (3.2 - 2.8 Ma) was by far richer than the recent one. Additional to the spicules of nearly all recent sponges, 24 spicules types of "extinct species", which have no analogues with recent ones, have been found. During the glacial period (2.8 – 2.5 Ma), which was accompanied by changes in tectonic conditions of Lake Baikal, dramatic and irreversible transformations took place in both families (Spongillidae and Lubomirskiidae). The majority of the "extinct species" disappeared. Later, the composition of the sponge fauna became closer to that of the present day, but it never reached the abundance that preceded the climate cooling. To summarize, according to our reconstruction of fossil sponge fauna development, the recent sponge fauna of Lake Baikal can be regarded as the impoverished fauna of the Late Pliocene. The well formed spicules from certain species of the Lubomirskiidae family (*Rezinkovia arbuscula*, *Baikalospongia fungiformis*, *Swartschewskia papyracea*, and *Lubomirskia incrustans*) were also found in Late Miocene sediments (6, 50 – 4, 75 Ma 10 Ma ago) (Weinberg et al., 2004). According to our preliminary investigation of the species composition in the sediment of this period (6, 50 – 4, 75 Ma and 10 Ma), sponge fauna of Late Miocene is poorer compared to the sponge fauna in Late Pliocene.

Porifera of Project MABIREH: new species and records of Acarnidae from King George Island, Antarctica

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In the 2007/2008 IPY and austral summers following thereafter (2008/2009 and 2009/2010), 101 specimens of sponges were collected by dredging and scuba-diving in King George Island by Project MABIREH (Marine Antarctic Biodiversity in Relation to Environmental Heterogeneity at Admiralty Bay, King George Island, and adjacent areas). In the materials collected by Project MABIREH, twelve were sponges belonging to Acarnidae - *Iophon unicorne* Topsent, 1907, *Iophon* sp.nov. 1-2, and *Megaciella annectens*. This material is briefly described here. *Iophon unicorne* (MNRJs 11051,11072, 11102, 11105 and, 11126) was found in Admiralty Bay and Maxwell Bay, collected between depths of 10 and 61 meters. Megaescleres are styles (358.9-533.5 x 3.1- 18.5 µm) and tylote (174.4-307.8 x 3.1-15 µm). As microscleres, the specimens has only palmate anisochelae (12.5-22.5 µm). This species has been reported in the literature with and without bipocilla. These microscleres, when present are 7-13 µm long, and were reported from specimens collected between 80 and 130 m depth. *Iophon* sp. nov. 1 (MNRJs: 11101, 11128 and, 11130) was also found in Admiralty Bay and Maxwell Bay, between 5 and 61 m depth. Spicules are styles (180-461.7 x 1.3-27.5 µm), tylotes (215.5-307.8 x 1.3-10.3 µm), palmate anisochelae (15-22.5 µm) and bipocilla (4.8-12.5 µm). The species approaches more closely *I. terranovae* and *I. timidum*, but differs in many respects related to spicules' categories and their dimensions. *Iophon* sp. nov. 2 (MNRJ 11134) was found only on Maxwell Bay in depth of 10 meters. Its spicules are styles (348.8-461.7 x 8.2-18.5 µm), tylotes (236-307.8 x 5.1-12.3 µm), and bipocilla (15-20 µm). *Iophon* sp. nov. 2 approaches *Iophon flabellodigitatus*, but besides the absence of anisochelae, here reported for the first time in the genus, it can be distinguished by several additional spicular traits. Assignment of this species to *Iophon* was made after a thorough revision of likely Microcionine and Myxilline taxa where it could fit. Notably, Acarnidae, Coelosphaeridae, Hymedesmiidae and Myxillidae. *Phorbas nexus* appears to have quite comparable microscleres, but the remarkable similarity to the spicule complement of *I. flabellodigitatus* led us to consider the present material more likely a new species of *Iophon*. *Megaciella annectens* (MNRJs 11061 and 11059B) has styles (379.6-718.2 x 10.3-35.9 µm), oxeas (300-440 x 10-20 µm), tylotes (200-455 x 4-10 µm), toxas (62.5-700 x 1-6.5 µm), and palmate isochelae (12.5-17.5 µm). The specimens were collected at 20 m depth in Maxwell Bay. The genus is reported for the first time in the Antarctic. The Argentinean holotype was studied with SEM for detailed morphologic comparison with the Antarctic materials. We concluded that both are conspecific, albeit a series of smaller differences. The holotype has true styles instead of subtylostyles, its ectosomal strongyles have much smaller spines, and its toxas approach more a V-shape and their terminations are blunt, nearly club-shaped, as opposed to acerate. This record strengthens the faunistic link between shallow Antarctic sponges and those in the deep continental slope of southeastern South America.

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Evaluation and monitoring of the sponge community health condition in Carrie Bow Reef, Belize

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Sponges and corals represent the sessile benthic organisms of highest biomass and abundance on Caribbean reefs. The Belize reef system is the second largest reef barrier in the world (250 km), extending from southern Mexico to northern Honduras. The main goal of this study was to evaluate the health status of sponge communities on the barrier-reef section near Carrie Bow Cay, Belize. There we distinguished four structural zones: the inner and outer reef, and for each of these, the ridge and slope zones. Observations were made each summer from 2007 to 2009, using two methodologies: permanent quadrats and timed transect swims. Abundance and area coverage was estimated along transect bands and quadrats at each reef zone. The reef bottom is dominated by algae (47.3%), followed by dead coral with algae (19.4%). Live coral represent 6.6% and sponges 1.9%. A total of 55 sponge species were found, not including encrusting cryptic forms; therefore, the count is an underestimate of the real richness of this reef. We found the following relative abundance of sponge species near Carrie Bow. On the inner ridge, *Scopalina ruetzleri* was the dominant species (15.38%), followed by *Niphates digitalis* (6.15%), *Aiolochroia crassa* (6.15%), and *Aplysina fistularis* (6.15%); on the slope of the inner ridge, the dominant species was *A. crassa* (19.85%), followed by *Svenzea zeai* (10.69%) and *Xestospongia muta* (9.16%); on the outer ridge, *N. erecta* (10.95%), *N. digitalis* (10%), and *Aplysina archeri* (7.62%) prevailed. The total prevalence of sponge syndromes/diseases (estimated from approximately 500 specimens) was similar between inner and outer reef and, overall, relatively low, ranging from 0.01% on the inner slope to 0.1% on the outer slope. Species affected were *Callyspongia plicifera*, *N. digitalis*, *Geodia neptuni*, *X. muta*, *Ircinia campana*, *I. felix*, *I. strobilina*, *A. fistularis*, *Aplysina cauliformis*, and *N. erecta*. Of these species, *N. digitalis*, *C. plicifera*, *A. fistularis*, and *I. campana* have not been reported previously with syndromes or diseases. *C. plicifera* and *N. digitalis* showed the most evident and particular pathological symptoms. Both syndromes appear similar by showing a thin red line separating necrotic and healthy tissue of the affected sponge, and by the capability of advancing in multiple directions. Tissue samples of both species are being examined by electron microscopy. Studies under way in Los Roques, Venezuela, have shown that the same species are affected. It is possible that, as with coral diseases, closely related sponge species can be subject to syndromes/diseases in many reefs of Caribbean.

First fossil sponge from Antarctica and its palaeobiogeographical significance

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Hexactinellid sponge *Laocoetis* sp. from the mid-Cretaceous (i.e. Albian – Cenomanian) of James Ross Island is the first record of a fossil sponge from Antarctica. Given the importance of sponges in the modern Antarctic benthic fauna it is somewhat surprising to find that they have such a poor fossil record in Antarctica. Thus the description of fossil specimens from Antarctica, even though they are incomplete and rather poorly preserved, has assumed some importance, and this is particularly so as they can be assigned to a modern genus, that is restricted in its occurrence at the present day to just Madagascar[1]. In this study we described *Laocoetis* sp. and briefly assessed both its palaeoenvironmental and palaeobiogeographical significance. The specimens of genus *Laocoetis* forming the subject of this study were collected from the extensive Lower – Upper Cretaceous sedimentary sequence exposed on the western and north-western margins of James Ross Island, Antarctica. It comprises part of a regressive mega-sequence of volcanic arc-related clastic and volcaniclastic marine rocks. All the studied material comes from a series of localities within the Whisky Bay Formation of James Ross Island. The stratigraphical age-range of *Laocoetis* sp. within this unit cannot at present be resolved to any narrower than Late Albian – Late Cenomanian. Generally, all of the Upper Cretaceous deposits with *Laocoetis* (central and western Europe, Russia, Australia) are interpreted to be littoral to sublittoral settings with no indication of sedimentation above normal wave base. Likewise, there are no obvious records of shallow-water settings in Cenozoic *Laocoetis*-yielding localities (southern Europe, northern Africa). Taking into account the bathymetric distribution of various Upper Cretaceous to Recent *Laocoetis* species and the character of the accompanying fauna of Whisky Bay Formation it is suggested that studied *Laocoetis* sp. inhabited water depths in the range of 100-400 m. It is clear that the sponge genus *Laocoetis* has retracted substantially in its distribution from the Late Cretaceous to the present day. The precise timing of this retraction is still unknown but this would appear to be another example of a formerly widespread Late Mesozoic – Cenozoic marine invertebrate genus that has gradually retracted through the Cenozoic to a very limited modern distribution in low-latitude regions[2]. From a variety of Late Cretaceous mid- to high-latitude occurrences, *Laocoetis* has retracted to just the Madagascar region of the Indian Ocean at the present day. R.V. work was done under the financial support of Ministry of Environment of the Czech Republic, project no. VaV SPII 1a9/23/07. J.A.C. would like to acknowledge financial support from NERC Antarctic Funding Initiative grant GR3/AFI2/38.

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The imperative need to include DNA phylogenies to revise the taxonomy of class Calcarea (Porifera)

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For certain metazoan taxa, establishing a robust phylogenetic classification can be a challenging task. Especially non-bilaterian metazoans such as sponges often lack informative morphological characters due to their simple organization. Calcareous sponges (Phylum Porifera, Class Calcarea) are an extreme example of this challenge. Despite numerous efforts to establish a well-supported phylogenetic system, the currently accepted classification is based on debatable hypotheses regarding the evolution of certain morphological characters. Several of these hypotheses were questioned by results of previous DNA analyses. By extending the currently available data, we now provide new and previously unconsidered hypotheses about the evolution of Calcarea. According to our results, the taxonomy of Calcarea is in desperate need of a thorough revision that cannot be achieved by considering morphology alone. We suggest that DNA data analyses with the largest possible taxon sampling should be included in every taxonomic study of calcareous sponges and in descriptions of new species.

First mitochondrial phylogeography of Calcarea: A case study from *Leucetta chagosensis*

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Many phylogeographic studies prefer mitochondrial (mt) DNA markers to nuclear (nc) markers for several reasons. For example, in most animals mt DNA evolves much faster than nuclear DNA, therefore mt markers provide a better resolution to trace more recent evolutionary events. More importantly, in contrast to nc DNA markers, which at least occur as two allelic copies in an individual, mt DNA is generally only inherited maternally. Therefore, mt haplotypes represent individuals, which simplifies the interpretation of the data, e.g., haplotype networks. So far, however, no mt marker was available for calcareous sponges. Because of this, the phylogeography of *Leucetta chagosensis* (Calcinea, Leucettidae) has been intensively studied by the analyses of nuclear rDNA data (ITS-region and 28S rDNA) and nuclear introns (ATPSb-iII). Here we present the first phylogeographic study of *L. chagosensis* using a mt DNA marker and compare the performance of the new mt DNA marker to the previous nc DNA based studies.

Evolution of non-coding repetitive elements in mt genomes of the genus *Vaceletia*

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Most animal mitochondrial (mt) genomes are densely organized and contain only small regions of non-coding DNA. In contrast to this typical situation, several sponge mt genomes –like the ones from genus *Vaceletia*- posses larger non-coding regions, while other, closely related sponges lack non-coding regions. In *Vaceletia*, the non-coding regions contain repetitive elements. Although *Vaceletia crypta* (Vacelet, 1977) is the only recognized species of its genus, DNA analyses and different growth forms suggest that *Vaceletia* consists of several species. Therefore, the genus is an excellent model system to study the origin and evolution of non-coding repetitive motifs in mitochondrial genomes.

Growth and cadmium stress in a cultivable species of *Haliclona*

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The identification of sponge species suitable for *ex situ* cultivation is essential for basic researches on biomass production and biomonitoring. The first goal of this work was to identify an *ex situ* cultivable sponge. The sponge species tested were: *Desmapsamma anchorata*, *Amphimedon viridis*, *Aplysina fulva*, *Hymeniacidon heliophila*, and *Haliclona* sp. Among these species, only *Haliclona* sp. showed an expressive long term growth (about 17 months). The taxonomy of *Haliclona* sp. was achieved by morphological and molecular (cytochrome C) methods. Biotic and abiotic parameters of the growing tank were measured in order to characterize the aquarium environment. Water temperature, salinity, pH, chlorophyll, Pheophytin, MOD, N, P, Fe, and Si were measured weekly during the last 3 months. Macroalgaes and macroscopic invertebrates were identified and quantified. These results indicated an eutrophic environment with no dangerous accumulation of toxic components. Since *Haliclona* sp. has proved to be *ex situ* cultivable, the second goal was to use it in a cadmium *in vitro* exposure experiment to assess the metal stress under histological and proteomic aspects. The cadmium exposure was made incubating healed fragments of *Haliclona* sp. for different periods (0, 17, 41, 137 and 326 hours) in a control tank and in a tank supplemented with 0.5 mg/L of Cd+2. The cadmium accumulation, detected by ICP-OES, was 0,007 ppb (17hs), 0,020 ppb (41 hs), 0,016 ppb (137 hs), and 0,018 ppb (326 hs). Structures at histological level were compared in order to assess the morphological effect of cadmium stress. The proteins from each fragment were extracted for proteomic characterization. Metallothionein like-protein (25 kDa) was detected by SDS-PAGE using a thiol specific fluorescent reagent, after 326 hs incubation. The 2D-PAGE was used to identify and assess changes in expressed cadmium stress proteins. A down expression was observed in three proteins ranging between 14.4 to 25 kDa. A set of proteins ranging between 14.4 to 45 kDa were over-expressed, however, two 25 kDa proteins showed a higher induction. These results showed that *Haliclona* sp. is a suitable species for *ex situ* cultivation and can be used to address Cd stress in *in vitro* biomonitoring experiments. Further investigations evaluating other stress conditions must be performed to confirm if these Cd induced proteins are general stress indicators for sponges.

Rising sea temperatures: implications for the sponge holobiont

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This research investigated the thermal tolerance of marine sponges by assessing the impact of elevated seawater temperature on both the sponge and its symbiotic microbial community (holobiont) for both larval and adult stages. Adult *Rhopaloeides odorabile* sponges were exposed to temperatures ranging between 27-33°C and larval *R. odorabile* to temperatures between 27-36°C. No effect of temperature was observed in adult sponges between 27-31°C. However, sponges exposed to 33°C exhibited minor surface necrosis after 24 h and major tissue necrosis (50 – 70%) after 3 d. In contrast, larvae showed remarkable thermal tolerances, surviving and metamorphosing in temperatures up to 38 °C, but were not functional at 40 °C. While larvae can metamorphose in as little as 6 hours at ambient temperature (27°C) this reduced to 2 hours at elevated temperatures (32-36 °C). No differences in bacterial community composition were detected in adult sponges between 27-31°C. However, adults exposed to 33°C exhibited a complete loss of the primary cultivated symbiont within 24 h. Furthermore, DGGE and 16S rRNA gene sequence analysis detected a dramatic shift in the bacterial population in adults between 31 and 33°C. Sponges exposed to 27-31°C hosted microbes with highest homology to known sponge symbionts whereas sponges exposed to 33°C were dominated by microbes having sequence similarity to coral pathogens. The microbial community in larval *R. odorabile* are less affected by elevated temperature, with DGGE showing completely conserved symbiotic populations in larvae at all temperatures up to 34°C. After 24 h at 36°C there was a loss of some symbionts and the arrival of alien microbial species. The breakdown of symbioses and stress in the adult sponge occurred at temperatures identical to those reported for coral bleaching, indicating that adult sponges may be similarly threatened by climate change. In contrast, larvae were able to survive and metamorphose in temperatures up to 38°C (albeit at lower levels than at ambient temperatures) and had a highly conserved symbiont population at temperatures up to 34°C. Larvae are positively phototactic, their upward migration at release placing them in the upper water column before their return to the benthos to settle. This behaviour places them in the warmest part of the water column where they presumably require thermal protection. Overall, this indicates that the larval life history stage may be less sensitive to the elevations in seawater temperature that are predicted with changing climatic conditions.

Great Barrier Reef sponges: symbiont variability between and within different host species

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Marine sponges are recognised for their often diverse and abundant communities of associated microorganisms. These microbes can comprise up to 40% of sponge volume and often serve important functions for the host including ammonia removal, a supply of fixed carbon and the production of secondary metabolites for use as chemical defences. Twenty bacterial phyla have now been documented from sponges and many of the associated microbes appear to be truly sponge-specific. Crucial to improving our understanding of sponge-microbe symbiosis is an accurate picture of microbial community composition and the variability that occurs within and between sponge species. We surveyed three replicate samples from 14 different Great Barrier Reef sponge species to assess the microbial community composition and determine the stability of the microbial associations. All sponges contained a conserved microbial population, with *Cinachyra* sp., *Cymbastella* sp., and *Phyllospongia* sp. having identical microbial profiles in all replicate specimens. There was some overlap of the microbial communities within *Luffariella variabilis*, *Haliclona* sp., *Paramyxilla* sp., *Ircinia* sp., *Coelocarteria singaporense*, *Hamigera* sp. and *Xestospongia testudinaria*. *Phakelia flabellata* and *Carteriospongia foliascens* contained a slightly different microbial community from these other species as did *Xestospongia exigua* and *Cinachyra* sp. which showed a high degree of similarity to each other. The sponges *Coscinoderma matthewsi*, *Phyllospongia* sp., and *Cymbastella* sp. hosted microbial communities that were clearly distinct from all other sponge species. These results highlight the strong influence of the host sponge on the composition of the microbial community and confirm the stability of these microbial associations in different individuals of the same sponge species. All sponges maintained populations of heterotrophic bacteria with sequence similarity to previously described sponge symbionts. In addition, *Xestospongia exigua*, *Cinachyra* sp., *Luffariella variabilis*, and *Hamigera* sp. all maintained a stable association with various archaeal species and *Xestospongia testudinaria*, *Carteriospongia foliascens*, and *Coelocarteria singaporense* hosted phototrophic microbial populations.

Taxonomy and biogeography of the shallow marine sponge fauna of Peru: The ESPER Project

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Porifera are well known for the effectiveness of their specialized diet on bacteria and similarly sized organic particles and have thus been connected to ecosystem health at several occasions. Sponges comprise today the richest natural source of new chemical structures, most of which present varied biological activities, and are currently under pharmaceutical screening for new drug leads against several human diseases. Until 2003, the south-east Pacific had one of the least studied sponge faunas in the world, and the little that was known about the species composition and geographic distribution of Peruvian marine sponges markedly contrasts to their recognized importance, both as keystone coenoses at several marine communities at nearly every latitude, as well as potential sources of economical revenue. In 2007, in the frame of the Global Taxonomy Initiative, the RBINSc has launched a cooperation project with the UPCH to inventory the diversity of sponges along the entire coast of Peru. Our scientific partners are the MNRJ and MHNG. Before our investigation, only 26 species were known, a large majority of which were deep sea Hexactinellids collected at the end of the 19th Century. Our three expeditions along the Peruvian coast, in 2007, 2008 and 2009, revealed a great variability of the sponge fauna from Tacna to Tumbes, with a larger diversity and abundance in the most northern zone (sub tropical). Our field work in 97 localities leads to the first reference sponge collection of the Peruvian coast which contains at present nearly 850 samples, all shared, deposited and curated in 4 different institutions (Peru, Belgium, Switzerland, and Brazil). Although identifications and descriptions are far from being completed, we estimate to have collected around 100 different species, among which several are new to science. Since the ESPER Project was initiated in 2007, 13 Peruvian students in Biology were trained in field work to collect sponges, 4 of them working under the supervision of the local partner of the project are presently developing their master theses (2 in taxonomy, 1 in genetics, and 1 in biochemistry). During this project, the Peruvian partner has had the opportunity to be trained on sponge taxonomy, collection management, including long term preservation methods and database management at the Royal Belgian Institute of Natural Sciences. A database including access to underwater pictures, maps, and micrographs has been developed to handle the sponge reference collection and a field guide of the commonest Peruvian sponges will also be an outcome of this project, filling a gap in the knowledge of the biodiversity of the South American marine fauna.

In situ hybridization: a proposed method to ensure accurate gene expression

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Evo-Devo studies in sponges have become more common in the last decade, but gene isolation and localization in sponge tissues remains difficult. This is particularly the case in species where cDNA libraries and genome projects are still not available. Here, we propose an improved methodological approach to isolate genes and determine their localization in sponges. To find genes involved in development we have taken a degenerate PCR approach using nested degenerate primers on cDNA from a mixture of sponge stages. Following cloning and sequencing, 5' and 3' ends of isolated fragments are found using RACE PCR techniques. Knowledge of the full mRNA sequence allows trustworthy phylogenetic analyses and longer templates for *in situ* hybridization (ISH) probe synthesis. Expression of genes in sponges is the ultimate goal of sponge evo-devo research, and yet difficulty remains in both generating believable expression patterns and interpreting them. We suggest a quick and straightforward PCR-based system for making ISH probes whereby the template is acquired with specific primers, and a T7-promoter site is added to the 5'-end of the reverse primer. Probes of 800 – 1000 bp are preferable; longer probes are more specific and give stronger signal even after high stringency washes. Simple synthesis of a riboprobe is accomplished using a commercially available RNA DIG Labeling kit. Probe quantity and quality is verified by absorbance at 260/280, and dot blots using known standards. Given the uniformity of tissues in sponges, use of an appropriate control is also essential. For demosponges, silicatein homologs are an excellent control since they are only expressed in sclerocytes and their precursor cells. Other commonly used controls (e.g., actin and beta-tubulin) should be expressed in the whole tissue and match what one might expect of pure background staining. We found that the use of 1% triethanolamine sufficiently reduces background, although it may affect tissue structure. This approach has given consistent results in our lab and can be applied for different systems in the future.

Global change and bioerosion – preliminary results

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In the recent discussion how biotic systems may react to raised CO2 and temperatures in the marine realm, we presently study the role of the most effective endolithic bioeroders – the boring sponges. Several hypotheses predict an increase of sponge-driven bioerosion along with weakening of the calcification potential of their hosts. Chemical etching of sponges is thought to be enhanced by ocean acidification. Using an integrated approach we aim to explain interaction effects of global ocean acidification and climate change with the main focus on how altered CO2 concentrations and raised temperatures may influence bioerosion rates. Experiments are conducted on bioeroding sponges as model organisms, because they have unique properties allowing simultaneous evaluation of different parameters: Symbiotic bioeroding sponges such as *Cliona orientalis* Thiele, 1900 are likely to be affected by climate change and can be compared to both symbiotic reef builders and asymbiotic bioeroding sponges such as *Cliona celata* Grant, 1826. By using field sites in two marine environments in different climates (Helgoland Island in the cold-temperate North Sea and Great Barrier Reef in the tropical Pacific), results will provide a large-scale impression of spatial variation. At the time of the conference, preliminary data will be available from Helgoland, and the first half of the field studies on the Great Barrier Reef will be completed. Ultimately, data will convey critical insights into global trends of biologically caused decalcification.

Molecular systematics and biogeography and biodiscovery of *Dysidea fragilis*

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Keratose marine sponges are a group notoriously difficult to classify. *Dysidea fragilis* has been recorded as a relative common and cosmopolitan species within this group. Due to the existing ambiguities in morphology-based classical systematics and ongoing difficulties in the more recent field of chemotaxonomy, the focus of this study is on applying molecular methods to investigate phylogenetic relationships of this group of sponges alongside investigations of their morphology and chemistry. Initial phylogenetic analyses of 28S rRNA gene sequences have indicated the possible presence of cryptic species and further work on additional gene loci is being undertaken to confirm this observation. Few studies have been carried out at the population level in sponges thus investigating micro-evolutionary processes in this group will contribute greatly to our growing understanding of sponge evolution. A number of *Dysidea fragilis* biopsies from locations along Ireland's coast and abroad, along with a smaller number of other keratose species have been collected for population genetic work. The ribosomal intergenic regions (ITS1 and ITS2), commonly used in population genetics appear to be problematic for use with *Dysidea fragilis*. The high levels of intragenomic polymorphisms in sequenced clones hinder distinction between individuals, directing the focus to the establishment of microsatellite analysis for this species. The population structure of this species along the Irish west coast will be presented.

A new bioeroding sponge associated to Northeast Atlantic cold-water corals: *Alectona tendali* sp. nov. (Porifera, Alectonidae)

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Sponges are an important component of the coldwater coral associated fauna and exhibit a remarkable diversity in these ecosystems. The genus *Alectona* constitutes a particularly important group of demosponges with a strong affinity for biogenic substrata. They bore into calcareous substrate and are commonly found associated to both shallow and deep-sea corals. *Alectona millari* Carter, 1879 is one of the most common excavating sponges associated with the main deep-water reef forming species - *Lophelia pertusa* and *Madrepora oculata* – on which it creates large endolithic chambers. Originally described from an area between the North of Scotland and the Faeroes, this species has been extensively reported throughout the NE Atlantic and Mediterranean. However, morphological and molecular examination of specimens collected in several reefs of the European margin (Rockall Bank, Mingulay, Sula reef and Vaagegrund) has revealed the existence of two *Alectona* species. Differences regarding shape and size of the spicules that constitute the species' skeleton, as well as a clear genetic gap between them has lead to the description of *Alectona tendali* sp. nov. In this study both species are described and illustrated.

On a Hexactinellid sponge ground at the Great Meteor seamount (Northeast Atlantic)

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Hexactinellids or glass sponges constitute a predominantly deep-sea sponge group typically occurring at bathyal and abyssal depths. Some species seem to form dense populations along the European continental shelf but the distribution and extent of these populations remains ill-known and the driving factors behind their occurrence poorly understood. Here we report an aggregation of the Hexactinellid sponge *Poliopogon amadou* Thomson, 1878 at approximately 2700 m depth on the Great Meteor Seamount, found in the course of a survey with the ROV Luso. This is to our knowledge the southernmost record of a hexactinellid ground in the Northeast Atlantic and the first to be associated to a seamount ecosystem. A description of the species, accompanied by scanning electron microscopy of its spicules, is provided.

Change in population densities of the marine sponge, *Oceanapia sagittaria* and its implications on impact of coastal development

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The marine sponge *Oceanapia sagittaria* is a common macrobenthic invertebrate in coral communities in the Gulf of Thailand. This sponge has been found living within the sediment and showing a special adaptation to life embedded in sediment. Population densities of *O. sagittaria* in coral communities of Laem Set, Samui Island (a popular tourist destination) and Taen Island, a small island south of Samui Island, in the Gulf of Thailand, were studied by using a quadrat method in 1998 and 2010. The sponge capitum was found sitting on top of a long fistule that was buried in sandy areas and coral rubble at depths 1 – 4 m of the study sites. The mean population density at Laem Set in 2010 (2.7 colonies/m^2) was much higher than that in 1998 (0.7 colonies/m^2). However the mean population density at Taen Island in 2010 (1.3 colonies/m^2) was slightly higher than that in 1998 (1.1 colonies/m^2). Intensive coastal development in Samui Island during the last two decades has resulted in high sedimentation in near-shore coral reefs. High population density of *O. sagittaria* in Samui Island may imply high sedimentation rates in coral communities of Samui Island.

Sexual reproduction in *Hippospongia communis*

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Hippospongia communis reproductive cycle has been studied over 2 years from October 2005 till November 2007. In total 146 specimens of different sizes (volume from 356 cm^3 to 16640 cm^3) were collected from Kerkennah Islands of Tunisia. The sexual reproduction was studied using histological sections from the samples of the bath sponges. The sex ratio, the reproductive effort and the reproductive cycle are presented in this work.

THE SPONGE GUIDE: A web-based interactive photographic guide to identify Caribbean sponges

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Taxonomical identification of sponges is usually difficult and restricted to experts, hampering their broader inclusion in ecological and other studies. Illustrated catalogs are important tools for scientists as well as for interested laymen to become familiar with the most common and important groups of marine organisms; but they are costly, which can limit both the scope and access to the information. With the coming of digital tools, catalogs may now have great size, can be updated periodically, and can be made available to all users through the Internet. Taking advantage of the experience gained in taxonomy of Caribbean sponges, we designed and built a relational database to catalogue and display the natural variability of Caribbean reef and lagoon sponge species through underwater photographs. For all species, one or more tissue fragments were collected for taxonomical identification in the laboratory from spicules, skeleton and specialized literature. The working database includes a data entry module for each photo with information on sample number, locality, habitat, color, consistency, and shape. The on-line system (www.spongeguide.org) displays for each species the available photos, a brief description, and taxonomical comments. Species can be accessed directly from a full taxonomic list (scientific or author names), or indirectly from a query window in which the form, color or consistency are selected, accessing all photographs with the combination requested. From the photographs that best matches the user query, the species can be accessed to consult the details. A full taxonomic reference list is also given to allow final identification after consulting the more specialized literature. Crosslink with the World Porifera Database (www.marinespecies.org/porifera/) are also given for each species. The database currently includes 1164 photos of 182 species-morphs, and will continue to be updated periodically. The working database has a module for inserting microscopic images of the skeleton, which is expected to be developed in the future. We are currently working in an off-line downloadable version to be brought to the field where there is no internet access. During the first year, the Sponge Guide has been accessed over 10,000 times, from 121 countries, with visitors spending an average of 5.5 minutes on the site during a visit.

The utility of proteins related to the immune system as pollution biomarkers using the marine sponge *Hymeniacidon heliophila* Parker, 1910 as bioindicator species

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Now-a-days most marine environments have suffered some form of anthropogenic impact. Among the most important ones are the discharge of industrial and domestic waste that increase eutrophization and hydrocarbons exposure, sometimes leading to the complete modification of a marine habitat. Therefore, the monitoring of marine ecosystems, through the use of sentinel organisms, has been essential to assess the environmental impacts caused by human activities. Currently, the use of biomarkers to predict and, in many cases, to limit the effects of these impacts has been greatly increasing. Proteins related to the immune system can be efficient biomarkers of stress due to pollution, since stress reduces the efficiency of the organism's immune system, leading many times to their death. The objective of this study was to test in marine sponges the efficiency of proteins related to the immune system as possible biomarkers of stress associated to the arrival of pollutants. The activation of the proteins p38/MAPK and the Allograph Inflammatory Factor (AIF-1) was assessed after the exposure of the marine sponge *Hymeniacidon heliophila* to pollutants, such as high concentrations of *Escherichia coli* and petroleum derivatives. The exposure to the lipopolysaccharide (LPS) of *E. coli* for 24 h and 48 h showed different results for p38/MAPK and AIF-1. For AIF-1, the activation was significantly higher than controls only 4 h after exposure, with highest values obtained in 24 h. Conversely, the highest activation of p38/MAPK was reached only 48 h after the exposure. The activation of both proteins as a response to the exposure to motor oil for 48 h was not significantly different from controls. To conclude, proteins related to the immune system in marine sponges are efficient biomarkers of stress caused by the presence of pollutants. However, *H. heliophila* may not be a good bioindicator species, since it might be resistant to the presence of petroleum derivatives. Additionally, each biomarker has different activation times and, thus, it is important to always test their response at various time intervals after the exposure to pollutants to determine the response profile of each biomarker.

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